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Phytoplankton-Nutrient Interactions in Lake Apopka

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EXECUTIVE SUMMARY

Phytoplankton-Nutrient Interactions in Lake Apopka were studied under an agreement between the St. Johns River Water Management District and the University of Florida. The study was instituted because Lake Apopka contains high concentrations of total phosphorus and excessive algal (phytoplankton) growth has been identified as the prime symptom of eutrophication in this lake. High standing crops of phytoplankton are the result of excessive phosphorus loading and contribute to low light penetration in the water column. Studies on this shallow, hypereutrophic lake were conducted from December 1989 to November 1991. Results are presented in five Chapters.

Introduction (Chapter 1)

Chapter 1 presents the purpose and scope of the report including a literature review on previous environmental conditions and management activities. The literature review includes discussion of the dramatic environmental changes that occurred in 1947 when the lake changed from a clear-water, macrophyte-dominated system to a turbid, phytoplankton-dominated system.

Phytoplankton Production and Ambient Lake Conditions (Chapter 2)

Results of a sampling program conducted from February 1990 to July 1991 are presented. Data collected twice-monthly include photosynthetically active radiation (PAR), water column light extinction, Secchi disc transparency, water temperature, dissolved

oxygen, and chlorophyll a. This chapter also includes monthly measurements of phytoplankton productivity.

Except for water temperature and lake level, no distinct seasonal patterns were found in the parameters measured during the study. Seasonal patterns, if present, are obscured by large between-sampling differences in data collected twice monthly. We obtained evidence that phytoplankton dynamics in this shallow lake are controlled by short-term dynamics in environmental processes. Algae and other materials that have settled to the bottom may be resuspended by wind-generated turbulence and periodically comprise a large fraction of measured phytoplankton and total suspended solids.

The high standing crops of phytoplankton are a major factor causing high turbidity in the lake. Measurements of Secchi disc transparency showed that the limit of visibility in the water ranged from 6-14 inches. Large ranges in other variables were found including K_t or light extinction (3.1-12.8 m^{-1}), water temperature (17.0-32.5°C), chlorophyll (44-217 mg m^{-3}), maximum gross primary productivity (133-1149 mg C m^{-3} h^{-1}), and areal gross primary production (116-568 mg C m^{-2} h^{-1}).

Although twice monthly sampling is not adequate to characterize the short-term variation in phytoplankton dynamics in Lake Apopka, our results are valid to describe average conditions in the lake. These results provide an important data base on which management decisions can be based and from which the results of management decisions can be assessed.

Nutrient Enrichment Bioassays (Chapter 3)

Results of monthly bioassays for nitrogen (N) and phosphorus (P) nutrient limitation are presented. The experimental approach was based on measuring responses of the algal community to nutrient enrichments of N, P, or N+P compared to non-enriched controls. Experiments were run in the laboratory under controlled light and temperature conditions using the natural phytoplankton assemblages present in the lake at the time water was collected. Comparison of experiments conducted in Lake Apopka with those conducted in the laboratory showed no statistically significant difference in response.

We conducted 20 experiments at approximately monthly intervals from December 1989 to July 1991 to determine if N or P was the primary limiting nutrient or a secondary limiting nutrient. Results from these experiments indicated that N was the primary limiting nutrient in 19 of the 20 experiments and that P was the primary limiting nutrient only once. Secondary P limitation was found in only 5 of the 19 experiments in which responses to N+P were statistically greater than the response to N alone.

We conclude that N is the primary limiting nutrient because the water of Lake Apopka generally contains large supplies of phosphorus that can be utilized for phytoplankton growth when the water is enriched with N. We also conclude that N limitation in Lake Apopka is the result of excessive P loading to the lake and that P supplies must be reduced to reduce standing crops of algae.

Nutrient-Dilution Bioassays (Chapter 4)

Chapter 4 presents results of bioassays for nitrogen (N) and phosphorus (P) nutrient limitation that were combined with dilution experiments. These studies are called nutrient-dilution bioassays because the experiments utilized nutrient enrichment with N, P, and N+P in combination with dilution of lake water to determine the effect of reduced nutrient supplies on phytoplankton growth and production. Lake water was diluted from 5-95% in these experiments. Effects of nutrient reduction must be known to predict or assess the impact of reduced nutrient loading on phytoplankton production and other ecosystem processes.

