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Abundance of Plankton Taxa and Their Effects on the Planktonic Community in Lake Auburn.

Dylan Ernest Larose
Bates College, dlarose@bates.edu

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Abundance of Plankton Taxa and Their Effects on the Planktonic Community in Lake Auburn.

By
Dylan Larose

Lewiston, Maine
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Abstract

Lake Auburn, the source of Lewiston and Auburn drinking water, is a dimictic mesotrophic lake located in Auburn Maine. The lake's pristine nature allows for water, under strict guidelines, to be processed for drinking without the need or requirement for filtration. The lake must maintain relatively clear water in order to continue to be a filterless drinking water supply. Past water treatments have been undertaken to maintain clear water. A closer look was taken at the community dynamics of Lake Auburn's plankton in an effort to better predict when costly and disturbing water treatments should be administered. Over fifty taxa were identified, and a total of 34 taxa were consistently counted and recorded. Of particular interest were also the reproductive habits of the cyanobacteria *Gloeotrichia.* It was found that the *Gloeotrichia population* as a whole divided about one week prior to reaching its maximum abundance late summer. Only a weak negative correlation was found between Secchi depths and *Gloeotrichia* abundance. Water clarity did not improve later in the season when Gloeotrichia abundance diminished suggesting that other plankton or abiotic factors are responsible for low visibility in the fall. *Dolichospermum* was found to have consistently high to medium abundance as was the prior findings of Ana Urbina in 2016. *Bosmina* cladocerans, nauplii copepods, and *vorticella* ciliates, all reached strong apexes during the low visibility period in the fall and could be a potential indicator of failing water conditions. Future studies in Lake Auburn's planktonic assemblage should focus on investigating these taxa and their relationships inside the plankton community.

2. Background

2.1 General Lake Ecology

No two lakes are the same. Their volume, latitude, altitude, morphology, tributaries, surrounding bedrock and soil types, age of succession, local biome, and proximity to urban development all play a role in the specific abiotic characteristics of that body of water. These abiotic factors in turn inform the biotic world. The biotic community of a lake is ever changing both across years, and seasonally. Tracking long-term trends in a lake's biota is useful to predict where the trophic status of that water may be heading, and can help inform local policy and planning. In particular, the assemblage of the planktonic community, hereafter referred to simply as community, is indicative of the trophic status of a lake, and careful monitoring of ratios of certain taxa have been well documented in helping determine the trophic status (Kalff, 2002). Primary producers respond directly to nutrient availability, and their composition can have a strong influence on water quality (Blomqvist, 1996).

Lake communities are the product of many abiotic factors including time in the form of succession and equilibration to perturbations in the local environment. Lake communities have been shown to change their composition over the span of years. For example, as a lake warms with climate change and accumulation of nutrients from shoreline development, that lake is expected to undergo eutrophication (Chorus et al., 2021), (Filiz et al., 2020). A lake characterized by cold water and nitrogen limitations might shift from diatom and copepod dominated blooms to cyanobacteria and cladocerans. Similar to the change that is tracked year over year, there is also a regular cycle in the community that happens annually that will be discussed in more detail later. Dimictic lakes are particularly interesting because their twice annual turnovers create contrasting environments that dramatically change water parameters such as solar radiance strength and duration, currents, nutrient availability, temperature, and dissolved oxygen content. Furthermore the strength, and location of the thermocline affect the communal composition along with turbulence in the water column (Arnott et al., 2021),(Sanford, 1997). Therefore, annual variations in the timing of ice-out and ice-in, major storms, and annual temperature maximums all change the way the community responds and alters their composition from year to year. It is for this reason that no two years have the exact same communal snapshot, and predicting the trajectory of the community is as challenging as predicting the weather. Nevertheless, there are basic trends that can be expected such as a diatom bloom shortly after ice-out, followed by a green algal bloom, and then, if conditions allow, a summer cyanobloom. The successions of primary producers are generally predictable in their order but timing is of a great challenge. Zooplankton, while also having a generally predictable order, do not have as predictable relation to the phytoplankton succession due to their higher trophic order and dependance on the producers. After considering intra-trophic forcings, the predictability of communal succession continues to break down as the season progresses and the richness of the community increases (Cirri and Pohnert, 2019).

This paper will not be focused on the changes in trophic status from year to year, but instead focus on the changes inside of one season, to determine if Lake Auburn's community changes in predictable ways. It is also important to be able to track a lake's community intra-year so that planning can be carried out in managing a lake for blooms.

2.2 Background

Lake Auburn, formerly known as Wilson Pond, bound entirely in the municipality of Auburn, Maine, was carved out by glaciers during the last glacial retreat at the end of the Pleistocene epoch (DACF, 2022). Its 48.2 km^2 watershed encompasses the towns of Auburn, Minot, Buckfield, Turner, and Hebron; with an average depth of 12.2 m, a maximum depth of 36 m, surface area of 9.1 km^2 and an approximate volume of 1.1x10^{11} liters **(**Dudley, 2004). The morphology and latitude of this lake had helped define it as an oligotrophic lake, however climate change and anthropogenic sources have turned this water into a mesotrophic status in recent years (Kalff,2002)(Reckhow,1973). The lake is dimictic with ice out dates ranging from a record-breaking March 18th since records began in the year 1836 to April 25th 2003, being the latest in the past 20 years, with the 20 year average ice out dates three weeks earlier than historical values (AWD/LWD, 2022). The lake has a watershed basin of 3832 ha, seventy-eight percent of which is forested (NLCD, 2016). The lake has two principal inflows; Basin Brook and Townsend Brook, and has a sole outlet that drains into Bobbin Mill Brook and further into the Androscoggin River. Lake Auburn water inflows from surface water and rain range around 825 ± 170 million cubic feet per year (Dudley, 2004) Lake Auburn serves as a source of recreational fishing, a scenic pull off, and the source of Lewiston and Auburn municipal drinking water. The volume of Lake Auburn is 3,920 mcf (Dudley, 2004). Residency time for water is 4.75 years.

2.3 Filtration Waiver

Due to Lake Auburn's exceptionally clean water quality at the time of this writing, the water district continues to hold a filtration waiver granted by the State of Maine and U.S. EPA. As long as the turbidity readings does not go above 5 Nephelometric Turbidity Units (NTUs, a basic measure of turbidity), in two events during a year, or five times in a decade amongst other non water clarity requirements in accordance with 40 CFR Section 141.71, Criteria for Avoiding Filtration under the National Primary Drinking Water Regulations (US EPA 1991).

Low turbidity is crucial to the understanding of how UV light can penetrate the water to sterilize coliforms such as *E. coli*. If the water is too turbid then UV light can not effectively reach all particles in a column and will serialize an inadequate fraction of coliforms present in the sample. Many different sources of particles contribute to turbidity such as suspended silt and clay, organic matter, and plankton (Cabaj, 1996). Planktonic blooms are of interest because they can be the most variable turbidity parameter, and can make up the largest fraction of the light absorbing particles in lakes, as in the case of Lake Auburn (FB Environmental, 2021). Therefore, it behooves policy makers to know as much about planktonic communities as possible so that they are better able to predict blooms giving them more warning time to plan a response.

2.4 Auburn's Treatments

Lake Auburn was historically an oligotrophic lake, but has turned mesotrophic in response to a changing climate and increased development inside of its watershed (Dudley, 2004). As a result, turbidity in the lake is on a rise, with high seasonal variability. Turbidity rates above 1 NTU were uncommon, and annual phosphate concentrations above 11 ppb were unheard of (CEI, 2010) until the bloom of 2011, and then in 2012 there was an even larger bloom. The resulting aerobic decay of this algae at the bottom of the lake resulted in the depletion of dissolved oxygen in much of the hypolimnion. The hypoxia killed a number of lake trout that could be observed washed up on the shores. Diagnostic studies followed in the succeeding two years to ensure that these conditions did not persist or recur. A plan was put in place to address future lake blooms following the 2012 event. In the late summer to early fall of 2018, a large algae bloom emerged and was treated that season with a copper sulfate (CuSO4) algaecide treatment in response to the ongoing bloom as a stop-gap measure. The following year in 2019 AWD and LWD partnered for an alum treatment mixture of aluminum sulfate $Al_2(SO_4)_3$ and sodium aluminate $NaAlO_2$ in a12,000 acres with a 2 mg/l dose that was administered in two phases to floc out excessive phosphate. Alum is costlier but a longer term treatment to address declining water quality with periodic treatments using algaecides. FB Environmental Associates expects that Alum treatments will be recurring to maintain desired water quality (AWD memorandum Dec 2022). At the time of this writing no other treatments have been administered to Lake Auburn despite recent increases in chlorophyll levels and a record surface temperature reading in 2022. Applications of algaecides and flocculants can be considered major disturbances when in the context of a limnic ecosystem. It is well noted that in the wake of a disturbance such as a pulse in nutrients, or the addition of a toxin the assemblage of a community changes (Cottingham et al., 2004), (Graham and Duda, 2011), (Klug, and Cottingham, 2001).

2.5 Prior Work

Communities are a complex web of interactions, often depending on keystone species for regulation but also rely on a myriad of other species in a chaotic array of interactions that can not be modeled properly unless all major species present are considered (Anderson, 2005). An extensive look into Lake Auburn's plankton community has not occurred since 2016, before the algicide and flocculant treatment. In 2016, Ana Urbina in association with Bates College studied the correlation of *Gloeotrichia* in relation to zooplankton with group data collected and counted from the spring of 2015 to the fall of 2016. No correlation could be found between *Gloeotrichia* and the copepods, cladocerans, and rotifers that they recorded. This study focused on potential grazers and ignored all other taxa including competitive species such as other cyanos (*Dolichospermum, Coelaspherium, and Worochinia*), desmids, golden algae, and diatoms.Continuous seasonal surface plankton sampling has been ongoing since the time of Urbina’s publishing, with the incorporation of a wider breadth of taxa and the addition of site #5, producing more data to be considered.

