

## **Effects of *Ceratophyllum demersum* on Phytoplankton Nitrogen Limitation in Twin Cities Metropolitan Area Lakes**

### **Introduction**

In aquatic ecosystem ecology, understanding the nature of resource limitation is a crucial step in being able to predict and manage changes to the environment. While it is an established paradigm that lakes are phosphorus-limited on the whole-ecosystem scale over long time periods, the biogeochemical mosaic hypothesis suggests that other factors, particularly nitrogen and trace metals, have important ecological effects on smaller spatial and temporal scales (Sturner 2008). The relationship between nutrient limitation and community composition involves physiological differences between species, and inter-species interactions. Allelopathy, or the secretion of chemical compounds by one organism that has beneficial or detrimental effects on another organism in the community, is one such interaction at play in aquatic communities.

The inhibitory allelopathic effect of the submerged macrophyte *Ceratophyllum demersum* on phytoplankton may maintain nitrogen limitation in lakes. As reviewed by Gross (2003), there is experimental evidence that *C. demersum* inhibits photosynthesis in phytoplankton through labile, lipophilic sulfur compounds and that *C. demersum* inhibits the growth of nitrogen-fixing cyanobacteria to a greater degree than other phytoplankton. In a Norwegian study of shallow lakes with varying phosphorus levels, lakes where *C. demersum* dominated had relatively low algal biomass and showed signs of nitrogen limitation (Mjelde and Faafeng 1997), indicating that allelopathic interactions between *Ceratophyllum* and phytoplankton may mediate nitrogen limitation. However, the effect of *C. demersum* on nutrient limitation has not been studied comprehensively and experimentally.

In the work for this paper, a set of nutrient enrichment experiments, including addition of *C. demersum* to algal cultures, was conducted on water collected from Como Lake in St. Paul, MN and Peltier Lake in Centerville, MN to test the effects of *C. demersum* on phytoplankton nitrogen limitation. Nutrient enrichment bioassays are a common method of assessing nutrient limitation in aquatic ecosystems (Hecky and Kilham 1988), and while bottle bioassays have limitations in their ability to explain whole ecosystems, they can give valuable insights into phytoplankton resource limitation on smaller scales. I hypothesize that cultures containing *C. demersum* or its extract will have lower algal biomass growth rates because *C. demersum*'s inhibitory allelochemicals biochemically inhibit algal growth. I also hypothesize that cultures incubated with *C. demersum* will show stronger signs of N limitation, i.e. a higher algal biomass growth response to N, because *C. demersum* competes for nitrogen and diminishes the ability of nitrogen-fixing algae to relieve N limitation.

## **Methods**

### **Site description and field collection**

Como Lake is located in St. Paul, MN next to a public park and golf course. It has a eutrophic nutrient status (Capitol Region Watershed District 2010), and a 2013 macrophyte survey found *C. demersum* at 20-30% of transect points over the course of the season (Capitol Region Watershed District 2013). Macrophytes were periodically harvested over the 2014 season for lake management purposes, though macrophyte biomass was still high near shore (personal observation). Peltier Lake is in Centerville, MN, a northern Twin Cities suburb. It is also fairly eutrophic, and despite infestation by both Eurasian watermilfoil and curly-leaf pondweed, the macrophyte community remains dominated by *C. demersum* in the shallow northern basin of the lake (Minnesota Pollution Control Agency 2009).

A total of four field sampling trips were carried out over July and August 2014, with two sampling events each for Como Lake and Peltier Lake. Field measurements included measures of lake depth and clarity, and depth profiles of temperature, dissolved oxygen, light, and conductivity. A water sample of approximately 5 liters was taken back to the lab to be used for lake chemistry analysis and as a medium for nutrient addition experiments, and roughly 70-100 grams wet mass of *C. demersum* was collected in a separate bottle for use in nutrient addition experiments.