Although complex interactions characterized these experiments, results provide insight on the relative roles of N and P in establishing and maintaining the present hypereutrophic conditions in Lake Apopka. Most of the phosphorus occurs in phytoplankton, and a large portion is stored in phytoplankton as polyphosphates. This finding is important because the stored polyphosphate in phytoplankton can be used for growth in the absence of phosphorus in the water. Phytoplankton in the lake, therefore, are generally N-limited because of phosphorus storage in phytoplankton cells.

Clear evidence for reduction of biomass with dilution was obtained only in N treatments because N enrichment allowed the utilization of stored phosphorus for phytoplankton growth and eventually caused phytoplankton to be P-limited.

In general for treatments other than N enrichment, the biomass yield increased with dilution. This effect was most dramatic with the combined treatment of N+P, but was also evident in the control

and P treatments. The unexpected increase in phytoplankton growth with dilution possibly can be attributed to one or more of the following factors: dilution increases the supply of nutrients for the diluted phytoplankton, zooplankton grazing on phytoplankton is relieved with dilution, artificial diluents like MIS may contain substances that enhance growth either directly or indirectly, allelopathic substances (growth-inhibiting compounds) may have been diluted, and light attenuation was reduced by dilution.

The cellular content of stored phosphorus in algae increased with dilution more than 3-fold. In the present marsh restoration project, increased P-storage capacity with dilution may aid in the removal of P from Lake Apopka by: (1) removing available P more swiftly in waters that have been diluted with effluent from the marsh, and (2) decreasing the internal P load by producing more P-rich particles to be deposited in the wetland. The relative effectiveness of this internal "P-pump", in concert with P removal by the wetland, requires additional experimentation in order to assess its role in the net export of P from Lake Apopka.

Indicators of Nutrient Status (Chapter 5)

Nutrient enrichment bioassays (NEB) appear to be the most reliable indicator of nutrient limitation to phytoplankton in Lake Apopka. In addition to NEB, physiological indicators of N and P limitation can be used to provide an independent validation. For indices to be used as predictive tools of phytoplankton nutrient status, they need to be calibrated in parallel with NEB for phyto-plankton assemblages of varying nutrient deficiency. The most

appropriate methods for this are HEP(hot-water extractable phosphorus)-storage or P-uptake for evaluation of P-limitation and N-uptake to corroborate N-limitation. All three of these assays are non-hazardous (avoid the use of harsh chemicals), simple to conduct (experimentation duration <24 h), and do not require any specialized equipment (e.g., radioisotopes) other than that used to measure basic water chemistry.

Routine chemical measurement of dissolved inorganic nitrogen in the water can be used to determine if phytoplankton are N-limited. The nutrient status of phytoplankton relative to N is not complicated by N storage in phytoplankton cells.

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Chapter 1

INTRODUCTION

Lake Apopka is a large (surface area = 125 km^2), shallow (mean depth = 1.7 m) subtropical lake located in central Florida, approximately 25 km northwest of Orlando in Orange and Lake counties. The original surface area of the lake was 18,000 ha (180 km^2, 45,000 acres), the second largest in Florida (Schneider and Little 1973). However, draining approximately 6,000 ha of marginal marshlands along the northern shore and converting them to muck farms reduced the surface area to 12,500 ha in the 1940s. It is considered to be hypereutrophic because its waters contain high concentrations of total phosphorus. During our study we measured concentrations of total phosphorus as large as 200 $\mu g\ P\ L^{-1}$ which is comparable to the average total phosphorus concentration reported by Lowe et al. (1992). This high degree of nutrient enrichment results in the production of large standing crops of phytoplankton (which averaged 100 $\mu g\ L^{-1}$ during our study) and contributes to low light penetration in the water column.