2.6 Phytoplankton

Lakes are an assemblage of life. At the base of every lake ecosystem lies its plankton which can be broken up into two groups: the photosynthetic phytoplankton which are the primary producers, and their predators, the zooplankton. Limnetic phytoplankton are composed principally of cyanobacteria and eukaryotic algae. These plankton are important to study because they fuel the rest of the food web. Each herbivore and their predators are specialized to some degree in feeding on a specific range of plankton; some feed on a single species some on a whole host of plankton (Bleiwas & Stokes, 1985). The initial assemblage of phytoplankton will invariably dictate the constraints on the zooplankton community composition and its stability or sensitivity to perturbations (Carey et al., 2017). The opposite can also be said in that phytoplankton are less sensitive to perturbations if there is a larger biomass of zooplankton or if the zooplankton are large (Cottingham et al., 2004), (Cottingham and Schindler, 2000). Furthermore, each phytoplankton has its own requirements in light, nutrients, temperature, and water chemistry. To provide an example: Cyanobacteria tend to dominate later in the summer when nitrogen concentrations have become depleted and temperatures are on the rise. Much can be learned about water parameters by looking at what is growing in the water, such as; changes in seasonal temperature, nutrient loading, pH levels or even dissolved organic carbon (Hehmann et al., 2001). The addition of nutrients has been shown to increase biomass overall, but can decrease the abundance of dinoflagellates and golden algae, as well as lowering the biodiversity of the community (Cottinghm et al., 1998). Plankton may be short lived, but they are not as ephemeral as some water quality parameters and can help smooth out day to day variations. Looking at planktonic assemblages may be more qualitative than sensor data, but they can give a more holistic and smoothed out picture of the environment opposed to noisy instantaneous probe readings. Methods such as the Planktonic Trophic Index or the Trophic Diatom Index can measure the health of a system (Fu et al., 2021).

2.7 Cyanobacteria

Oxygenic photosynthesis evolved only once (Fischer et al., 2016). Cyanobacteria are the original oxygenic photosynthesizers, making them the simplest and most primitive plankton, all life that utilizes this pathway evolved from cyanobacteria (Schopf 2011). Cyanobacteria also differ from other algae in that many are diazotrophs (nitrogen fixers) and are able to sequester diatomic nitrogen from air or dissolved in water and convert it into useful bioactive ammonia/ammonium. Plants can not fix nitrogen because nitrogenase, the enzyme required for this process, having been evolved before the presence of atmospheric oxygen is permanently denatured by free oxygen, something that plants produce as a waste product of photosynthesis (Buick 2008); (Gallon, 2006); (Luo et al., 2016). Therefore, this chemical process occurs inside of specialized anaerobic cells with thickened walls called heterocysts; any cyanobacterial colony containing heterocysts is diazotrophic (Fey et al., 1968). Nitrogen is often quoted as the limiting nutrient for terrestrial plants. This is as true in water as it is on land, however with the presence of nitrogen-fixing cyanobacteria in water and the ease of gas diffusion within surface waters, phosphate is also a co-limiting nutrient in epilimnetic lacustral zones and is often the primary limiting nutrient due to the prevalence of nitrogen fixation (Kalff, 2002). This sets the stage in dimictic lakes where algae is the first bloomer after ice out and consumes significant amounts of bioactive nitrogen in relation to phosphates. At this point the rarer nutrient, phosphate, becomes the limiting nutrient and the more energy intensive nitrogen fixing cyanobacteria gains a competitive advantage. It is at this point where cyanobacterial blooms become prevalent in lake systems that have high free phosphate (Kalff, 2022).

Cyanobacteria are small single-celled organisms. For this reason light microscopy is limited to only identifying taxa that form colonies (Pound and Wilhelm, 2021). Visually identifiable colonies common to New England include *Dolichospermum* (formerly known as "*Anabaena*"), *Aphanizomenon, Gloeotrichia, Microcystis, Woronchinia, Coelosphaerium, Dolichospermum* and *Nostoc* both form long trichome filaments, *Nostoc* associates with bottom surfaces and so can be assumed to not be part of a surface plankton sample. *Gloeotrichia* colonies are unique in that they form filaments radially with each filament beginning with a nuclear heterocyst in the center of the colony, followed by an akinete, and finishing distally with several increasingly smaller vegetative cells (Schopf, 2000). *Microcystis, Woronchina*, and *Coelosphaerium* can be difficult to distinguish from each other because they all form roughly spherical masses of cells. *Coelosphaerium* can be distinguished by its denser mass of cells around the periphery but an almost hollow appearance as its name implies. *Woronchina's* perfect sphere is usually broken forming a shape more like a kidney bean or grain of pine pollen, often these colonies have a halo-like extracellular matrix with scores or satellite cells studded into the matrix. *Microcystis, the* most cosmopolitan of the three, tends to be less perfect sphere and more of an amorphous blob of homogeneous cells (Xie et al., 2003). *Aphanizomenon* and *Oscillatoria* also form linear trichomes but are easily identified by *Aphanizomenon's* nature to consolidate like a mat of lawn clippings, and *Oscillatoria's* well defined sheath over the length of the trichome.

2.8 Nutrients

Nitrogen and phosphorus are often the limiting nutrients in limnic phytoplankton blooms, diatoms and other algae depending on higher ratios of nitrogen to phosphorus than cyanobacteria. This is due to most colonial cyanobacteria's ability to sequester useful nitrogen from atmospheric diatomic nitrogen inside their heterocysts (Gallon, 1992). When phosphorus levels get particularly low, cyanobacteria, like *Gloeotrichia*, can go into a vegetative state, and rest on the bottom until more favorable conditions come along. Sometimes these conditions manifest themselves as anoxic benthic events which release phosphate from transition metals like iron when it is reduced from ferric to ferrous iron (Whitton, 2012). These colonies can then incorporate gas into their akinetes making the colony buoyant and allowing it to migrate to the photic zone in a lake, bringing with them an upwelling of nutrients in a positive feedback loop (Paerl 1988). *Gloeotrichia* recruitment in mesotrophic lakes has been shown to increase biomass of other phytoplankton, likely from the upwelling of P that they bring with them (Carey et al., 2014). This motility further complicates the understanding of the plankton community and adds difficulty to predicting planktonic blooms.

Qualitative analysis ("tons, lots, many, few, and none") is useful to know if a taxa might be present at a certain time, but to know the quantity in a population the following simple equation is used… A population is a summation of total recruitment (births-deaths) +(immigration-emigration). In the case of *Gloeotrichia*, immigration and emigration can be summarized by the aforementioned dormant cycle, births or cell division is going to be a function mostly of sunlight, temperature, and nutrients (Fogg, 1973). Death in a controlled environment is more or less well understood(Karlsson-elfgren 2005), but deaths in the wild are complicated by the presence of grazing. Grazing has been observed amongst various cladocerans although it is shown that *Gloeotrichia* is not a choice forage (Fey et al., 2010). What is not known is the preferences that these zooplankton have in their selection of phytoplankton to graze upon, and even less is known about the effects that each food choice has on the organism.

Many cyanobacteria produce cyanotoxins, and many more cyanotoxins are being discovered at a quick rate to where very few cyanobacteria are believed incapable of producing such toxins under the right conditions. It has been found that most cyanotoxins are not secretions but exotoxins released through cell lysis. The introduction of cyanotoxins into the environment appears to be similar to that of lipid-A, and therefore the primary function of many cyanotoxins may not be as a toxin (Chorus, 1999).

It is not known if cyanobacteria are capable of poisoning their predators as a deterrent but if they are this would add another layer of complexity into cyanobacterial blooms. Some predators would likely be less affected than others; therefore imparting a skew in the predatory effects of the presence of some zooplankton over others. Some evidence suggests that Daphnia populations are negatively affected by *Mycrocystis* abundance (Wojtal-Frankiewicz et al., 2014).

2.9 Diatoms

Diatoms are a rapidly multiplying single celled photosynthesizer belonging to the class Bacillariophyceae. As a result, they tend to comprise the first blooms after ice out. They are limited in their persistence by the availability of low soluble dissolved orthosilicate which they use to make their frustules. During blooms diatoms will consume this nutrient at a faster rate than its dissolution into the water column (Godhe and Rynearson, 2017). Common species encountered were Asterionella, Fragilaria, surellia

2.10 Desmids

Desmids are a ranking of green algae, as a result they tend to be either green or sometimes small species appear black due to their relatively large nucleus. They are single celled and easily distinguished by their bilateral symmetry with a connective isthmus bridging their two lobes. Their biomass does not rival that of diatom, cyanobacteria, or Dinobryon blooms, due to the small size of most taxa, but they do have substantial blooms where several specimens can be found on a single frame. Desmids are a persistent plankton and usually a few can be found in any of the samples. Common species encountered include Staurodesmus, Staurastrum, Xanthidium, and rarely Micrasterias, Cosmerium, and Pediastrum.

2.11 Golden Algae

Golden algae, Chrysophytes, can be identified in part by their golden hue coming from their unique pigment fucoxanthin (Peng et al., 2011). similar in structure to xanthophyll or carotene. They are prolific bloomers during colder months on Lake Auburn, but will bloom in warmer months in other waters that are typically labeled as dystrophic. Dinobryon, a colony shaped like a bifurcating tree of champagne flutes, is the principle species found in blooms and in the spring can be so numerous as to obscure the presence of other plankton, often co-blooming with Asterionella. Synura is a species associated with poor water quality, said to smell bad (Healey, 1983). Synura levels are low in Lake Auburn, and are seen in the warmer months.

