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Assessment of Pelagic Food Webs in Mendums Pond, NH

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Abstract
This study focused on the relationship between plankton in Mendums Pond, NH. A grazing experiment was conducted to determine the effect of zooplankton on the phytoplankton population. The phytoplankton were largely composed of net plankton (75 %) and this fraction was dominated by cyanobacteria (84.5 %) even though this was a slightly acidic system. Data indicated that the mean body length of zooplankton increased with depth. The average body length of Daphnia ranged from 1.4 mm in the epilimnion to 1.9 mm in the hypolimnion. Copepods followed a similar trend increasing in average body length from 0.85 mm to 0.95 mm. The high numbers of cyanobacteria and copepods resulted in a 17.92 % day^-1 grazing rate indicating that almost 18 % of the total lake water was filtered every day by the zooplankton. This also suggests that the phytoplankton are reproducing at a higher rate than they are being consumed by grazers. This may raise concerns about the future diversity of the food web as cyanobacteria reproduce and become more dominant in this system.

Introduction
The effects of zooplankton grazing on both phytoplankton and cyanobacteria are poorly understood despite a vast amount of information on the organisms themselves (Lampert and Taylor 1985). Grazing by zooplankton affects the dynamics of a phytoplankton community ranging from the density to the composition of that population. Phytoplankton such as cyanobacteria may contain toxins that deter predation and can even have adverse effects on humans. The interaction between zooplankton and phytoplankton along with nutrient input can greatly affect the trophic status of a lake.

The trophic status of a lake is indicative of primary productivity and thus dependent on the concentration of inorganic nutrients, such as phosphate (Dodson 2005). Lakes with high rates of primary productivity are enriched with key nutrients such as nitrogen and phosphorus, which could inadvertently initiate algal blooms (Peter, 2004). A strict increase of phosphorus into a low-nutrient lake dramatically increases the relative abundance of phytoplankton (Schindler 1974). Eutrophication is a slow, natural process that involves an increase in nutrients, primary production, and algal biomass (Lampert and Sommer 1997). Minimal impact from human development can help prevent eutrophication. Oligotrophic lakes have small amounts of nutrients, low primary production, well oxygenated clear water, and low algal production. Low productivity prevents accumulation of organic sediment on the bottom of oligotrophic lakes, resulting in high levels of oxygen present throughout the water column. Mesotrophic lakes have an intermediate level of productivity. Organic sediment accumulates and oxygen levels decrease deeper in the water column, but these changes do not drastically affect plankton (Dodson 2005).

Although nutrient concentrations influence the phytoplankton communities (Vanni 1987), zooplankton grazing can also have influential affects on phytoplankton relative abundance and community composition (Porter 1973, 1977, Gliwicz 1977, Lynch & Shapiro 1981, Vanni et al. 1987). Zooplankton grazers are typically larger, >375 µm, than their prey, <50 µm, and many ingest whole cells by pushing water through filter baskets (Dodson 2005). Predation can be avoided through defenses such as extracellular spines, used by many species of Daphnia, or secreting toxins, used by many cyanobacteria, as well as being too large or small to interest predators (Kirk and Gilbert 1992). Defenses to predation are important for the survival of individuals within a population and may
contribute to lowered grazing rates, which may result in a less diverse food web or eutrophication. The objective of this study was to analyze the food web and determine the interaction between organisms in Mendums Pond, NH.

Methods

Study Site – Mendums Pond is located at 43°10′24″N 71°3′54″W in Barrington, NH. It has a surface area of 102.39 hectares, a mean depth of 6.4 m and a maximum depth of 15.9 m. Sampling of this oligo-mesotrophic lake was performed on September 25, 2008 between 1500 and 1700 h.