Different approaches have been used to assess the importance of nitrogen and phosphorus supplies on the production of phytoplankton in freshwater lakes. A statistical approach has been used to predict phytoplankton biomass from water column concentrations of total phosphorus and total nitrogen (e.g., Jones and Bachmann 1976). The correlative relationship, however, offers limited information on underlying causal factors. Soluble nutrient concentrations also may not be a valid index of nutrient status of the phytoplankton because

cells can store phosphorus in excess of immediate growth requirements (e.g, Healey 1979; Lean and Pick 1981). In addition, the nutrient demand for phytoplankton production, especially in eutrophic lakes, can deplete soluble inorganic nutrients to limiting levels for phytoplankton growth. Excessive loading of phosphorus, for example, may induce nitrogen limitation by increasing the demand for this nutrient until supplies in the water column are depleted (Schelske 1984). Thus, identifying nutrients that limit phytoplankton growth in hypereutrophic waters may be complicated by persistent nutrient excesses (Paerl and Bowles 1987). Other approaches to study nutrient limitation of phytoplankton include bioassays using natural phytoplankton assemblages (Chapters 3 and 4), whole lake fertilization (e.g., Schindler and Fee 1974), or the use of physiological indicators of nutrient deficiency (Chapter 5).

Debate exists on which primary factors mediate phytoplankton dynamics in tropical and subtropical lakes (Kratzer and Brezonik 1981 1982; Osgood 1982; Canfield et al. 1989). Relationships between total phosphorus and total nitrogen and phytoplankton biomass across a suite of Florida lakes support the idea that phytoplankton biomass in these lakes is regulated by nutrients (Canfield 1983). However, additional work has questioned whether phytoplankton abundance is under strict nutrient control, given the high levels of biomass supported in some of these lakes (Canfield and Hoyer 1988; Canfield et al. 1989; Duarte et al. 1990; Agusti et al. 1991). Here we have used several approaches to assess the importance of nutrients in the production of large standing crops (biomass) of phytoplankton in Lake Apopka.

This report presents results obtained from an agreement between the St. Johns River Water Management District and the University of Florida to study "Phytoplankton-Nutrient Interactions in Lake Apopka." Studies under the agreement were instituted because excessive algal (phytoplankton) growth has been identified as the prime symptom of eutrophication in this lake. The general objectives were to estimate rates of phytoplankton production, to measure standing crops of phytoplankton in terms of chlorophyll concentration, to investigate factors that limit phytoplankton production, and to evaluate methods of assessing nutrient status of phytoplankton. Most of the data presented here were collected from December 1989 to July 1991.

ORGANIZATION OF THE REPORT

In presenting data obtained during the study, results are organized in four separate chapters (Chapters 2-5). This organization, includes a summary of all findings (Executive Summary).

In Chapter 2, results of a twice-monthly sampling program are presented. Data include measurements of photosynthetically active radiation (PAR) and calculations of water column light extinction, Secchi disc transparency, water temperature, dissolved oxygen, and chlorophyll a. This chapter also includes data on monthly measurements of phytoplankton production using light and dark bottle methodology and data on photosynthesis (phytoplankton production) and irradiance (P vs. I) relationships.

In Chapter 3, results of monthly natural assemblage bioassays for nitrogen (N) and phosphorus (P) nutrient limitation are presented.

This chapter includes an evaluation of the protocol used in the experiments.

In Chapter 4, results of additional natural assemblage bioassays for nitrogen (N) and phosphorus (P) nutrient limitation including dilution experiments are presented. In dilution experiments, raw lake water was diluted with distilled water, artificial lake water containing the major ionic components of Lake Apopka water, and Lake Apopka water filtered to remove suspended particulates and phytoplankton. These experiments were designed to evaluate the potential effect of reduced nutrient concentrations on phytoplankton growth and production.

In Chapter 5, an evaluation of the methodology and utility of using physiological indicators to assess the nutrient status of Lake Apopka phytoplankton is presented and discussed.

Conclusions are presented in each chapter. Possible applications of our findings on nutrient and light limitation and their interactions for the management and restoration of Lake Apopka are discussed in each chapter.