2.12 Succession

General patterns in seasonal planktonic succession have been observed in deep coldwater non eutrophic lakes and the following is a sequence of those events (Malchow et al., 2001). Some taxa follow more predictable patterns than others. As the season progresses the community tends to become more rich and a compounding of factors makes the middle and end of the season harder to predict. The succession is driven by allogenic and autogenic factors. When considering autogenic succession the general trend is as follows. Fast growing algae and diatoms bloom in spring, followed by zooplankton with high fecundity or winter resting stages that abruptly hatch out which then in turn graze upon these phytoplankton, there numbers varying partly due to the length of the ice-in period (Hrycik,2022). The slower growing of the zooplankton species then become more populous and better represent amongst the quick growing zooplankton. An exponential zooplankton growth rate consumes phytoplankton at a greater rate than phytoplankton growth. There is a crash in phytoplankton that results in a clear water equilibrium phase that persists as inedible or hard to eat phytos like *Gloeotrichia* or filamentous algae build up their numbers. Zooplankton lose weight, become small, and reproduce more slowly, thus reducing their numbers. Predation on grazers, presumably by fish, selects preferentially the larger zooplankton, decreasing the size of specimens but also the abundance in larger crustaceans. Zooplankton recycle nutrients, and these become nutrients non limiting to the growth of phytoplankton again if there was a deficiency earlier in the season. The growth of the plankton community becomes ever complex with larger chlorophytes becoming common. These phytoplankton deplete free nitrogen to a seasonal low and nitrogen comes closer to parity to phosphate levels. As the ratio between nitrogen and phosphate becomes closer to 1:1 large relatively inedible slower growing diatoms replace green algae. Diatoms deplete the water of silica and are replaced by cyanobacteria or dinoflagellates depending on nitrogen and phosphate levels (Dagenais-Bellefeuille and Morse, 2013). Ultimately any large blooms in dinoflagellates give way to cyanobacteria colonies. Pressure from predators and food scarcity continues on larger crustaceans and these plankton give way to a rise in rotifers which are better adapted against mechanical clogging in a filamentous rich environment. Weather starts to cool and autogenic factors start to dictate the planktonic assemblage. Diatoms and algae filaments that are hard to eat and well versed in being mixed in the column increase in numbers. The mixing event brings with it a resurgence of nutrients and small edible fast growing phytoplankton, which feed on zooplankton increasing their numbers and size. Lower light and temperatures of autumn slows down production, and inturn decreases the numbers of primary producers and their consumers. Stress conditions lead to sexual reproduction in pathogenic species and the creation of resting stages such as ephippia (Sommer, 1986). In general phytoplankton populations are more variable in relation to phosphorus with nutrients being a greater limiting factor than light in oligo and mesotrophic lakes cyanobacteria and dinoflagellates appear to be more prevalent in higher biomass lakes (Sommer, 1986). The short life cycles, loading of vegetative states, fast reproduction, and high fecundity of plankton make weekly sampling an incomplete snapshot of the continuous succession, for example, grazers can multiply so quickly that they consume 100% of available forage daily (Haney, 1973).

3.Methods

3.1 Sample collections

Samples of Lake Auburn water were collected, typically in mornings without heavy wind or bad weather that would preclude the boat from launching, via a plankton net (type and size 80 micron) vertically towed at a depth of 0.5 meters. Samples were washed into amber bottles with tap water and a dropper's worth of Lugol's solution added. Bottles of samples were labeled and stored on a shelf at room temperature till needed.

3.2 Sample analysis

Samples were counted one at a time by first straining contents in a fabricated filter made out of a 80 micron mesh screen sandwiched in between two pieces of 2" PVC pipe placed inside of a beaker. Lugol's solution filtrate was disposed of according to hazmat protocols. Tap water was then added to the sample bottle, bottle capped, shaken, and then drained into filter, and repeated 2 more times. Using a wash bottle filled with tap water, samples were squirted and concentrated to a corner of the filter cup where they were next irrigated out of the filter by inverting the cup over a sample tray while flushing with a limited amount of water from an irrigation bottle.

The sample tray used was a Falcon 35112 square intergrid petri dish, 100mm square with 15mm cells. It was important to consider that sample components had different densities and cross sections, and therefore sorted themselves when the tray was sloshed. To avoid concentrations in regions of the tray that would skew qualitative analysis that arise when only considering a taxa for a portion of the tray, the tray was aggressively sloshed to thoroughly suspend sample particles, and then allowed to settle. With some skill a relatively even distribution could be achieved after a few tries with one rock immediately after aggressive agitation.

The tray was viewed under a binocular microscope under 2x objective for counting purposes and could be zoomed up to 6x for identification purposes. Trays were traversed across the stage in a lawnmower type pattern with the margins being included last for assurance in a complete and accurate count.

Gloeotrichia colonies were recorded using a single battery of clickers and were annotated for two parameters, cell size/condition and species. Size/condition options were either Complete, Grazed, <½, or >½. Complete was defined as a mature colony that appeared to have the vast majority of vegetative trichomes intact with a center that was not in advanced stages of division (could not see light through the length of division fissure). Grazed colonies were those that had fully intact centers but had heavily grazed vegetative cells. Any colony that did not have a fully intact center and or was missing a wedge of akinetes/heterocysts were categorized as either <½ or >½, depending on if the majority of a sphere was present or not. Any bundles of filaments or single trichomes were considered to be <½. The second annotation was for speciation of *G. echinulata or G. pisum* where species could be discerned.

Final counts were recorded in a log book, along with qualitative assessments of other identifiable members of the community or other significant organic structures. The following five tiered scale was used; tons, lots, many, few, and none. Tons was defined as any taxa that dominated the tray in such a way that it appeared scores of times per grid. Lots was defined as a taxa that appeared more than 4 times per grid. Many were defined as a taxa that appeared more than 2 times per grid. Few was defined as appearing less than twice per grid while also being more frequent than other taxa that could not be listed in the limited space in the notebook table. None for larger sized objects and taxa were defined as being absolutely none per sample, while None for smaller objects meant that they were present less than any significant amounts of Few. None was seldom recorded, instead if an object or taxa was not listed then it was assumed to be None. The entire tray was not counted for each qualitative measurement. Instead, it was typical to count one pass in the lawnmower grid fashion to get an idea of the value. After sample trays were counted, samples were returned to the amber bottle via plastic funnel and wash bottle.

For qualitative analysis, A sample site picture was recorded on binocular settings for the microscope via Captivision software for every sample counted. A complete even distribution of particles across the trey could not be achieved and so one of the four center grids was chosen for the picture to get a more standardized comparison across sites. It should be noted that the center of the dish tended to show an over representation of particles, especially of denser specimens.

4. Results

Dolichospermum and *Gloeotrichia* are the dominant species found in Lake Auburn in the summer of 2022. *Aphanizomeno*n was often present but rarely in numbers more than 3-6 per sample. *Merismopedia* was rarely found in samples and it was almost never found in abundance of more than one colony per sample. *Coelosphaerium* and *Woronchinia* matching closely in the shape and size of their individual colonies also matched closely with their relative abundance, not being present for most of the season but becoming more abundant than *Gloeotrichia* at the conclusion of the summer. *Microcystis* underwent two blooms, one corresponding with the bloom from *Gloeotrichia* the other occurring during the end of the year as the lake cooled and on track to become the dominant coldwater cyanobacteria in the winter months.

Dolichospermum was present in three distinct morphs: straight single stranded trichomes, single stranded spring-like spirals, or single stranded “spaghetti”-like knots that often included colonies of *Vorticella* or occasionally was a foothold for *Conochilus*. Although *Gloeotrichia* biomass was greater than *Dolichospermum* at the height of its late summer bloom, *Dolichospermum* colonies were always the most abundant cyanobacteria for the months sampled in 2022 across all sites (Figure 1). *Dolichospermum* was the most ubiquitous taxa; being present as either tons or lots in all samples, accept being found a few times as many or few at the onset of springtime sampling, therefore *Dolichospermum* also had the least variability of all the common taxa in this study and lesser exaggerated blooms ranging in corrected relative abundances of 6-16.

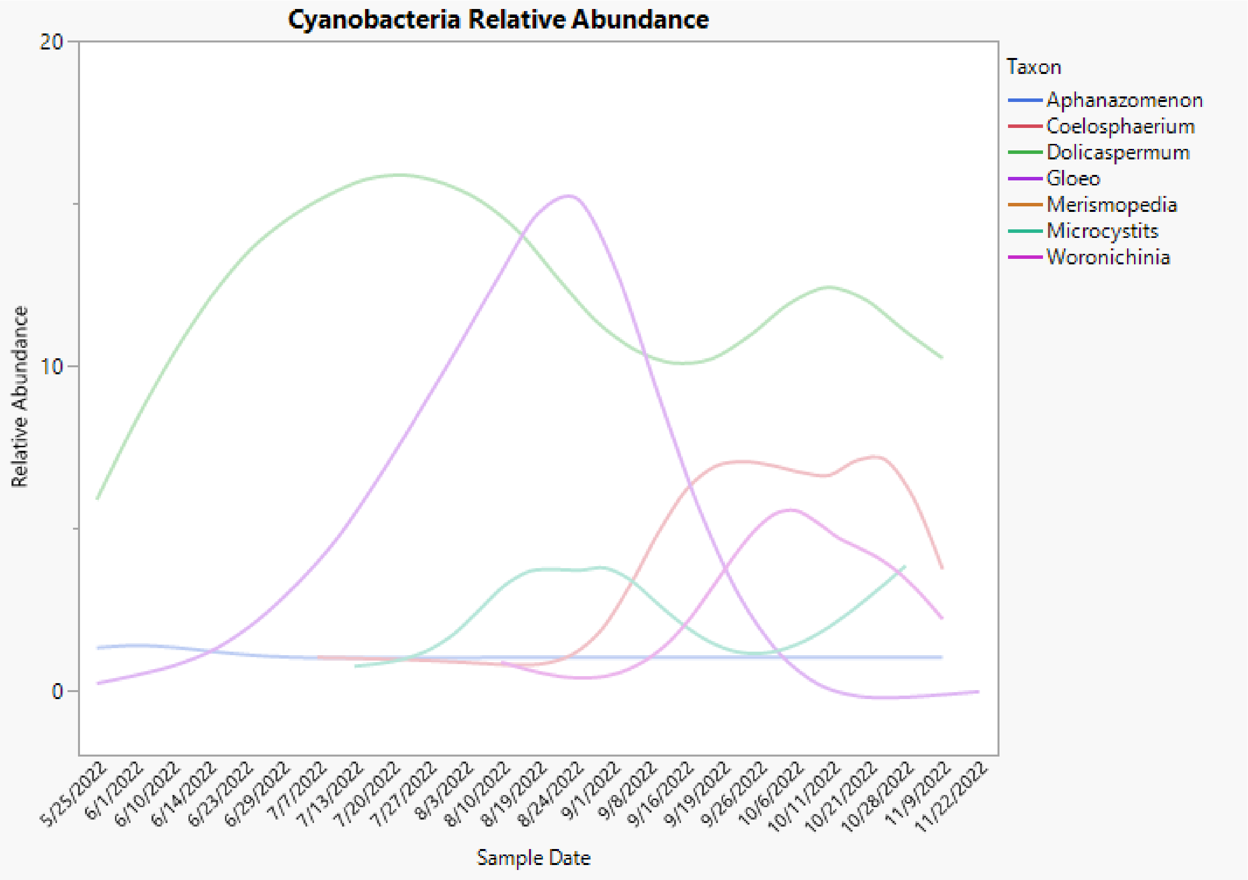

Figure 1. Relative abundance of cyanobacteria in Lake Auburn open water season 2022. Abundance of Non-*Gloeotrichia* was scaled from semi quantitative counts (see Methods). *Gloeotrichia* colony counts were divided by 30 to display on the same scale as other cyanobacteria. Counts across sites #4,5,8, and 32 were averaged and are displayed with spline fit.