Grazing – A mesocosm grazing experiment was conducted to determine the rate at which water was filtered by zooplankton. The zooplankton were concentrated into a closed environment with a known volume of filtered lake water and chlorophyll measurements were taken prior to adding zooplankton and after a known time interval. Water samples were taken at a depth of 1.9 m in triplicate using an integrated tube sampler (6 cm in diameter and 1.9 m long) and were then filtered through a 375 µm mesh to remove the large zooplankton. A control container was then filled with 3 L of filtered water. A zooplankton tow was taken with a 375 µm net in order to gather large zooplankton to enrich the water sample. The zooplankton were mixed into the remaining water which was separated into 3 L containers. Multi-parameter (MP) readings for chlorophyll and phycocyanin were taken for each sample using a Yellow Springs (YSI) Sonde 6600M multi-parameter probe. A fluorescence measurement was also taken on subsamples from each container using an AquafluorTM Handheld 2-Channel Fluorometer, Turner Designs, testing chlorophyll A and phycocyanin. One sample was then filtered through a 50 µm mesh and MP and fluorometer readings were taken. The containers were then submerged for approximately 1 h before they were brought to the surface where further readings were taken. The water in the containers was then filtered through both a pre-weighed 235 µm funnel mesh and a 50 µm ring mesh. The 235 µm mesh was placed in polycons and frozen for calculating the zooplankton biomass and the 50 µm mesh was used to filter out the large phytoplankton. After 50 µm filtration MP and fluorometer readings were taken for each replicate.

Other Field Methods – The MP was lowered at a rate of 0.5 m min⁻¹ to take vertical profiles of dissolved oxygen, chlorophyll a, phycocyanin, temperature, specific conductance, turbidity, oxidation-reduction potential, pH, and depth at the deepest part of the lake (~13 m) which was determined with a hand-held sonar. The data was recorded on an YSI 650 datalogger at 3 s intervals.

The light intensity profile of the lake was measured using a LI-92SA Underwater Quantum Sensor (LI-COR). The data were recorded, in triplicate, on a LI-1400 (LI-COR) data logger starting at 0.1 m and continuing at 0.5 m intervals to approximately 8 m. Secchi disk (20 cm black and white) transparency depths were measured in triplicate on the sunny side of the boat, with the aid of a view-scope.

Discrete plankton samples were taken using an 80 µm closing net at 2 m increments. The depths sampled were 0-2, 2-4, 4-6, 6-8, 8-10 and 10-12 m. Zooplankton and phytoplankton biomass were also estimated using samples taken from 0-12 m with a 50 µm net. The samples were taken in triplicate and placed in 2 L separatory funnels, which were then covered in order to separate the zooplankton from the phytoplankton. The zooplankton migrated to the bottom of the funnel where there was light. After 20 min the zooplankton were passed through a pre-weighed 50 µm funnel mesh and the remaining phytoplankton were then passed through another pre-weighed 50 µm funnel mesh. The meshes containing the plankton were then placed in polycons and frozen for later analysis. Live plankton samples were also collected with a 50 µm net and stored in a cooler for lab analysis.

Unfiltered whole lake water (WLW), sampled in the epilimnion (0-3 m), was gathered using an integrated tube and placed in 3 L containers (Reliance Freezepak) in triplicate. Subsamples were taken and stored in a cooler. Chlorophyll and phycocyanin were measured from the subsamples.
using the hand-held fluorometer. Volumes of 100 mL of water from each triplicate sample were filtered through a 2 µm or 0.45 µm membrane filter. The filters were placed in desiccators for drying and chlorophyll analysis then the filtrate was used to determine chlorophyll and phycocyanin in the soluble fraction. Whole lake water was collected for analysis of total phosphorous.

Lab Methods – Live plankton were identified and images were taken using a compound light microscope from each 2 m subsample using a Sony Handicam (10.2 megapixel HD) at a resolution of 0.01 mm. Data recorded on the YSI 6600M were downloaded from the YSI 650 and LI-COR data loggers and saved in Excel. The data were then graphed and statistically analyzed using SigmaPlot 11.0.

One-way ANOVA’s were used to analyze how average crustacean body length varied with depth. A t-test was performed to determine if there was a significant difference between the experimental clearance rate and the predicted rate from Lampert’s Equation. Additionally, regressions were used to determine the relationship between phycocyanin and chlorophyll, the relationship between phycocyanin or chlorophyll and turbidity, and chlorophyll and dissolved oxygen.