THE LAKE APOPKA ECOSYSTEM: HISTORICAL PERSPECTIVE

Lake Apopka was a premier bass fishing lake with exceptionally clear water in the 1940s (Clugston 1963), at which time the vegetation was dominated by a lush growth of submerged macrophytes (Dequine 1950). A dramatic change in water quality occurred after a hurricane caused massive plant mortality in 1947. Within a week after the hurricane the first plankton bloom was reported. Since then, decaying plant material has produced a lake

bottom characterized by loose unconsolidated peat, silt, and decaying organics (Schneider and Little 1969; Reddy and Graetz 1991). Dense beds of rooted aquatics were never reestablished, probably because they could not compete with planktonic algae in the nutrient-rich waters (Chesnut and Barman 1974), and the lake has changed from a highly regarded sport-fishing lake to a lake with few desirable sport fish. The historical perspective reported here has been adapted from the account by Schelske and Brezonik (1992).

Human influence on the lake was evident by 1920 when citrus groves were being planted in central Florida (Schneider and Little 1973). The well-drained southern shoreline was an excellent site for groves, but the marshland on the northern shore was not developed. In 1920 the town of Winter Garden constructed a sewerage system and two large septic tanks permitting wastes to enter the lake directly. Nutrients from municipal waste and runoff from the citrus groves seemed at first to be beneficial to a popular sport fishery. Lush growth of submerged macrophytes, such as *Vallisneria americana* and *Potamogeton illinoensis*, that covered the lake bottom (Dequine 1950) provided cover for young fish and tied up nutrients.

Several factors may have led to the abrupt shift to phyto-plankton dominance of the food web in 1947. A plan for draining and farming part of the marshland in 1942 called for construction of a dike along the north shore and draining about 6000 ha of fertile lake bottom to be used for muck farm agriculture (Schneider and Little 1973). Water pumped out of the farming areas may have added nutrients and contributed to siltation in the lake. Draining marshlands destroyed spawning grounds. Clugston (1963) stated that

a combination of external factors probably increased the fertility of the lake that led to the first algal bloom in 1947. A water hyacinth control program resulted in large amounts of decaying vegetation and increased nutrient supplies. The capacity and release of waste products of a citrus processing plant at Winter Garden increased considerably between 1946 and 1950. Muck farms at the north end of the lake were expanded greatly in the 1940s. Citrus groves located along the eastern and western shores may have contributed nutrients. A sewage treatment plant at Winter Garden was pumping effluent into the lake and increasing the supply of nutrients.

The game fish population comprised 35 percent of the species present and gizzard shad made up 20 percent of the total fish population by weight in 1947 when the plankton bloom was first noted (Clugston 1963). Sports fishing for largemouth bass, speckled perch, bluegill, and other panfish provided record size fish and a half million dollar annual income for 13 fishing resorts and camps. A thriving commercial fishery yielded more than 3 million kg (dressed weight) of catfish in one 8-month period. The planktivorous gizzard shad probably increased greatly in number in response to the persistent plankton bloom, but were small in size providing excellent forage for game species. By 1956-57, however, the game fish population dropped to 18 percent. Shad, which comprised most of the remaining 82 percent, are thought to have become too large and numerous to be cropped by game species. In an effort to alleviate the shad problem, the Game and Fresh Water Fish Commission treated the lake with rotenone in three successive years, 1957, 1958, and 1959. An estimated 9 million kg of gizzard shad were killed with the three

treatments. These fish were left in the lake to decompose and release nutrients.

An FWPCA study begun in 1968 revealed that 90% of the bottom was covered with unconsolidated bottom sediment (muck) averaging 1.5 m thick (Schneider and Little 1973). FWPCA made a crude nutrient budget and emphasized that restoration of the lake must include reduction of nutrient input. Although direct rainfall on the lake and high nutrient input from citrus grove runoff were important, the principal controls on inputs emphasized by FWPCA were point sources such as agricultural runoff pumped directly into the lake from muck farms and municipal and industrial wastes. In addition to control of external nutrient sources, several solutions for improving lake water quality included: dredging to remove nutrient-rich unconsolidated bottom sediments to increase lake depth and reduce internal nutrient recycling, using lake drawdown to expose and subsequently consolidate large areas of lake bottom by oxidation and compaction, adding an inert sealing material to stabilize bottom sediments, employing hydroponic farming to remove dissolved nutrients, and harvesting algae and fish to remove nutrients.