### **Lab methods**

Four nutrient limitation assay experiments were carried out corresponding to the field sampling events. Each nutrient addition experiment consisted of a factorial addition of nitrogen, live sprigs of *C. demersum*, and *C. demersum* extract, with three replicate flasks for each treatment (i.e. a control flask and treatments of N, N+plant, N+extract, plant+extract, and N+plant+extract).

The flasks contained 100 mL of diluted lake water (9 parts 0.45  $\mu\text{m}$  filtered water: 1 part whole water) and were incubated for three days in temperature and day-length conditions approximating those of the lake at sampling time with a light level of 200  $\mu\text{mol photons/m}^2$ . Water was filtered on cellulose nitrate filters for chlorophyll *a* analysis before and after incubation. The extract was made using modified methods from Gross et al (2003); approximately 30 grams wet weight of *C. demersum* was ground in a mortar and pestle, filtered through an 80-micron mesh, and diluted in Nanopure<sup>TM</sup> Type I ultrafiltered water to a final volume of 300 mL. Each extract treatment flask received 10 mL of extract. The nitrogen treatment flasks received a nutrient enrichment of 70  $\mu\text{M}$  ammonium nitrate.

Comparison of the growth response of different treatments, approximated by the change in chlorophyll *a* levels before and after incubation, estimates the nutrient limitation of phytoplankton. Growth rates ( $\text{day}^{-1}$ ) were calculated using the expression

$$\left( \log \left( \frac{\text{final chl-a concentration (}\mu\text{g/L)}}{\text{initial chl-a concentration (}\mu\text{g/L)}} \right) \right) / 3$$

, which represents exponential algal growth over three days. Experimental data was analyzed using statistical software from the R project (R Core Team 2014).

## Results

In order to maximize the interpretive power of the relatively small data set in this study, the two sample dates and experiment runs for each lake were combined so that there was one analysis of variance (ANOVA) model for Como Lake and one model for Peltier Lake, with sample date and the experimental treatments as factors. Results from the ANOVA describe both direct effects of each factor and the interactions between factors; for example, the presence of nitrogen and *C. demersum* together in a culture may have an effect greater than the additive effects of nitrogen and *C. demersum* alone, i.e. a positive interaction.

Patterns in growth rate response to experimental treatments are variable, both between lakes and between dates. The only consistent effect on phytoplankton growth rate across lakes and dates is a positive effect of live *C. demersum*. The July experiment on Como Lake shows a positive effect of *C. demersum*, and a small positive effect of N addition alone that seems to disappear with addition of *C. demersum* and/or extract (Figure 1). Patterns in this data set are also obscured by considerable spread in the data points within experimental treatments (Figure 1). As for the August Como Lake experiment, Figure 1 suggests that N addition had a minimal effect on phytoplankton growth, while *C. demersum* and its extract had strong positive effects. The Como Lake ANOVA showed nitrogen, extract, *C. demersum* (“plant”), date, and the extract-

by-plant, extract-by-date, and extract-by-plant-by-date interaction terms to be significant at the 0.05 level (Table 1).

The Peltier Lake July data shows a clear, strong positive effect of *C. demersum*, and a positive effect of nitrogen, on chlorophyll-*a* growth rate (Figure 2). August experimental results for Peltier Lake showed a negative effect of extract on phytoplankton growth, a positive effect of live *C. demersum*, and no discernible effect of nitrogen (Figure 2). The ANOVA for Peltier Lake found N, extract, plant, date, and the N+plant, extract+plant, N+sample date, extract+ sample date, and plant+ sample date interaction terms to be significant at the .05 level (Table 3).

## **Discussion**

Nitrogen had a significant positive effect in the Como and Peltier experiments (Tables 1 and 2), suggesting that both lakes were N-limited to some degree. This effect was more evident in Peltier Lake than in Como Lake. Extract of *C. demersum* had a negative effect in some cases, most strikingly in Peltier Lake in August. The negative effect of extract there is consistent with the hypothesis that allelochemicals from *C. demersum* inhibit phytoplankton growth. Though these results are less clear in the treatments including live plants, they are certainly suggestive of allelopathy and warrant further exploration to tease apart variables.