The clearance rate for the grazing experiment was calculated using the fluorescence data gathered with the MP. Using the formula, Clearance = ((Ln(Efinal/Einitial)-(Ln(Cfinal/Cinitial))*Volume/dry weight*time(hrs))) (Peters, 1971). Where C is the control and E is the experimental trial. The individual clearance rates from the replicate containers were used to calculate the grazing rate in % day⁻¹. First, (Ln(Einitial)-Ln(Efinal))/time(hrs) was calculated for the control and the three replicates. Then, Volume*(E-C)/dry weight was used. This number is then placed in the formula: (n*24hrs*calculated biomass)*1L/1000mL*100. This stepwise process converts mL mg⁻¹ hr⁻¹ to % per day.

Chlorophyll was extracted from the 2 µm and 0.45 µm filters using the acetone chlorophyll extraction method using 90% alkalized acetone and following the procedure outlined by Lind (Lind 1974). WLW samples were also analyzed for total phosphorous content.

Results

The temperature of Mendums Pond ranged from 19°C to 6.2°C with a thermocline between 4.8 m and 8.1 m. The epilimnion, between 0 m and 4.8 m, has relatively high levels of phycocyanin and chlorophyll, 21000 - 8000 Microcystis (Mic) cells L⁻¹ and 6.1 - 3.0 µg Chl a L⁻¹ respectively (Fig. 1).

Both phycocyanin and chlorophyll decrease in the metalimnion (4.8 m -8.1 m), from 4.9 to 0.8 µg L⁻¹ for chlorophyll and from 10700 to 2300 Mic cells L⁻¹ for phycocyanin (Fig. 1). In the hypolimnion, the phycocyanin and chlorophyll levels rose slightly but remained in a low range, 4.4 to 0.4 µg L⁻¹ and 22,700 to 4,100 Mic cells L⁻¹ respectively (Fig.1). The relationship between the chlorophyll and phycocyanin found in Mendums Pond was described by the equation PC conc. = 657.328 + (644.528 * chlorophyll). The adjusted R² was 0.0841 and the p-value<0.001 (Fig. 2).
The dissolved oxygen in Mendum’s Pond decreased from approximately 9 mg L\(^{-1}\) (100% saturation) at the surface to approximately 2 mg L\(^{-1}\) (15% saturation) at 13 m. The chlorophyll and phycocyanin concentrations follow the oxygen concentration closely at nearly all depths including the negative heterograde, an area of high oxygen surrounded by two areas of low oxygen (Fig. 1). The only exceptions were the increase in chlorophyll from 0 – 2 m and the decrease in phycocyanin at approximately 1 m (Fig. 1).

Turbidity maintained relatively consistent readings through the metalimnion averaging 2.77 NTU, but rapidly increased at 0.75 m to 9.4 NTU and at approximately 1.45 m to 5.1 NTU. There is a statistically significant correlation \((p<0.001)\) between turbidity and both chlorophyll \((R^2=0.09)\) and phycocyanin \((R^2=0.36)\). Turbidity increased steadily from 10.75 m to 12.75 m reaching 25 NTU (Fig. 3). Corrected oxidation-reduction potential \((E_7)\) averaged 132.5 mV until it sharply dropped at approximately 10.75 m. The pH was acidic throughout the water column, with 5.7 at the lake surface and decreased to 4.7 at the sediment surface, with the largest change occurring within the thermocline.

The photosynthetic compensation depth, arbitrarily defined here as the depths between 1% and 10% of the surface light intensity, was between 4.38 m and 2.19 m (Fig. 4). The average secchi disk depth was 2.78 m which corresponds to a decrease in chlorophyll (Fig. 4).
Phytoplankton – Cyanobacteria, consisting primarily of *Anabaena* and *Aphanocapsa*, dominated the water column with a relative abundance in the net phytoplankton of 59.1% (Fig. 5). Large quantities of *Kirchaeriella* and *Sphaerocystis*, of the class green algae or *Chlorophyta* were also prevalent throughout the water column (33.1%). Discrete samples within the epilimnion (0-4 m) revealed a high abundance of cyanobacteria (84.5%) followed by green algae (11.6%).