The governor of Florida assigned complete responsibility for a 1970 restoration of Lake Apopka to the Florida Air and Water Pollution Control Commission. This agency decided to proceed with the lake drawdown plan by allowing gravity drainage to lower the lake level 60 cm beginning December 1970. The effect of this lowering was to be evaluated and the lake would then be drained further by pumping to 25 percent of its original area. It was anticipated that two beneficial effects would result from the drawdown. First,

nutrient recycling would be reduced or eliminated from dried, compacted sediments. Second, suitable substrate for rooted aquatic vegetation would also be a result. This plan to lower the lake about seven feet below normal water level was not implemented, however, because the projected cost of the final pumping and sediment removal was $20 million and because of concern about environmental and economic impacts (Lowe et al. 1985). For example, reduction in lake volume minimizes freeze protection citrus growers receive from the large heat capacity of the lake.

In the 1970s additional studies were conducted on water quality problems and on restoration of Lake Apopka (Brezonik et al. 1978, Lowe et al. 1985). Studies of techniques that might be used to restore Lake Apopka also have continued. Biomanipulation of algal standing crops with gizzard shad may actually increase standing crops of undesirable algae (Crisman and Kennedy 1982).

Divergent views can be found concerning the restoration of hypereutrophic lakes in general and Lake Apopka in particular (see Schelske and Brezonik 1992). One viewpoint is illustrated by the need to reduce nutrient inputs to prevent accelerated eutrophication. Schneider and Little (1973) comment that the history of "Lake Apopka is not atypical" because other lakes in Florida and reservoirs all over the south are being subjected to similar attacks. They state the lake can be restored but only with great expense and difficult decisions, particularly whether a $10 million marginal muck-farming operation can expend money for nutrient removal. The other viewpoint is that it may not be possible to restore some lakes or, if it is, it may not be practical due to economic considerations.

At the present time, the St. Johns River Water Management District is beginning a pilot study on using marsh restoration to improve water quality in the lake (Lowe et al. 1989, 1992). Approximately 5,000 acres of muck farm land has been purchased and will be flooded to restore the wetland and use the wetland as a filter to remove nutrients. The hydrology of the wetland will be manipulated so highly nutrient-enriched water will flow from the lake into the wetland and then nutrient-depleted water from the wetland will be directed back to the lake. If successful, this project would result in both a restored wetland and a restored lake.

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Chapter 2

PHYTOPLANKTON COMMUNITY PRODUCTION AND RELATED VARIABLES, FEBRUARY 1990 TO JULY 1991

INTRODUCTION

The factors that regulate and control growth and production of phytoplankton populations in shallow lakes are not well known. Little work has been done on subtropical lakes, including Lake Apopka. Lake Apopka is interesting to study because it has changed from a highly productive sport fishery lake with a production base that was supported by submerged macrophytes and attached algae to a hypereutrophic lake with a production base dominated by phytoplankton (see Chapter 1). The work undertaken in this chapter was used to 1) describe ambient conditions in the lake including lake level, water temperature, light absorption, water transparency, and standing crop of phytoplankton measured as chlorophyll a, 2) measure volumetric rates of phytoplankton photosynthesis (primary productivity), 3) estimate phytoplankton primary production integrated over the water column, 4) relate phytoplankton primary productivity and photosynthetically active radiation (PAR), and 5) investigate sources and causes of short-term variability in ambient conditions and phytoplankton primary productivity. These data are needed to assess trophic conditions in the lake, to model phytoplankton production using light and chlorophyll data, and ultimately to be able to predict how nutrient reduction will affect phytoplankton production, phytoplankton standing crop, and light

penetration in the water column. These data combined with results presented in Chapters 3 and 4 can be used to make management decisions and to predict how much nutrient reduction will be required to achieve a given set of trophic state conditions.