As for higher-order interaction terms, the most important ones to consider are N+plant and N+extract, which correspond to *C. demersum*'s effect on N limitation. The N+plant interaction term was significant and negative in both the Como Lake and Peltier Lake models, which suggests that the presence of *C. demersum* inhibits phytoplankton growth even when there is adequate nitrogen. This interaction is interesting in light of the fact that both N and *C. demersum* alone have positive effects on chl-*a* growth rate. The N+extract term was not significant, so it is inconclusive whether allelochemicals in the extract have any direct effect on

phytoplankton response to N addition. The interaction term data support the hypothesis that cultures incubated with *C. demersum* had lower phytoplankton growth rates in nutrient-enriched conditions than nutrient-enriched cultures incubated without *C. demersum*. However, it is difficult to differentiate allelopathic inhibitory effects from stimulated growth due to nutrient enrichment.

The sample date interaction terms are relevant to seasonal effects on N limitation and the relative growth of *C. demersum* and phytoplankton. The Peltier Lake data seems to show an overall pattern of negative interaction of N addition with sample date, which would mean that N limitation diminishes later in the season. This is consistent with experimental findings of community shifts to favor N fixation in response to strong seasonal N limitation (Schindler et al. 2008), which would lead to a decline in N limitation over the course of the season.

The strong positive effect of *C. demersum* on phytoplankton growth in the experiments was unexpected. Given that epiphytic algae have less sensitivity to *C. demersum*'s allelochemicals (Hilt and Gross 2008), it could be that the sprigs of *C. demersum* used in the experiment carried epiphytic algae on their leaves, and these additional algae contributed to the chlorophyll *a* levels in the water. Nutrient leakage from *C. demersum* has previously been cited as a confounding factor in laboratory experiments (Gulati et al. 2007), and it could have contributed to the positive effect of *C. demersum*. The strong positive effect of extract on phytoplankton growth in Como Lake in August may be due to nutrient content of the extract. If Como Lake is phosphorus-limited, the algae may be responding to available P in the extract. If it is the case that the inclusion of *C. demersum* in culture increased phytoplankton growth, then this finding warrants further study to better understand the physiological interaction between *C. demersum* and the algae in question.

It is possible that the different patterns observed in growth responses to experimental treatments between the two lakes were a result of differences in background lake chemistry or phosphorus limitation, which was not assessed in this set of experiments. It may be helpful to revisit these results later with more chemistry data and other experimental results from Como Lake and Peltier Lake to provide more clarity. It is also difficult to separate the allelopathic effects of the extract from other compounds possibly present in the extract (e.g. organic N and P) without resorting to higher-level chromatography techniques; the explanatory power of my experiments was constrained by the amount of time and resources available for me to conduct research.

### **Conclusion**

The course of experiments provided variable evidence for allelopathic inhibition in the negative effect of *C. demersum* extract on phytoplankton biomass growth in Peltier Lake, but the influence of *C. demersum* on N limitation was even less clear. The observed results could be due to fundamental differences in nutrient chemistry and/or community composition between Como Lake and Peltier Lake. Further study should provide insight into the nutrient chemistry of the two lakes, the role of phosphorus in these inter-species interactions, and other community-level effects on nitrogen and phosphorus cycling and availability.