Cyanobacteria was the most abundant net phytoplankton between 0-2 m (1429 cyanobacteria mL⁻¹; Fig. 6), consisting solely of *Anabaena* (496 mL⁻¹) and *Aphanocapsa* (487 mL⁻¹). Green algae, composed mostly of *Kirchaeriella*, was also abundant (114 mL⁻¹) between the surface and 2 m. Green algae was dominant between 4-6 m and 8-10 m (474 mL⁻¹ & 694 mL⁻¹ respectively).

The percent composition of phytoplankton size fractions was 75% net plankton (>50 µm), 11.4% nanoplanckton, (2-50 µm), and 13.6% picoplankton (0.2-2 µm, Fig. 7).

![Fig. 5: Relative abundance of net phytoplankton throughout the entire water column. Mendums Pond was dominated mostly by cyanobacteria (59.1%) and green algae (33.1%).](image)

![Fig. 6: Vertical distribution of the three dominant groups of phytoplankton in Mendums Pond. The cyanobacteria are dominant numerically at all depths except 8-10 m, where green algae were the dominant group. The density of cryptophytes is greatest at 4-6 m.](image)

![Fig. 7: Size fractionation indicated netplankton as the dominant size (75%), followed by picoplankton (13.6%), and nanoplanckton (11.4%). This data is based on chlorophyll a estimates using fluorescence.](image)
Zooplankton - The densities of the dominant types of zooplankton ranged from 0.4 L\(^{-1}\) for cladocerans to 36.6 L\(^{-1}\) for rotifers (Fig. 8). Rotifers are most abundant throughout the majority of the water column until the 8-10 m sample at which point the copepods are dominant (Fig. 8). Rotifers, primarily *Kellicottia* and *Keratella* made up approximately 60\% of the zooplankton while copepods composed approximately 37\%, and cladocerans approximately 2\%.

Subsamples of the zooplankton were taken to measure individuals and obtain an average size of zooplankton at each depth (Fig. 9).

![Graph of zooplankton densities in Mendums Pond](image.png)

**Fig. 8:** Densities of the dominant zooplankton found in Mendums Pond. Rotifers of the genus *Keratella* and *Kellicottia* numerically dominated the water column to 8 m at which point the Copepods became dominant.

The average body length of *Daphnia* ranged from 1.4 mm at 2-4 m to 1.9 mm at 10-12 m (Fig. 10) while the average body length of copepods ranged from 0.85 mm at 0-2 m to 0.95 mm at 4-6 m (Fig. 11). One specimen of an invertebrate predator, *Leptodora kindti*, was found at 8-10 m (Fig. 12). Its estimated body length was 20 mm and although *Leptodora* is a cladoceran its body is highly modified to make it an efficient predator (Fig. 12).

Grazing Experiment – Clearance rates of net phytoplankton averaged 33.53 mL mg\(^{-1}\) hr\(^{-1}\) between the three replicates. The average grazing rate was found to be 17.92 \% day\(^{-1}\).

Discussion

Freshwater food webs can be affected by many different factors. These include temperature, oxygen concentration, and turbidity among others.

One hypothesis for the negative heterograde is the presence of respiring organisms, such as bacteria whose presence would significantly increase the turbidity. However, there was no increase in turbidity at the negative heterograde, thus rejecting the hypothesis. Additionally, turbidity increases at 0 – 2 m (Fig. 3). These could be due to bacteria or silt from runoff but without further testing of the water at those depths we cannot be certain. Although common with heterogrades, the significant
correlation between chlorophyll with oxygen ($P<0.001$, $R^2=0.83$) indicated that phytoplankton were not dense, which otherwise would have created an algal plate below the thermocline (Dodson 2005).

Other possible hypotheses include: 1) oxidizable material that is produced decomposes while it sinks, thus when it encounters the dense metalimnetic water, it slows down and demands a greater abundance of oxygen, 2) respiratory demands from large concentrations of zooplankton, and 3) where a gradual bottom slope coincides with the metalimnion and initiates horizontal mixing, which can spread the reduction laterally (Scrivener & Brownlee 1989).