Data presented in this chapter were collected on 33 dates extending from February 1990 to July 1991. Most of the data were obtained as part of the twice-monthly sampling program established in February 1990 (see Chapter 1). Some additional data were obtained that are very useful in understanding phytoplankton dynamics and production in Lake Apopka. Of particular interest are data collected on consecutive days from 27 February-3 March 1991 and from 24-26 July 1991. Data collected on consecutive dates at two different times show that characteristics of Lake Apopka can change dramatically in 24 hours. We believe that this type of variability is not uncommon in the lake and that it explains much of the variability that we found during 18 months of sampling. We believe that this type of short-term variability tends to obscure seasonal patterns of phytoplankton production. We conclude from data presented in the final section of this chapter (see Short-Term Variability) and from other information that episodic events play major roles in controlling short-term production dynamics of phytoplankton in Lake Apopka.

MATERIALS AND METHODS

A single station was chosen as the site for our study because our preliminary investigations indicated that spatial variability was not great on any given date. Other data also indicated that spatial

variability in the lake was much less than temporal variability (see Brezonik et al. 1978, Newman 1991). Our station was located approximately 500 m southwest from the boat launching ramp at Magnolia Park (Fig. 2.1). This station was approximately 1.0 m deep, but during the course of the study the depth varied 37 cm (1.2 ft) as the result of seasonal changes in lake level.

Physical and Chemical Characteristics

Physical and chemical characteristics were sampled routinely prior to collecting water for primary productivity experiments. Data were usually obtained during mid morning (1000-1100 h). Water temperature and dissolved oxygen were measured with a Yellow Springs Instruments Meter (Model 57) at 10-25 cm intervals in the water column. Secchi disc transparency was measured with a 20-cm disc (black and white quadrants).

A LiCor Quantum Meter and Data Logger were used to record data simultaneously for upwelling, downwelling, and incident photosynthetically active radiation (PAR) using separate cosine sensors for each measurement. Data for upwelling and downwelling light were obtained immediately sub-surface and at 10-cm intervals for depths below the surface. To minimize short-term variability, data were integrated and averaged for 60 seconds. Absorption (extinction) coefficients, K_t, of PAR were calculated at each depth relative to the surface using the following equation