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Figures and Tables

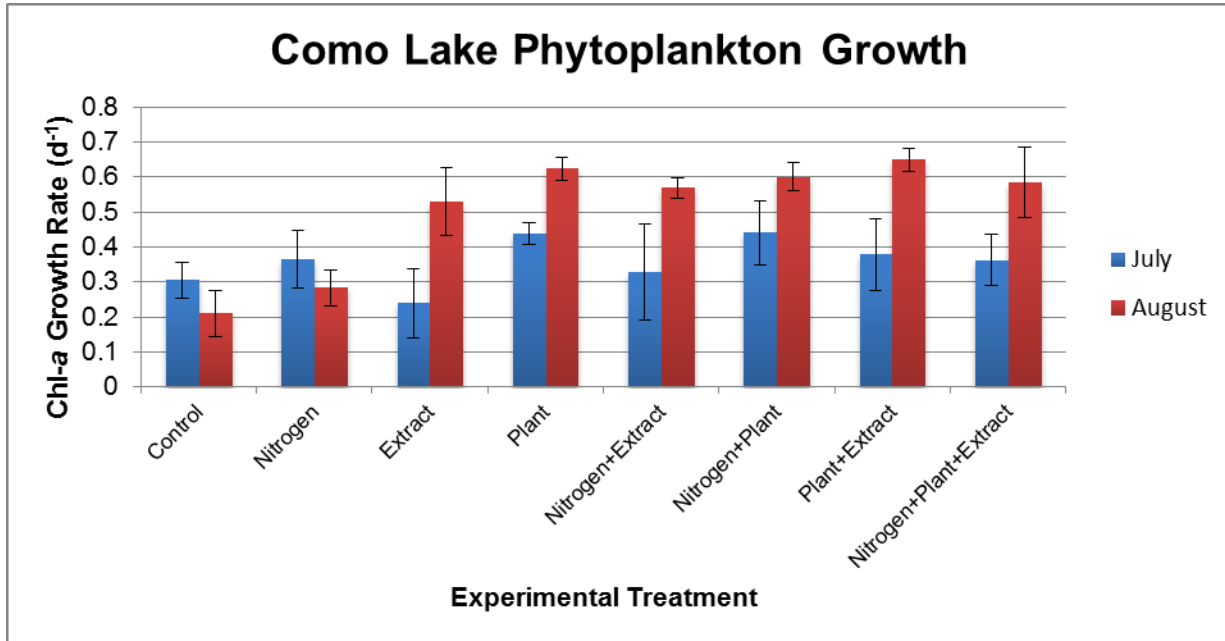


Figure 1. Mean chlorophyll *a* growth rates (day<sup>-1</sup>) in response to factorial addition of nitrogen, *Ceratophyllum demersum* (“plant”), and extract of *C. demersum* (“extract”) to water taken from Como Lake. Blue bars correspond to the July sampling date and red bars are for the August sampling date. Error bars indicate standard deviation.