Gloeotrichia contrasts with *Dolichospermum* as being the most pronounced bloom of all the cyanobacteria and only second to *Asterionella* as the most dramatic planktonic bloom in the study. *Gloeotrichia* has zero presence in the spring and fall, and is unique to be the only cyanobacteria counted to have zero presence at the conclusion of 2022 season's sampling. (Fig. 2). Colonies appear to divide at about the same time they migrate to the surface as seen by the matching shapes and temporal placement of the plots in Figure 2, with the colony fragments peaking a week before full colonies peak.

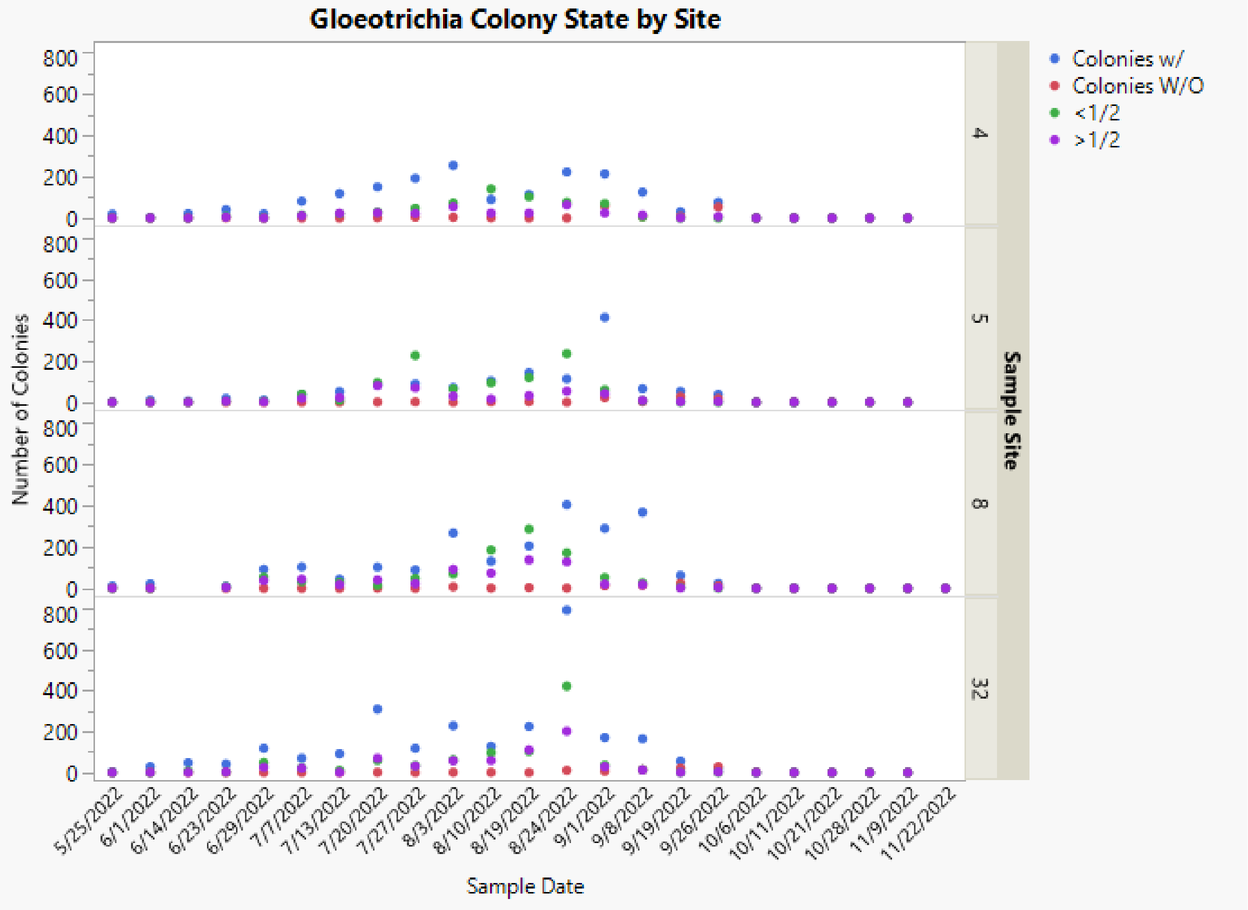

Figure 2. Mean total count of Gloeotrichia colony types found in Lake Auburn open water surface samples across sites #4,5,8,and 32 from season 2022 displayed with spline fit. Full colonies (blue), colonies that have been grazed on (red), divided colonies that are more than half size of mature colony (purple), colony less than half sized mature colony (green).

Water clarity varied from 12 meters to 0.8 meters from the open water season (Figure 3). Water clarity hit its maximum at the conclusion of the diatom bloom in the early summer (Figure 4.), and finished the clear-water phase at the end of July. Water clarity continued to diminish throughout the sampling season until fall turnover. Gloeotrichia colonies peak once Secchi readings drop below 5 meters, but subsequent drops in colony count do not correspond to an increase in Secchi depths (Figure 3). *Gleotrichia* colony count and Secchi depths were only negatively correlated to a value of -0.18 using the REML method.

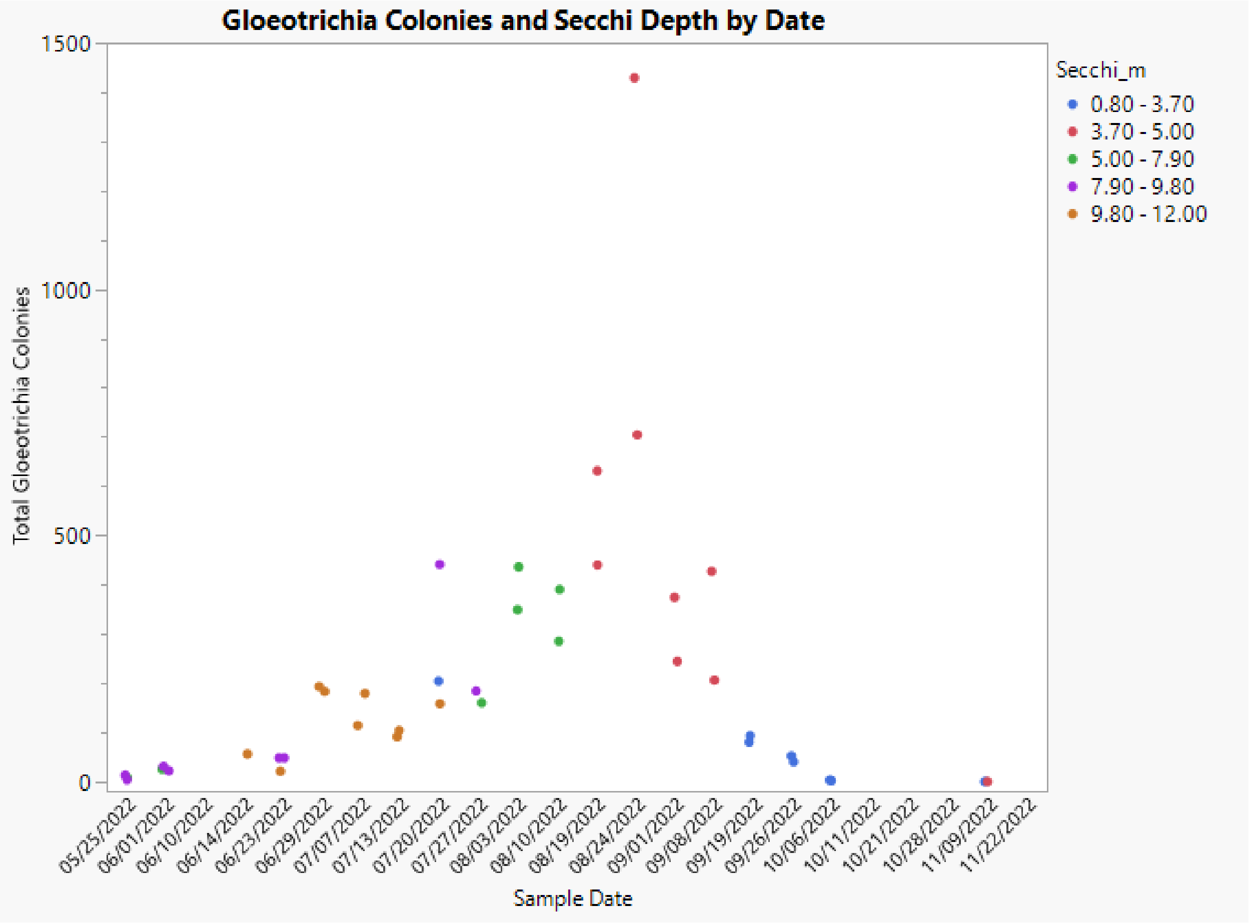

Figure 3. Scatter plot of total count Gloeotrichiatrichia colonies per site found in Lake Auburn open water surface samples for sites #8 and #32 from season 2022. Corresponding Secchi depth for water at sample site displayed by color.

The major non-cyanobacteria Algae taxa found were the diatoms; *Asterionella*, and *Fragilaria*, the golden algaes; *Dinobryon* and *Synura*, the desmids; *Staurastrum, Staurodesmus*, and *Xanthidium*, the long filamentous algae such as *Spirogyra, and Zygnema*, collectively recorded as “Algae Filaments”, and the flagellated green algae colonies; *Eudorina*,and *Volvox*. Taxa that were present in some samples but not common were Closterium, Cosmarium, Gonium, Pediastrum, Pleodorina, and Surirella. Furthermore it was almost certain that Chlorella was present but taxa of this size were too small to identify and so were ignored.