Chlorophyll and phycocyanin have a significant correlation ($P < 0.001$), although nearly 92% of the variation was not accounted for by the given equation ($R^2 = 0.084$, Fig. 2). The majority of the points for each particular layer fall into tight groups (Fig. 2) which indicates that the Chlorophyll:Phycocyanin ratio changes with each limnetic layer in turn representing a change in the composition of the phytoplankton. The epilimnion was mostly composed of cyanobacteria, while the metalimnion contained both *Chlorophyta* and *Cryptophyceae* and the hypolimnion was dominated by cyanobacteria and *Chlorophyta* (Fig. 6).

There were relatively low levels of chlorophyll and phycocyanin in the hypolimnion and any phytoplankton or cyanobacteria living here require very low light levels. Most phytoplankton grow best at 10% of the maximum surface light levels (Dodson 2005), which was consistent with chlorophyll being most abundant at this depth (Fig. 3).

The phytoplankton were dominated by cyanobacteria, *Chlorophyta*, and *Cryptophyceae* (Fig. 6). While other taxa were apparent they did not make up any significant portion of the total phytoplankton (1.8%). The net cyanobacteria were highly dominant through nearly the entire lake, which indicates that it may be advantageous to contain toxins that repel potential predators (Lampert & Sommer 1997, Kirk & Gilbert 1992). The domination of cyanobacteria throughout the water column suggests lower grazing rates (% day$^{-1}$) due to the inedibility of cyanobacteria, which make up 84% of the netplankton. Although some of these phytoplankton may still be ingested by zooplankton, some have a thick cell wall (*Sphaerocystis*) that prevents digestion which leads to being egested alive. This allows the phytoplankton with thick cell walls to dominate in field experiments with high zooplankton densities (Porter 1977). The dominant green alga between 4-12 m was *Sphaerocystis*, which may also explain a lower grazing rate. While passing through the crustacean’s gut, the algae cells may take up nutrients within the intestine, thus enhancing their growth and domination (Cole 1994).

The percent composition of the size fractions of phytoplankton indicates that the majority of the phytoplankton are greater than 50 µm in length (Fig. 7) forcing zooplankton to consume larger than average food particles (Dodson 2005 and Wilson 1973). At 8-10 m green algae was dominant rather than cyanobacteria (Fig. 6). At this same depth the number of copepods also increased dramatically (Fig. 8). This may be indicative of a more easily utilized food source in the green algae.

The zooplankton data indicated that rotifers had the highest density followed by copepods, and cladocerans in descending order (Fig. 8). This relationship suggests a tendency towards smaller zooplankton which may indicate either a selective pressure of fish predation or evolution to avoid...
such predators (Mills and Green 1987). The copepods had a higher density than the cladocerans indicating an advantage in speed for predator avoidance. The larger cladocerans (1.9 mm) tend to be found in deeper waters, this is consistent with the findings of Murby (2006), whereas the largest copepods (0.95 mm) were found at depths between 2 m and 6 m (Fig. 10 and 11). Zooplankton have a direct relationship to fish production, thus zooplankton size can determine the structure of fish population within a lake (Mills & Green 1987, Mills & Schiavone 1982). According to the study performed by Mills & Green (1987), the size of the zooplankton increased with the mean length of planktivorous fish (yellow perch). Zooplankton can be indirectly affected by pressure of piscivores when they prey heavily on planktivorous fish (Mills & Green 1987). Mean zooplankton body size has a strong correlation between chlorophyll a and total phosphorus compared to animal biomass, which is significantly uncorrelated (Currie et al. 1999). The large copepods may be relatively hard for planktivorous fish to catch because of their speed. This would allow them to remain higher in the water column during the day than large cladocerans which are relatively slow.