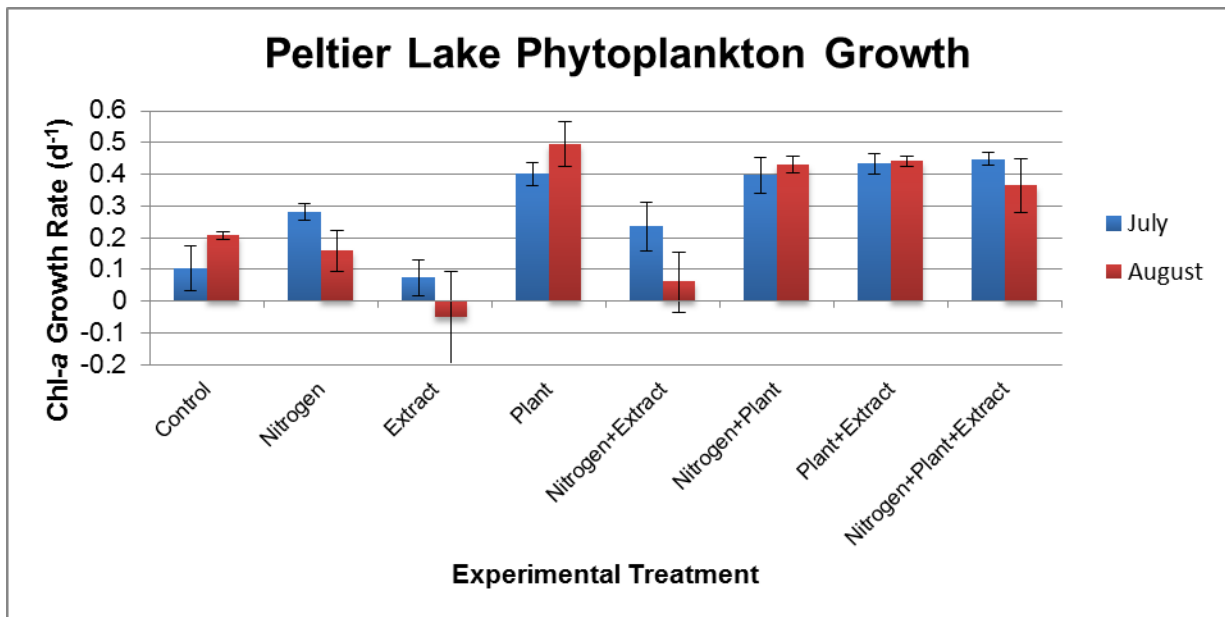


Figure 2. Mean chlorophyll *a* growth rates (day<sup>-1</sup>) in response to factorial addition of nitrogen, *Ceratophyllum demersum* (“plant”), and extract of *C. demersum* (“extract”) to water taken from Peltier Lake. Blue bars correspond to the July sampling date and red bars are for the August sampling date. Error bars indicate standard deviation.

Factor	df	Mean Square	F-statistic	p value
Nitrogen	1	0.0260	4.147	0.045387*
Extract	1	0.0988	15.739	0.000170*
Plant	1	0.5171	82.361	1.51e-13*
Sample date	1	0.3965	63.154	1.93e-11*
Nitrogen + Extract	1	0.0082	1.309	0.256382
Nitrogen + Plant	1	0.0236	3.764	0.056281
Extract + Plant	1	0.1169	18.613	5.01e-05*
Nitrogen + Sample date	1	0.0063	1.008	0.318818
Extract + Sample date	1	0.2904	46.256	2/57e-09*
Plant + Sample date	1	0.0689	10.971	0.001449*
Nitrogen + Extract + Plant	1	0.0038	0.609	0.437825
Nitrogen + Extract + Sample date	1	0.0069	1.105	0.296656
Nitrogen + Plant + Sample date	1	0.0004	0.066	0.798194
Extract + Plant + Sample date	1	0.0985	15.685	0.000174*
Nitrogen + Extract + Plant + Sample date	1	0.0005	0.081	0.776587
Residuals	72	0.0063		

Table 1: ANOVA results for the effects of sample date and factorial addition of nitrogen, *Ceratophyllum demersum* (“plant”), and extract of *C. demersum* (“extract”) on chlorophyll *a* growth rate in Como Lake. Asterisks indicate  $p < 0.05$ .

Factor	df	Mean Square	F-statistic	p value
Nitrogen	1	0.0288	6.776	0.011034*
Extract	1	0.0797	18.731	4.37e-05*
Plant	1	2.0560	483.128	<2e-16*
Sample date	1	0.0226	5.304	0.023910*
Nitrogen + Extract	1	0.0066	1.559	0.215506
Nitrogen + Plant	1	0.1012	23.783	5.48e-06*
Extract + Plant	1	0.0615	14.443	0.000283*
Nitrogen + Sample date	1	0.0643	15.106	0.000210*
Extract + Sample date	1	0.0851	19.990	2.57e-05*
Plant + Sample date	1	0.0512	12.020	0.000855*
Nitrogen + Extract + Plant	1	0.0070	1.655	0.202050
Nitrogen + Extract + Sample date	1	0.0076	1.790	0.184804
Nitrogen + Plant + Sample date	1	0.0059	1.393	0.241373
Extract + Plant + Sample date	1	0.0024	0.569	0.453033
Nitrogen + Extract + Plant + Sample date	1	0.0161	3.773	0.055643
Residuals	79	0.0043		

Table 2. ANOVA results for the effects of sample date and factorial addition of nitrogen, *Ceratophyllum demersum* (“plant”), and extract of *C. demersum* (“extract”) on chlorophyll *a* growth rate in Peltier Lake. Asterisks indicate  $p < 0.05$ .