Asterionella was the dominant taxa in the spring, appearing to have climaxed its bloom immediately before the sample season began in the spring. The abundance of Asterionella and Dinobryon was so intense in the first weeks of sampling that they had a tendency to obscure other taxa and made counting difficult, furthermore the quantities of Asterionella made filtering a slow process and a bloom of any higher magnitude would have required splitting the sample to count. Asterionella abundance quickly trailed off and was no longer detectable in samples beyond July. Fragilaria had the inverse relation and started off undetectable though possibly present in small quantities and its corrected mean abundance increased linearly until abruptly disappearing from the samples for the rest of the season. Asterionella drop off and Fragilaria's abrupt disappearance mark the end of the diatoms in the samples and do not reappear even after the fall turnover.

Because of the similarity in morphology and the assumed physiology and niches across the green algae filaments combined with their lower individual abundances they were all counted as one. Algae filaments follow a similar trend to the diatoms, but show two different peaks before finally extinguishing in the fall.

The problematic Synura underwent two blooms; one in the spring, and one in the fall with a complete disappearance in the summer. Dinobryon went through a similar pattern but was more prevalent. Furthermore, Dinobryon did not abruptly disappear in the fall but had a hockey stick like growth ending the year denser than it started and presumably peaking after the conclusion of sampling in the fall of 2022.

The Desmids, a challenge to distinguish amongst themselves, all follow a similar pattern for not being present in samples at the onset of sampling, then blooming in mid summer to taper off in the fall in a classic bell shaped curve. Flagellated colonies of *Volvox* and *Eudorina* bloomed in the fall and were ontrack to increase in their numbers in the coldwater phase of the fall beyond the sampling window.

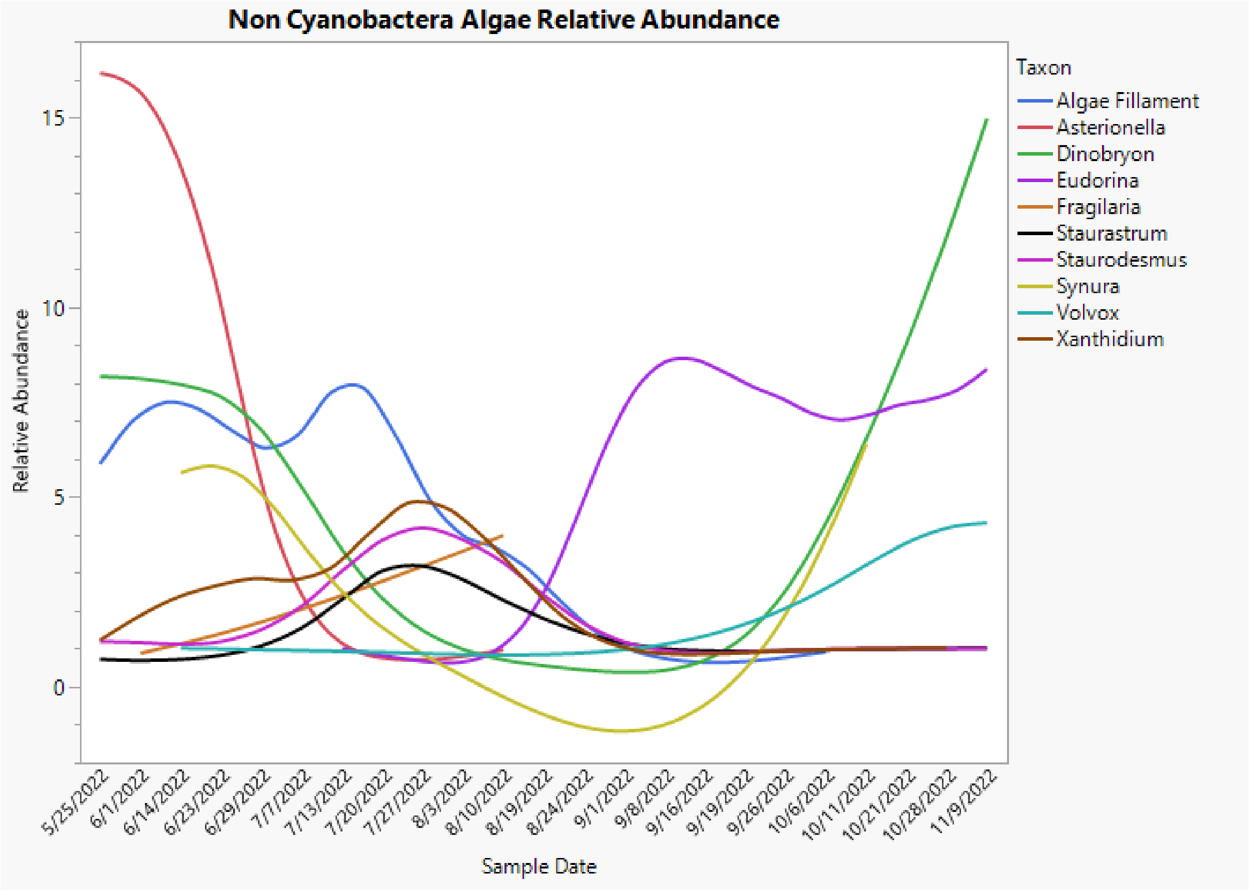

Figure 4. Relative abundance of Non Cyanobacteria algae in Lake Auburn surface samples from open water season 2022. Counts across sites #4,5,8, and 32 were averaged and are displayed with spline fit.

Bosmina is the most abundant cladoceran throughout the sample period with exception to the warm water period of summer where *Daphnia* becomes more abundant followed briefly by *Diaphanosoma*. *Chydorus* is the only cladoceran on an upward trend at the conclusion of sampling. (Figure. 4).

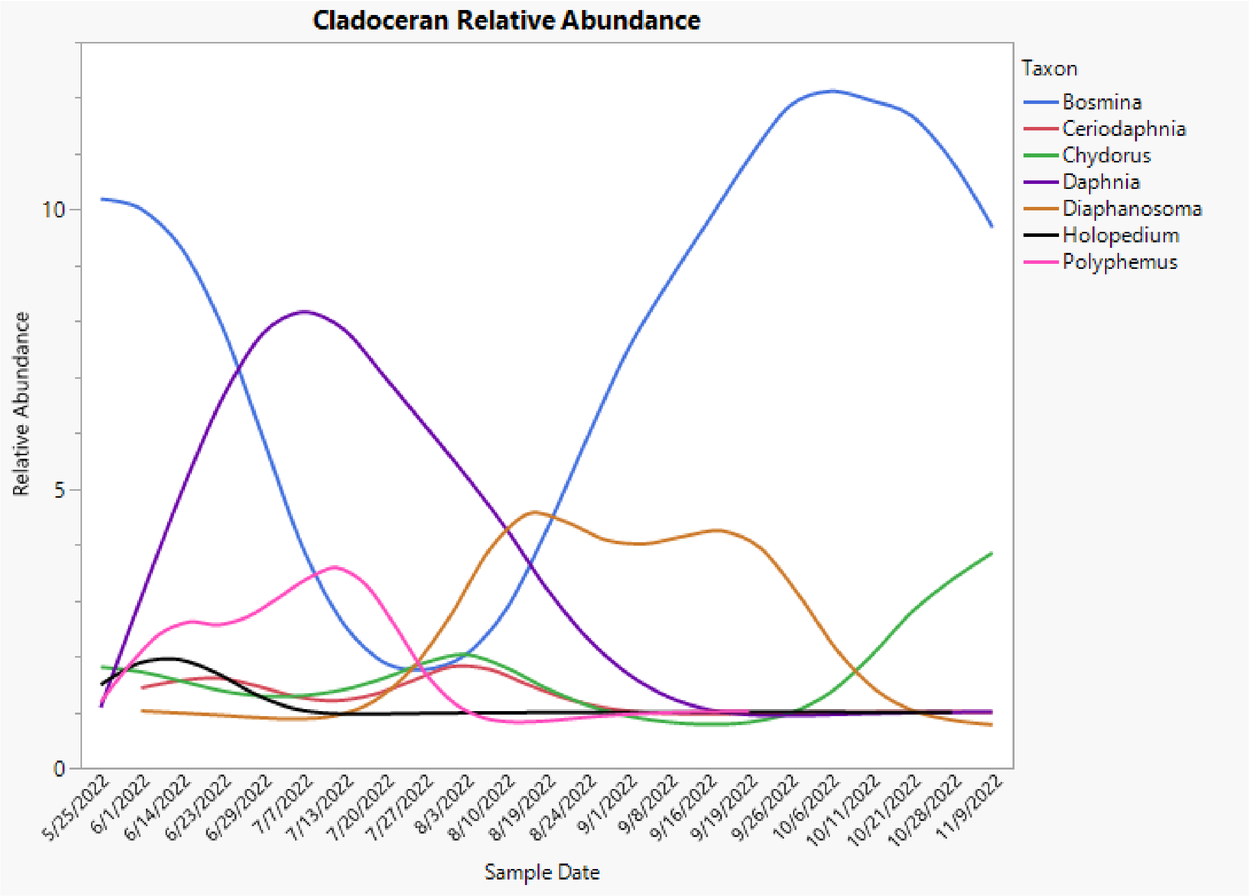

Figure 5. Relative abundance of Cladoceran in Lake Auburn surface samples from open water season 2022. Counts across sites #4,5,8, and 32 were averaged and are displayed with spline fit.

Adult copepods in Lake Auburn during the 2022 open water sampling period were observed to be exclusively *Calanoida* and *Cyclopoda*. Nauplii instars were often indistinguishable to a further taxon beyond copepods and so are the combination of both cyclops and calanoids. All three taxa types increase throughout the open water season with nauplii having a pronounced bloom and dieback in the fall (Figure. 4).