If predatory fish control prey populations then they will indirectly affect the zooplankton community (Mills and Green 1987). Lakes with low predator to prey ratios (<0.2) will have many small panfish whereas those that have high ratios (>0.2) or predators to prey will have large panfish and large zooplankton (Mills and Green 1987). Mendum Pond had a high ration of predators to panfish (0.48) indicating that the panfish population is controlled by predators and that there should be a relatively small population of large panfish. The average length of zooplankton in Mendum Pond (0.95 mm) also supports this conclusion.

Fig. 11: Average body length of calanoid copepods from all sampled depths. Average body lengths ranged from 0.85 mm to 0.95 mm.

Fig. 12: morphology of *Leptodora kindti*. The relative size of *Leptodora* to copepods found at the same depth is indicated in (A). (B) *Leptodora* under a dark scale, allowing an excellent visual of the organism’s body without interference from other objects. Image (C) Image of one of the swimming appendages. The setae are easily visible and are used to help propel the organism through the water. (D) The multifaceted eye. This allows vision in all directions because the body of *Leptodora* is transparent.

Similar to the large *Daphnia* that seek refuge from predators deep in the lake (Fig. 10) some invertebrate predators, such as *Leptodora*, can also be found in deep waters during the day before moving shallower to feed on the zooplankton (Herzig 1990). *Leptodora* prefers food items larger than 500 µm which it can detect by using the setae on its thoracic limbs as mechanoreceptors (Fig. 12). It then grasps the prey and moves it into the head area which in conjunction with the thorax and other limbs forms an open basket preventing the escape of prey (Herzig 1990). In large densities these predators could significantly affect the
population of prey items such as other cladocerans and, rarely, copepods (Herzig 1990). This is interesting because it adds a unique level into the food web. In a study performed by Costa and Cummins (1972) the gut contents of several common species of panfish were analyzed. They determined that only the gut contents of Black Crappie contained the remains of *Leptodora* indicating that most planktivorous fish do not prey on *Leptodora*. However, further analysis of the fish population is necessary to determine how *Leptodora* fits into the food web of Mendums Pond.

The food web of Mendums Pond indicates a flow of energy from the phytoplankton through several trophic levels before reaching the piscivores (Fig. 13). The phytoplankton are consumed by zooplankton represented by the cladocerans and copepods. These organisms are then subject to predation by both invertebrate predators and planktivorous fish. These fish are then preyed upon by the dominant predators of the system (Fig. 13).

Zooplankton grazing exceeds the rate at which phytoplankton can grow when it reaches 80 % day$^{-1}$ (Reynolds 1984, Lampert 1988). This sets a standard for the effects of grazing on phytoplankton growth when calculating the grazing rates. The experimental grazing rates averaged 17.92 % day$^{-1}$ indicating a net growth of phytoplankton compared to a predicted value of 39.84 % day$^{-1}$ using Lampert’s equation. A two-way t-test analysis indicated that these two grazing rates (% day$^{-1}$) were not statistically significantly different ($P = 0.319$). This suggests that 17.92 % of the lake water is filtered every day by the zooplankton present in the lake. Although a relatively low number this may be explained by two different hypotheses 1) the dominance of copepods and rotifers which have a much lower grazing effect than cladocerans and 2) the grazing experiment was conducted during the afternoon meaning there was a relatively low grazing rate as photosynthesis takes place during the day and grazing occurs at night (Haney & Hall 1975, Lampert & Taylor 1985).

Through this study we were able to diagram the food web in Mendums Pond. The trophic level has important effects on the food webs. Mendums Pond had a high ratio of piscivores to panfish (0.48) and a high mean length of zooplankton (0.95 mm). this is typical of oligotrophic lakes which have many piscivores (Mills and Green 1987) resulting in significant pressure on the planktivores. Planktivorous fish are as a result present in low densities allowing for higher numbers of large zooplankton and decreasing concentrations of phytoplankton (Mills and Green 1987). These selective pressures also allow for the dominance of certain types of organisms that are more likely to avoid predation. The toxins produced by cyanobacteria and the speed of copepods allow these organisms to avoid predators and increase their densities within a system. This is evident in Mendums Pond because even though it is slightly acidic it is dominated by cyanobacteria which prefer alkaline environments.
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