**Appendix: Nutrient Addition Experiment Data**

Sample Date	Lake	Replicate Flask	Treatment	Replicate Filter	Growth Rate (day <sup>-1</sup> )
7/11/2014	Como	1	Control	1	0.2258213
7/11/2014	Como	1	Control	2	0.2732234
7/11/2014	Como	2	Control	1	0.3235323
7/11/2014	Como	2	Control	2	0.2963372
7/11/2014	Como	3	Control	1	0.3726236
7/11/2014	Como	3	Control	2	0.333373
7/11/2014	Como	1	N	1	0.2410654
7/11/2014	Como	1	N	2	0.3950754
7/11/2014	Como	2	N	1	0.4683236
7/11/2014	Como	2	N	2	0.4271638
7/11/2014	Como	3	N	1	0.3265987
7/11/2014	Como	3	N	2	0.3239743
7/11/2014	Como	1	Plant	1	0.4626783
7/11/2014	Como	1	Plant	2	0.4260795
7/11/2014	Como	2	Plant	1	0.4247636
7/11/2014	Como	2	Plant	2	0.3953453
7/11/2014	Como	3	Plant	1	0.4393993
7/11/2014	Como	3	Plant	2	0.4788426
7/11/2014	Como	1	E	1	0.0947299
7/11/2014	Como	1	E	2	0.2459429
7/11/2014	Como	2	E	1	0.1739049
7/11/2014	Como	2	E	2	0.2316386
7/11/2014	Como	3	E	1	0.3605723
7/11/2014	Como	3	E	2	0.3276088
7/11/2014	Como	1	N+Plant	1	0.3507049
7/11/2014	Como	2	N+Plant	1	0.5472998
7/11/2014	Como	2	N+Plant	2	0.5301897
7/11/2014	Como	3	N+Plant	1	0.3729387
7/11/2014	Como	3	N+Plant	2	0.4021903
7/11/2014	Como	1	N+E	1	0.076414
7/11/2014	Como	1	N+E	2	0.2637222
7/11/2014	Como	2	N+E	1	0.3853971
7/11/2014	Como	2	N+E	2	0.4250578
7/11/2014	Como	3	N+E	1	0.3842373
7/11/2014	Como	3	N+E	2	0.4380685
7/11/2014	Como	1	Plant+E	1	0.2954469
7/11/2014	Como	1	Plant+E	2	0.3367818
7/11/2014	Como	2	Plant+E	1	0.2568702
7/11/2014	Como	2	Plant+E	2	0.3807014
7/11/2014	Como	3	Plant+E	1	0.4992666
7/11/2014	Como	3	Plant+E	2	0.4984754
7/11/2014	Como	1	N+Plant+E	2	0.2740599
7/11/2014	Como	2	N+Plant+E	1	0.4530256
7/11/2014	Como	2	N+Plant+E	2	0.4049538
7/11/2014	Como	3	N+Plant+E	1	0.3739856
7/11/2014	Como	3	N+Plant+E	2	0.3032933