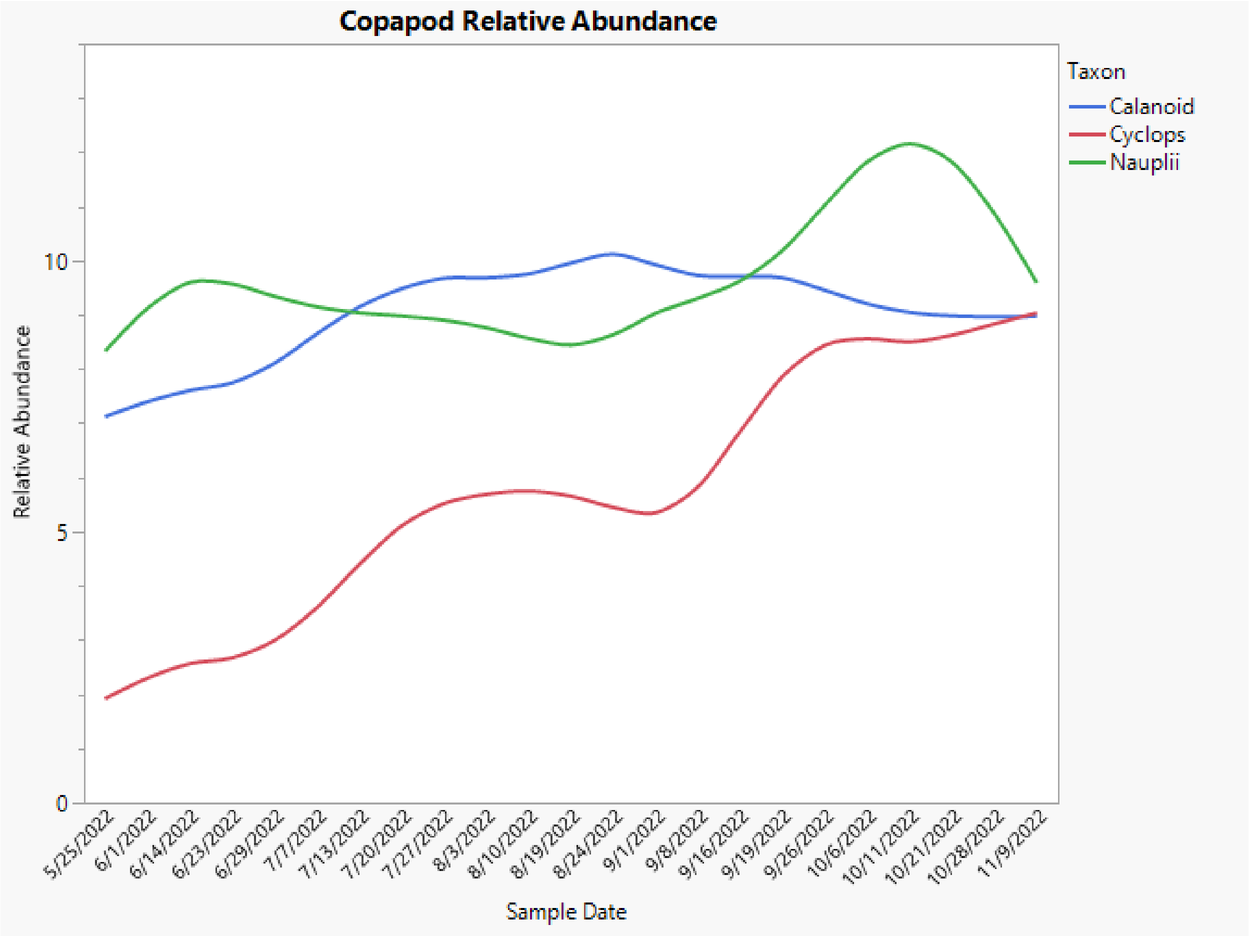

Figure 6. Relative abundance of Copepods in Lake Auburn surface samples from open water season 2022. Counts across sites #4,5,8, and 32 were averaged and are displayed with spline fit.

Rotifers and ciliates represented by the sole taxon *Vorticella* are most abundant at the beginning and end of the 2022 open water sampling season, with the exception of the colony forming *Conochilus* which is most abundant late summer (Figure. 5)

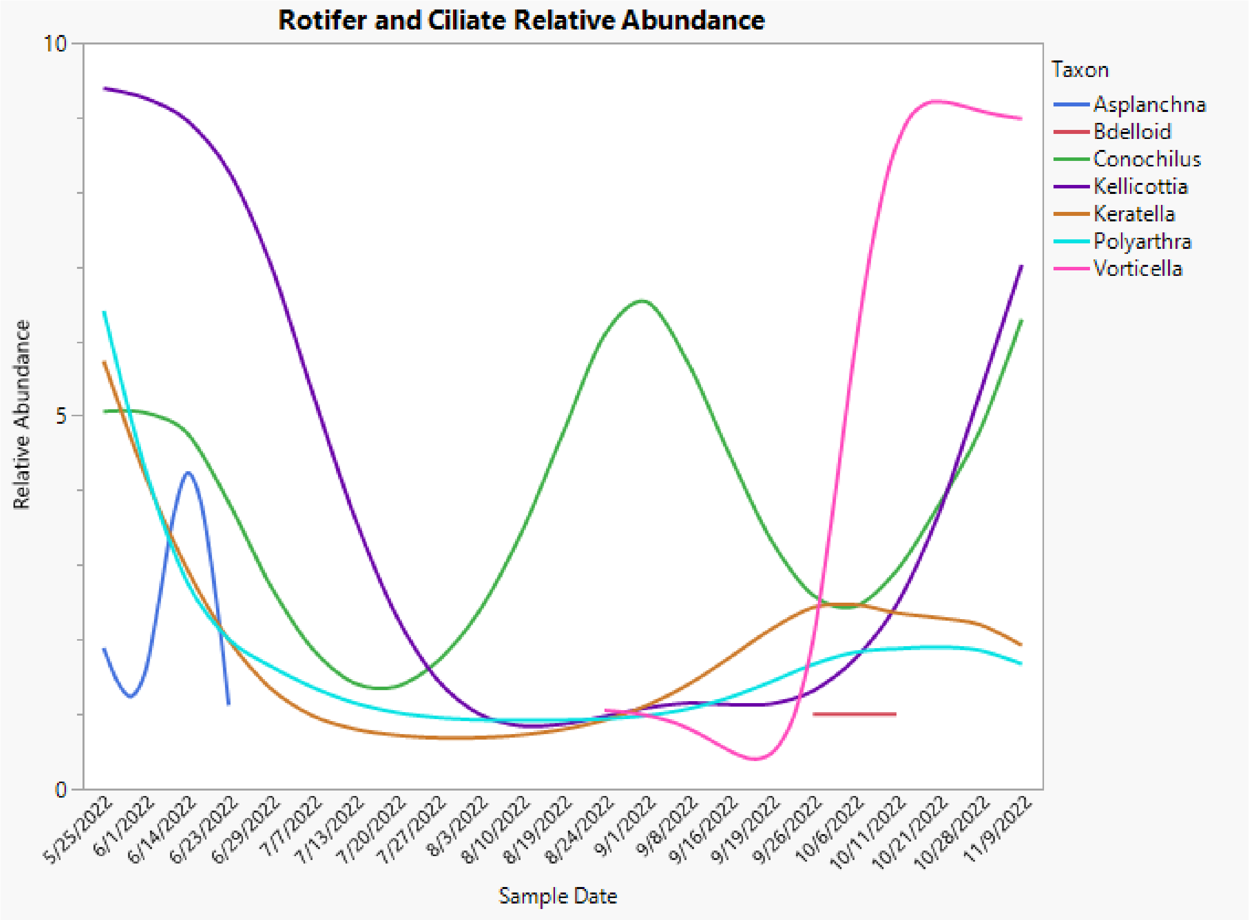

Figure 7. Relative abundance of rotifers and ciliates in Lake Auburn surface samples from open water season 2022. Counts across sites #4,5,8, and 32 were averaged and are displayed with spline fit.

Copepods are the most abundant zooplankton throughout the open water season with the exception of rotifers being exceptionally abundant in the springtime. Cyanobacteria tend to be the most abundant primary producer, but are not as prevalent in the colderwaters of the spring where the mixotrophic dinoflagellates and fast colonizing diatoms are more abundant (Figure 7).

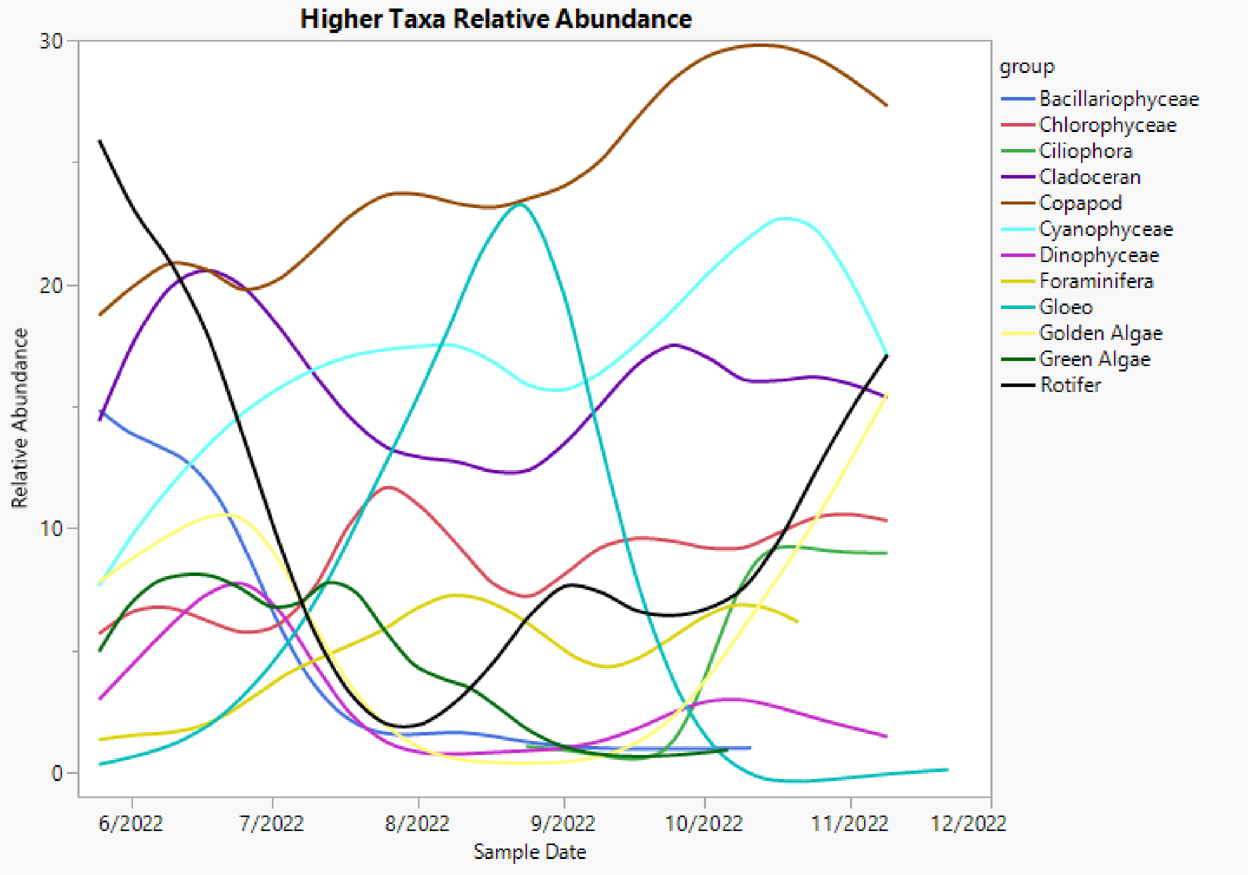

Figure 8. Relative abundance of higher taxonomic groupings of phytoplankton and zooplankton in Lake Auburn surface samples from open water season 2022. Counts across sites #4,5,8, and 32 were averaged and are displayed with spline fit.