$$K_t = \frac{\ln(I_0) - \ln(I_z)}{Z}$$

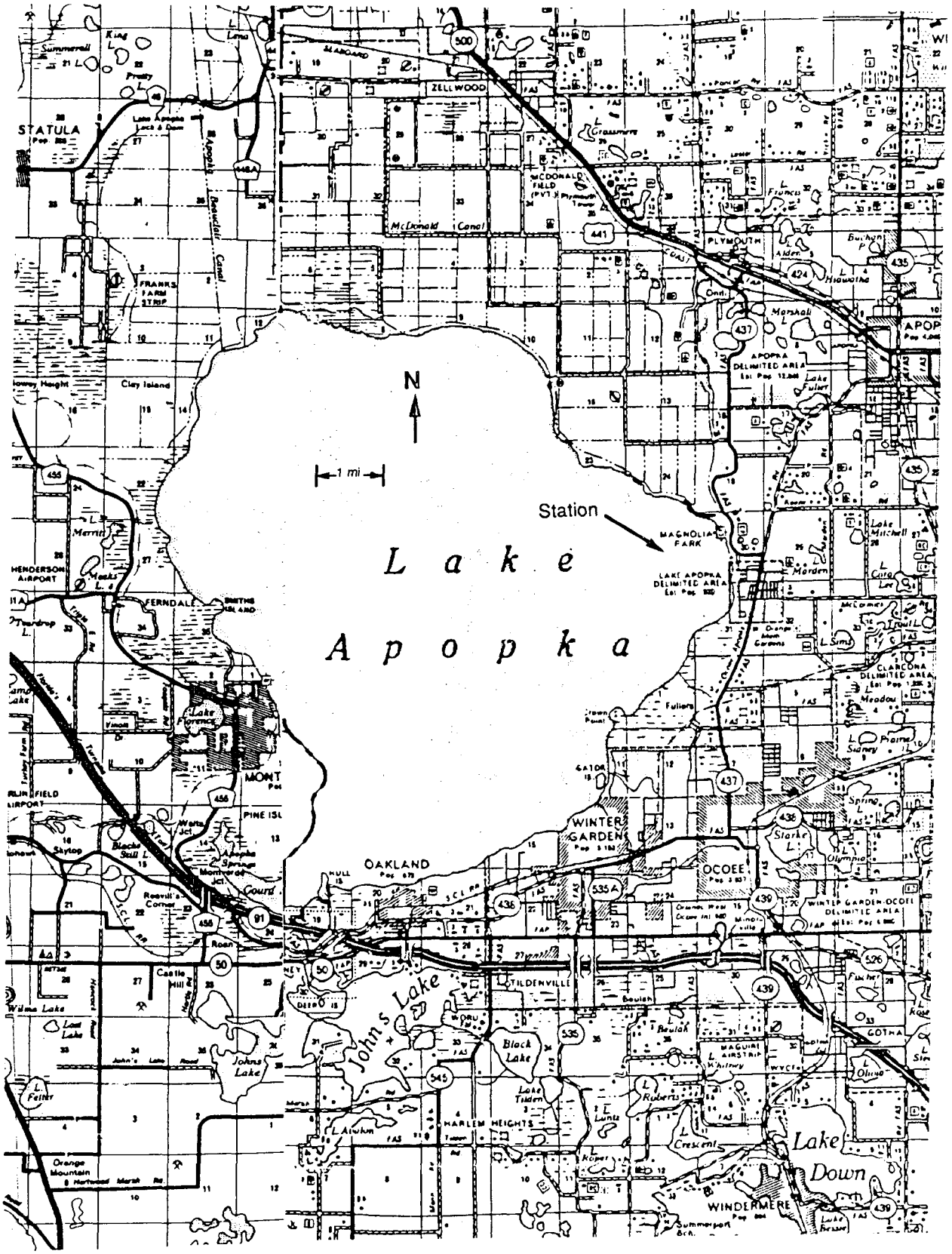

Figure 2.1. Lake Apopka. Sampling station was located 500 m Southwest of Magnolia Park.

where I_0 is subsurface PAR and I_Z is the irradiance at depth Z (Appendix A). I_0 used with each I_Z was obtained by multiplying the value of the deck cell by a correction factor determined each time a profile was measured. The correction factor was obtained by calculating the ratio of PAR for the immediate sub-surface sensor to that for a deck cell. An average K_t (ln units) was calculated from data for each sampling date using values for K_t from depths ranging from 20 to 80 or 90 cm (Appendix C). Data for 0-10 cm were not used in the average because the depth range was so small and the stratum 10 cm above the bottom was excluded in calculating the average because this value was biased by high concentrations of near-bottom suspended materials.

Phytoplankton Primary Productivity

The experimental design for primary productivity and related parameters was based on several considerations. In preliminary experiments, we determined that photosynthetic activity in Lake Apopka produced measurable changes in dissolved oxygen in 1-2 h. We, therefore, decided that methods based on changes in dissolved oxygen could be used to study primary productivity and related parameters. In addition, effects of containment of phytoplankton could be minimized because these measurements could be completed in 2-3 h.

Our first measurements of light penetration using either a Secchi disc or a quantum meter showed that the photic zone would not be deep. From these data we concluded that it would not be feasible to incubate bottles *in situ* at fixed depths because light

intensity changes rapidly with depth. Under these conditions, to obtain a profile of photosynthesis vs. depth, bottles would have to be incubated within 50 to 100 cm of the surface at depths that would be difficult to establish precisely. To avoid these problems, we decided to incubate bottles at constant relative light intensity using a series of neutral density screens to absorb known fractions of incident light. Knowing the fraction of light transmitted by each screen and K_t allowed us to establish the equivalent depth in the water column for each light level employed in the series of neutral density filters. Finally, we also determined that the water column, at least to the depth of the photic zone, was well mixed. As a result we could obtain samples for primary productivity for all treatments from one fixed depth.