Sample Date	Lake	Replicate Flask	Treatment	Replicate Filter	Growth Rate (day <sup>-1</sup> )
7/18/2014	Peltier	1	Plant	1	0.385933
7/18/2014	Peltier	1	Plant	2	0.3678911
7/18/2014	Peltier	2	Plant	1	0.3795802
7/18/2014	Peltier	2	Plant	2	0.3818622
7/18/2014	Peltier	3	Plant	1	0.4643715
7/18/2014	Peltier	3	Plant	2	0.4269312
7/18/2014	Peltier	1	E	1	0.1756061
7/18/2014	Peltier	1	E	2	0.0846345
7/18/2014	Peltier	2	E	1	0.070472
7/18/2014	Peltier	2	E	2	0.0521983
7/18/2014	Peltier	3	E	1	0
7/18/2014	Peltier	3	E	2	0.0646333
7/18/2014	Peltier	1	N+Plant	1	0.4390879
7/18/2014	Peltier	1	N+Plant	2	0.3991647
7/18/2014	Peltier	2	N+Plant	1	0.3065277
7/18/2014	Peltier	2	N+Plant	2	0.4489681
7/18/2014	Peltier	3	N+Plant	1	0.3506196
7/18/2014	Peltier	3	N+Plant	2	0.4315395
7/18/2014	Peltier	1	N+E	1	0.321323
7/18/2014	Peltier	2	N+E	1	0.234766
7/18/2014	Peltier	2	N+E	2	0.2630792
7/18/2014	Peltier	3	N+E	1	0.2426417
7/18/2014	Peltier	3	N+E	2	0.1145335
7/18/2014	Peltier	1	Plant+E	1	0.3812632
7/18/2014	Peltier	1	Plant+E	2	0.423926
7/18/2014	Peltier	2	Plant+E	1	0.4800274
7/18/2014	Peltier	2	Plant+E	2	0.4430263
7/18/2014	Peltier	3	Plant+E	1	0.415414
7/18/2014	Peltier	3	Plant+E	2	0.4508419
7/18/2014	Peltier	1	N+Plant+E	1	0.460053
7/18/2014	Peltier	1	N+Plant+E	2	0.4307352
7/18/2014	Peltier	2	N+Plant+E	1	0.4656846
7/18/2014	Peltier	2	N+Plant+E	2	0.4662805
7/18/2014	Peltier	3	N+Plant+E	1	0.4185487
7/18/2014	Peltier	3	N+Plant+E	2	0.4396708
7/18/2014	Peltier	1	Control	1	0.1767092
7/18/2014	Peltier	1	Control	2	0.1733745
7/18/2014	Peltier	1	N	1	0.2389363
7/18/2014	Peltier	1	N	2	0.2867504
7/18/2014	Peltier	2	Control	1	0.0154408
7/18/2014	Peltier	2	Control	2	0.0628348
7/18/2014	Peltier	2	N	1	0.2967983
7/18/2014	Peltier	2	N	2	0.3170105
7/18/2014	Peltier	3	Control	1	0.1505885
7/18/2014	Peltier	3	Control	2	0.0441899
7/18/2014	Peltier	3	N	1	0.2804495
7/18/2014	Peltier	3	N	2	0.2665174

Sample Date	Lake	Replicate Flask	Treatment	Replicate Filter	Growth Rate (day <sup>-1</sup> )
8/8/2014	Como	2	Plant	1	0.5995567
8/8/2014	Como	2	Plant	2	0.6468285
8/8/2014	Como	1	E	1	0.5277627
8/8/2014	Como	1	E	2	0.5281601
8/8/2014	Como	2	E	1	0.5269646
8/8/2014	Como	2	E	2	0.3525472
8/8/2014	Como	3	E	1	0.6143162
8/8/2014	Como	3	E	2	0.6249333
8/8/2014	Como	1	N+Plant	1	0.6554814
8/8/2014	Como	1	N+Plant	2	0.585395
8/8/2014	Como	2	N+Plant	1	0.5982584
8/8/2014	Como	2	N+Plant	2	0.634105
8/8/2014	Como	3	N+Plant	1	0.5478758
8/8/2014	Como	3	N+Plant	2	0.5788343
8/8/2014	Como	1	N+E	1	0.5821826
8/8/2014	Como	1	N+E	2	0.5739366
8/8/2014	Como	2	N+E	1	0.5937328
8/8/2014	Como	2	N+E	2	0.5257591
8/8/2014	Como	1	Plant+E	1	0.6402921
8/8/2014	Como	1	Plant+E	2	0.6804935
8/8/2014	Como	2	Plant+E	1	0.6982901
8/8/2014	Como	2	Plant+E	2	0.6186994
8/8/2014	Como	3	Plant+E	1	0.6282177
8/8/2014	Como	3	Plant+E	2	0.6230911
8/8/2014	Como	1	N+Plant+E	1	0.6459128
8/8/2014	Como	1	N+Plant+E	2	0.6669466
8/8/2014	Como	2	N+Plant+E	1	0.6337442
8/8/2014	Como	2	N+Plant+E	2	0.4059088
8/8/2014	Como	3	N+Plant+E	1	0.624458
8/8/2014	Como	3	N+Plant+E	2	0.5199969
8/8/2014	Como	1	Control	1	0.1832026
8/8/2014	Como	1	Control	2	0.2919092
8/8/2014	Como	1	N	1	0.3541727
8/8/2014	Como	1	N	2	0.3398456
8/8/2014	Como	2	Control	1	0.2414268
8/8/2014	Como	2	Control	2	0.1381664
8/8/2014	Como	2	N	1	0.2262795
8/8/2014	Como	2	N	2	0.2623679
8/8/2014	Como	3	Control	1	0.265649
8/8/2014	Como	3	Control	2	0.1401099
8/8/2014	Como	3	N	1	0.2619524
8/8/2014	Como	3	N	2	0.2533834