Trendlines tend to follow the same patterns across sites regardless of stream influence. Some notable variability is the high abundance of springtime golden algae that was only recorded at site #32. Site #32 is also notable in that it has the highest *Gloeotrichia* to copepod ratio in the summer bloom out of all four sites. *Gloeotrichia* blooms are larger and more pronounced in the deeper water sites than from the lotic sites. The largest non *Gloeotrichia* cyanobacteria bloom occurs in October at site #5 (Figure #7).

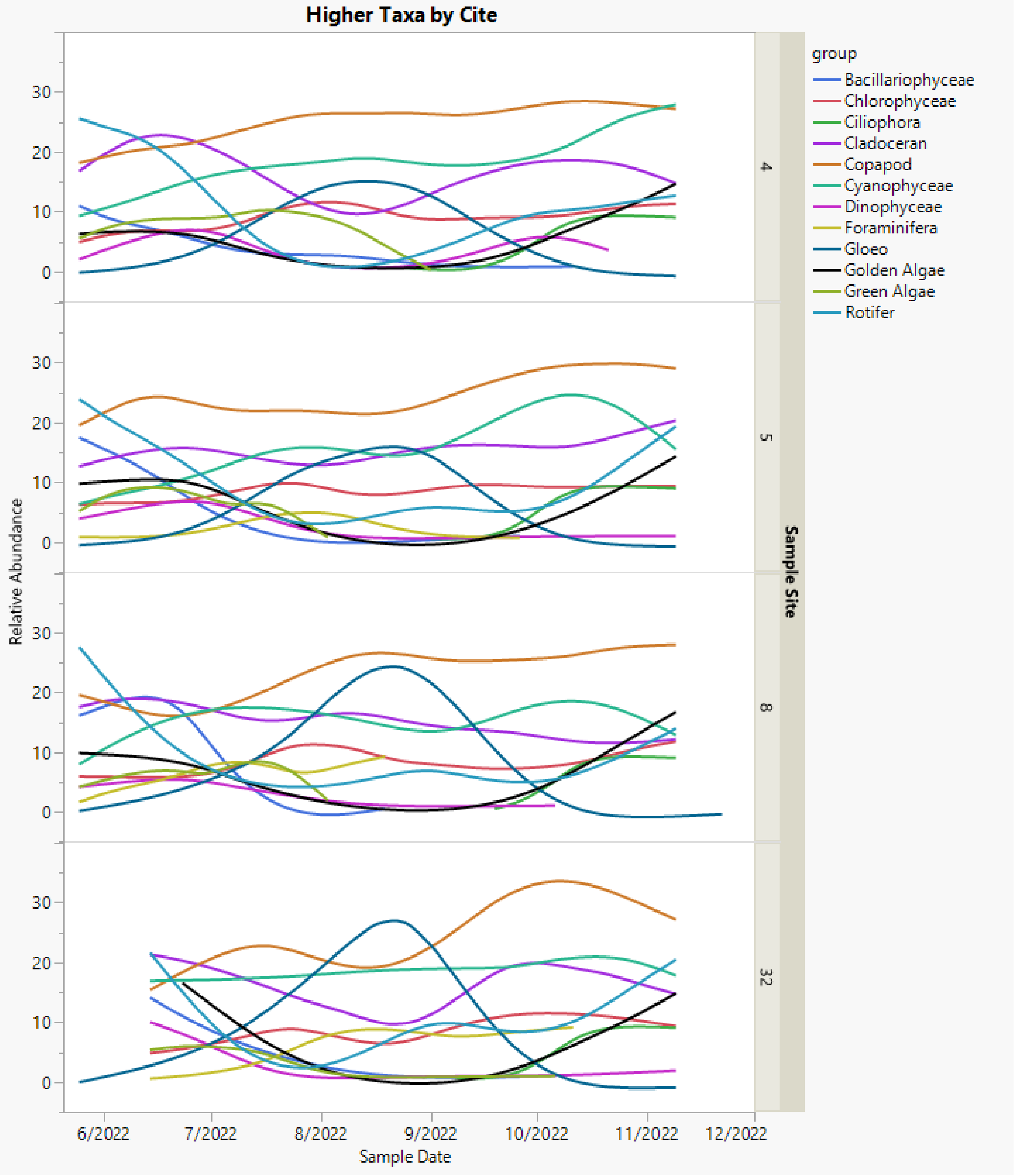

Figure 9. Relative abundance of higher taxonomic groupings of phytoplankton and zooplankton in Lake Auburn surface samples from open water season 2022. Counts across sites #4,5,8, and 32 are reported separately and are displayed with spline fit. Sites #4 and #5 are heavily influenced by logic inputs; sites #8 and #32 are deepwater limnic sites.

5. Discussion

5.1 General Observations

Lake Auburn largely followed expected trends in planktonic succession. Diatoms bloom out in the spring and are quickly depleted. Asterionella, although not the only diatom recorded, was the only diatom to occur in great enough numbers to be reported in Figure 5. Due to the lack of richness in the diatoms they appear to be under-reported as a group. This is due to Asterionella being so abundant during their bloom that there wasn't a value equitable to their density in the samples. A taxon grouping of extreme high abundance when compared to a taxon grouping such as copepods composed of three lower taxa of high abundance will appear to be less abundant even though total abundance of the one extreme taxa is observed to be higher. This is the case for the diatoms. Field notes bare out that Asterionalla alone accounted for the majority of identifiable taxa in springtime samples.

The fast reproducing crustacean *Bosmina* is in a bloom at the same time as the diatoms. Bosmina is an efficient grazer with a preference for forage in the size range of up to the size of *Asterionella* colonies, and is capable of dismantling the colonies by use of its legs and mandibles, making *Bosmina* one of the most capable graziers of larger phytoplankton colonies (Bleiwas & Stokes,1985). Although direct herbivory of *Asterionella* was not observed due to samples being preserved before viewing, it is likely that *Bosmina*, and to some extent, the rest of the adult crustaceans are responsible for the initial rapid decline of diatoms, before the unfavorable warm water period sets in. It has been shown that when planktivorous fish are introduced into a mesocosm experiment that the larger cladocerans are eliminated and chlorophyll levels increase suggesting that the larger crustaceans are more efficient at eating the diatoms (Sorf et al., 2014). *Bosmina*'s summer minimum aligns closely with the disappearance of diatoms in the water column and their two curves match closely during this time period. It is perhaps the depletion of diatoms as a forage and the longer life cycle of *Daphnia* that allows *Daphnia* to outcompete *Bosmina* and become the most abundant cladoceran. *Holopedium*, While not nearly as abundant as *Bosmina,* follows a similar spring maximum. *Holopedium* has delicate arms and many *Holopedium* are considered to be nano planktivores. It is perhaps this niche that explains why *Holopedium* is not more prevalent, and why we see its maximum in the spring during the decline of the diatoms.

It is unclear where the early summer *Daphnia* were recruited from. *Daphnia* can be life-birthed via parthenogenesis or can be hatched out from sexually produced resting eggs called ephippia (Thorp et al., 2014). *Daphnia* are capable of surviving the winter in low numbers and it's these adult females that could have seeded the population of summer *Daphnia*. It is also possible that ephippia hatched out of sediments and that these juveniles matured and reproduced to give rise to the summer maximum. To answer this question for future seasons trap samples can be taken from the bottom of the lakes and audited for intact ephippia and empty ephippia cases to give a sense of hatching rates and timing. Cladocerans can also be recorded as either small or large, with the assumption that the small cladocerans would be freshly born or hatched juveniles. The third measure that can be taken to discern if *Daphnia* are being recruited from ephippia or parthenogenesis would be to record the percentage of fall and winter females that are laden with ephippia to give us a sense of the prevalence of ephippia production.

The end of the clear-water phase is marked by the increase of *Dolichospermum*. *Dolichospermum* and *Gloeotrichiatricha* both were observed to be in higher abundance than any non nitrogen fixing phytoplankton immediately following the clear water phase. Green algaes were expected to become dominant during this period if N:P ratios were relatively high which is typical in the lakes of this region. The absence of green algaes being abundant in this period suggests that there could be a higher concentration of P relative to N than typically found in dimictic mesotrophic lakes. The typical trend of the non nitrogen fixing phytoplankton to reach their apex in early summer is evident in our samples. These maximums can be seen through July for green algae filaments and the desmids; reinforcing the process of non-nitrogen fixing phytoplankton depleting the water of N relative to P, and thus turning P into the limiting nutrient.

As the season progresses photosynthetic plankton scrub nutrients from the water. *Dolichospermum* abundance marginally declines likely due to epilimnetic P becoming the principle limiting nutrient for most cyanobacteria. *Gloeotrichia's* population surpasses *Dolichospermum* to become the most abundant phytoplankton and cyanobacteria in August, and remains the most abundant until its downward benthic migration in September. The sudden surge is likely do to *Gloeotrichia's* ability to migrate in the water column; lying on bottom in the colder months and absorbing high concentrations of P to use later when it fills its cells with gas and buoys itself to the surface of the lake where it undertakes photosynthesis and consumes its stocks of P in rapid cell growth and division. The observation that *Dolichospermum* population reaches its apex before *Gleoetrichia* colonies suggests that while we are concerned with elevated levels of P in Lake Auburn; that epilimnetic P is still a limiting nutrient, otherwise *Dolichospermum* might peak around the same time when *Gloeotrichia* reaches it apex. *Gloeotrichia'*s population is able to reach its peak when surface water P is more depleted because of its higher cellular concentration of P that it brought with it from the hypolimnion. An increase of P in Lake Auburn could result in a closer temporal overlap in *Dolichospermum* and *Gloeotrichia* population maximums in surface samples.