Sample Date	Lake	Replicate Flask	Treatment	Replicate Filter	Growth Rate (day <sup>-1</sup> )
8/15/2014	Peltier	1	Plant	1	0.5625937
8/15/2014	Peltier	1	Plant	2	0.5630394
8/15/2014	Peltier	2	Plant	1	0.5158459
8/15/2014	Peltier	2	Plant	2	0.5109873
8/15/2014	Peltier	3	Plant	1	0.3991617
8/15/2014	Peltier	3	Plant	2	0.4166148
8/15/2014	Peltier	1	E	1	0
8/15/2014	Peltier	1	E	2	0
8/15/2014	Peltier	2	E	1	0
8/15/2014	Peltier	2	E	2	-0.2657194
8/15/2014	Peltier	3	E	1	0.1394283
8/15/2014	Peltier	3	E	2	-0.1741045
8/15/2014	Peltier	1	N+Plant	1	0.4583189
8/15/2014	Peltier	1	N+Plant	2	0.4198664
8/15/2014	Peltier	2	N+Plant	1	0.4437336
8/15/2014	Peltier	2	N+Plant	2	0.4560654
8/15/2014	Peltier	3	N+Plant	1	0.3888839
8/15/2014	Peltier	3	N+Plant	2	0.4161552
8/15/2014	Peltier	1	N+E	1	0
8/15/2014	Peltier	1	N+E	2	0
8/15/2014	Peltier	2	N+E	1	0.1701678
8/15/2014	Peltier	2	N+E	2	0.1944489
8/15/2014	Peltier	3	N+E	1	0
8/15/2014	Peltier	3	N+E	2	0
8/15/2014	Peltier	1	Plant+E	1	0.4666737
8/15/2014	Peltier	1	Plant+E	2	0.4245756
8/15/2014	Peltier	2	Plant+E	1	0.4369847
8/15/2014	Peltier	2	Plant+E	2	0.4475516
8/15/2014	Peltier	3	Plant+E	1	0.4195679
8/15/2014	Peltier	3	Plant+E	2	0.4505466
8/15/2014	Peltier	1	N+Plant+E	1	0.3989037
8/15/2014	Peltier	1	N+Plant+E	2	0.4388987
8/15/2014	Peltier	2	N+Plant+E	1	0.4166157
8/15/2014	Peltier	2	N+Plant+E	2	0.4056578
8/15/2014	Peltier	3	N+Plant+E	1	0.3075237
8/15/2014	Peltier	3	N+Plant+E	2	0.2158498
8/15/2014	Peltier	1	Control	1	0.2214041
8/15/2014	Peltier	1	Control	2	0.1946262
8/15/2014	Peltier	1	N	1	0.2280249
8/15/2014	Peltier	1	N	2	0.2477248
8/15/2014	Peltier	2	Control	1	0.2155434
8/15/2014	Peltier	2	Control	2	0.2023882
8/15/2014	Peltier	2	N	1	0.1399462
8/15/2014	Peltier	2	N	2	0.1010296
8/15/2014	Peltier	3	Control	1	0.1970859
8/15/2014	Peltier	3	Control	2	0.2138466
8/15/2014	Peltier	3	N	1	0.1047034
8/15/2014	Peltier	3	N	2	0.1327923