Gloeotrichia colonies were expected to migrate to the surface as mature and intact colonies, before dividing and growing. What was observed was that partial colonies, those of greater than or less than half a colony, reached a maximum about a week prior to the full colonies (Figure. 2). There are several explanations for this. It's likely that the colonies start dividing before reaching the surface and so they initially appear at the surface as less than a full colony, and then mature into a full colony at the surface in about a week's time. No other rounds of division commonly occur before the full colonies migrate back down into the sediment. The second, and less likely scenario, is that some *Gloeotrichia* colonies divide while still benthic before recruiting to the surface population and rise up first, followed a week later by colonies that recruit as full colonies. For this second scenario to be plausible we would likely see this behavior in other lakes and this is not what we see in Lake Sunapee trap samples where rising benthic colonies are almost entirely observed as full colonies. *Gloeotrichia* terminal vegetative cells are wispy and provide a good substrate for other more nutritious microplankton. The implications that the colonies are dividing sooner than the full colonies are present in mass at the surface means that *Gloeotrichia* is a less nutritious food source than predicted and benefits its grazers less. A bloom in cladocerans, split almost exclusively between *Diapanasoma* and *Bosmina,* temporarily corresponds to the maximum number of grazed *Gloeotrichia* colonies (Figures 4,6). This reinforces that *Bosmina* is a heavy feeder of Gloeotrichia when other forage is scarce and also implies that *Diaphanosoma* could be feeding on *Gloeotrichia* which was not discovered in the literature. If *Diaphanosoma* did not feed on *Gloeotrichia* then we would expect for the maximum of grazed colonies to temporarily line up more closely with the fall bloom of *Bosmina*. *Bosmina* is likely resorting to feeding on *Gloeotrichia* at this time because the other available phytoplankton forage candidates observed are too large and smooth to break pieces off of in the case of the flagellated green algaes, too large and rigid to do the same as in the case of *Dinobryon*, or coated in a mucilage as in the case of the other plotted cyanobacteria colonies. Bosmia also is likely to avoid feeding on *Dolichospermum due to its* toxicity and is thought that feeding heavily on Gloeotrichia can stunt growth and sometimes results in sterility (Wilson et al., 2006).

Towards the cooling phase in October the community becomes more diverse reaching its maximum observed diversity at the conclusion of the 2022 sampling period (Figure. 6). With this there is an increase in the effectiveness of herbivorous plankton to graze on or consume a specific taxa of plankton of their preference. We see a trend for smaller plankton to either not be present at this time, to dwindle in numbers, or to form protective colonies. Those taxa which naturally form defensive shape and sized colonies tend to reach their maximums at this time in the fall. The cyanobacteria *Worochinia* and *Coelosphaerium* are both roughly spherical colonies held together by a gelatinous matrix exudate whose size can be comparable to a small *Volvox,* precluding it from grazing by the zooplankton herbivores (Moustaka-Gouni and Sommer, 2020). *Worochina* and *Coelosphaerium* reach their maximum in October culminating in the apex of cyanobacteria abundance (Figures. 1,6). The flagellated green algae taxa represented by *Eudorina* and *Volvox* increased in abundance a month earlier than *Worochinia* and *Coelosphaerium*, but unlike those colonies the flagellates increased in abundance up through the last day of sampling. It's possible that these two taxa could persist in large numbers alongside the copepods through part of the coldwater season; sampling later in the season would be needed to confirm this.

Conochilius is an interesting rotifer in that the adults are colony forming. *Conochilius* population maximums display a sinusoidal pattern. During the course of the clearwater season, *Conochilius* population decreases and then reaches a maximum during peak summer only to taper off at the end of summer and increase again in the fall. *Conochilius* appears to have two maximums, one in the summer, and the other in the winter. *Conocilius* is an unusual rotifer in that the adults that can be seen in abundance in the winter months from spherical colonial units, whereas the juveniles exhibit the typical solitary behavior and can be seen in abundance in the summer. Calanoids are a common predator of Conocilius, however almost no correlation can be seen between these two populations (Figures 4, 5). This is likely due to the protection offered to the adult *Conochilius* when in a colonial setting. The weak apex of Calanoid abundance corresponds to the pronounced summer maximum of Conochilius and this could be a principal driving force in the populations of the longer lived Calanoid. The biomass of Conocilius may be under represented in the colder months, this is due to the nature of counting; a colony of Conocilius is weighted equal to a solitary individual and so the number of winter adults is higher than reported. This should be taken into account when considering *Conochilus* populations in comparison to other plankton in the community.

5.2 Shortcomings

The vast majority of *Gloeotrichia* identified down to the species were G. *echinula* with only rare examples of *G. pisum* confirmed. About half of *Gloeotrichia* colonies could not be identified down to the species due to the colonies being in the midst of dividing or being a post divided fractions of a full colony. Identification down to the species level is feasible in trap samples that collect the full benthic Gloeotrichia colonies, but it was found to not be practical in surface samples where visual species identification is often not possible. For this reason the recording of Gloeotrichia species was halted part way through the counting process and species were not recorded for all sites and are not reported here.

Filamentous algae trichomes can be a challenge to identify. Not all trichomes could be identified to a taxon beyond *Charophyceae* and so it was decided that all Charophyceae trichomes were to be recorded as filamentous algae. The double apex of the taxa plot suggests that there could be two taxa blooming at two different times. To gain a better understanding of these timings it is suggested that future studies record the taxa Spirogyra and Zygnema, two easy to identify taxa, as separate taxa from the other filamentous algae.

Synura can be a challenge to identify. Each cell in the colony has a classic teardrop shape and the colonies are typically spherical, the trouble lies in noticing it in a field of *Dolichospermum* that is consistently a background taxon. *Synura* in Lake Auburn were significantly less abundant than the spaghetti form of *Dolichospermum* and so could easily be looked over as *Dolichospermum*. Future studies should take note of the slight difference in color with *Synura* being a deep golden color. Under full brightfield and front lighting the color difference becomes apparent and *Synura* are readily identifiable. As a result of this challenge in identification *Synura* were likely looked over in the first sites counted, (sites 4, 8, and 32), and are likely under-reported.

Plankton samples were only taken from surface grabs. Although we have an understanding of what plankton are in this layer of water at these general times, we do not have a true sense of what the whole column looks like. The assumption that the planktonic assemblage is homogenous is not a fair assumption, and we have addressed this thus far with the migrations of various cyanobacteria, but this is true for most plankton. There is a phenomenon of zooplankton patchiness which explains that zooplankton will not only migrate up and down the column in search for: food, avoidance of predators, and optimum temperatures; but also that currents can microclimates can cause pockets of different taxa densities throughout the lake (Folt and Burns, 1999). For this reason future sampling concerned with auditing the whole plankton community should consider sampling from multiple depths at several locations.

There are many forcings that shape the planktonic community they were not looked at in this study, and are a challenge to measure. Such a list would include but not be limited to allelochemicals in the water (Gross, 2003), algicidal bacteria (Meyer et al., 2017), nutrients and exudates in the phycosphere (Gubelit and Grossart, 2020), plankton too small to identify with 120X magnification including parasites (Grami, et al., 2011), non-cyanobacterial diazotrophs (Riemann et al., 2022), and Fungal parasites like chytrid (Sime-Ngando, 2012).

5.3 Future steps

Dolichospermum has many species and polymorphs. Some species' trichomes are straight, some are spring shaped, and some still are tangled like cooked spaghetti. *Dolichaspermum* had an early summer and a late fall apex. It would have been interesting to note if these two blooms were from separate or similar taxa, by recording polymorphs of *Dolichospermum* we are able to tell with a greater degree of certainty if this is the same species blooming twice or two separate blooms from two separate species. Furthermore *Vorticella* was most commonly found in association with the spaghetti morph of *Dolichaspermum*.It would be interesting to note any correlation they had by being able to plot their populations together.

Sampling was concluded shortly after fall turnover in 2022, but did not continue to ice-in of that season. As a result the picture of the open water succession is not complete. To get a more accurate depiction of the late fall trends it is important to continue sameling further into the year. It is recommended that in future years of Lake Auburn study, that include in depth looks into the planktonic community, should sample subsequent weeks into the season. There are several taxa of interest for the coldwater season and corresponding questions. Such as, do Copepods become the dominant zooplankton as we might expect from the literature and data (Figure 5.)? At what point in time and maximum do Chydorus, *Kellicolita, Conochilius, Dinobryon, and Synura* stop increasing in abundance? Is there a second bloom in the cold water for diatoms or maybe a small green algae that would support the herbivorous plankton that overwinter the water column.

In future samplings size bins for plankton can be created and counted. THis can be in addition to regular taxa count, or in lieu of in the case of an inexperienced counter. It has been shown that the shift in size of the plankton community is an excellent predictor of the changes in nutrient availability with small phytoplankton increasing in abundance at a greater rate than larger phytoplankton or small zooplankton (Cottingham,1999). Furthermore it was found that selecting for larger grazers, specifically cladocerans, allowed for more efficient grazing and less abundance and biomass of phytoplankton (Cottingham et al., 1997). Not only can these observations tell us about water quality but might also be able to tell us about Lake Auburn's planktivore community which has yet been studied from plankton samples.

5.4 Conclusion

An ability to predict future blooms on Lake Auburn would be helpful in managing the lake's water quality and protecting against unexpected rises in NTU's. The planktonic community is complex and has proven difficult to predict given the parameters considered. In an effort to expand on Ana Urbina's work a larger breadth of taxa were considered and counted. Thirty-four taxa were plentiful enough to report on and plot on graphs for the 2022 open water season on Lake Auburn, while over 50 taxa were documented. The limitations for adding abundant taxa to the taxa already considered is no longer the skill in identification, but the power of the microscope. So far no major trend has been discovered to predict future NTU's or of the timing of cyano bloom maximums.

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