Primary productivity was determined using the light and dark bottle oxygen method (Wetzel and Likens 1991). Water was collected from a depth of 30 cm in 750-ml polystyrene tissue culture flasks using a submersible pump that was non-metallic and powered with a 12-V battery. Water was pumped through a specially designed manifold that allowed simultaneous filling of triplicate flasks. Triplicate flasks were filled in a plastic 10-L container. Thus when the overflow from the flasks filled this container, the bottles had been flushed at least three times. Triplicate samples were filled for light treatments of 0, 1, 2, 4, and 6 screens, the dark treatment, and the initial sample. The fraction of light transmitted by the series of screens was 0.57, 0.24, 0.12, and 0.013, respectively. Screens in the form of sleeves were designed so flasks could be incubated with the flat side parallel to the water surface. Dark bottles were painted

with black paint and then wrapped with black tape to ensure no light leakage. Samples were then attached to a rack suspended on floats and incubated at a depth of 10 cm for nominal times, generally 2-3 hours. Short experiments were used because high rates of production on some dates produced supersaturated oxygen levels in light bottles exposed to optimum light for photosynthesis. Bubbles of oxygen under such conditions may not be "fixed" when dissolved oxygen concentrations are measured and thus result in underestimates of photosynthesis. After the incubation, water samples were fixed using the azide method (APHA 1989) and dissolved oxygen concentrations were determined by titration of 250-ml samples with 0.05 N sodium thiosulfate.

Rates of community photosynthesis, net primary productivity (NP) and gross primary productivity (GP), and community respiration (CR) were determined using the following relationships:

$$NP\ (g\ C\ m^{-3}\ h^{-1}) = \frac{(LB - IB)\ (1000)\ (0.375)}{(PQ)\ (t)}$$

$$CR\ (g\ C\ m^{-3}\ h^{-1}) = \frac{(IB - DB)\ (1000)\ (0.375)\ (RQ)}{(t)}$$

$$GP\ (g\ C\ m^{-3}\ h^{-1}) = NP + CR$$

where IB is the initial bottle dissolved oxygen concentration (mg O_2 L^{-1}) at time zero, LB and DB are the light and dark bottle dissolved oxygen concentrations (mg O_2 L^{-1}) at time t, t is the incubation time (h), 1000 is factor used to convert volumetric rates from liters (L) to cubic meters (m^3) (1000 L m^{-3}), 0.375 is the weight ratio of C:O_2 produced during photosynthesis or utilized during respiration (assuming a theoretical molar ratio of 1:1 for photosynthesis and respiration), PQ is the photosynthetic quotient, and RQ is the respiratory quotient. We used a PQ of 1.2 and a RQ of 1.0 which are considered to be typical of algal populations exposed to moderate light intensities (Wetzel and Likens 1991).

Data for GP and NP were plotted as a function of equivalent depth (calculated from the mean K_t value for each profile, Appendix A, and the fraction of light transmitted by each set of screens) and integrated over the depth of the photic zone to obtain areal gross productivity (AGP) and areal net productivity (ANP). The depth of the photic zone was defined as the depth (compensation point) at which GP or NP was zero. On three dates, NP at all depths was negative indicating that CR was greater than net photosynthesis; and on one date NP was slightly positive at one depth and negative at the others (Appendix B). Because rates were low, productivity was not estimated on these four dates (Appendix C). CR determined with this method, however, is the sum of all oxidative processes including phytoplankton respiration. Therefore, CR can be affected by processes other than phytoplankton respiration such as heterotrophic respiration and chemical oxidation.

Rates of NP and GP were plotted as a function of photosynthetically active radiation (PAR) at equivalent depths to obtain photosynthesis vs. irradiance (P vs. I) relationships. These plots also could be used to obtain estimates of maximum net primary productivity (NPmax) for each experiment, but the limited number of irradiance levels used complicated this approach. To obtain estimates of NPmax, we used the highest rate measured for each experiment (see Appendix B). Maximum gross primary productivity (GPmax) was calculated from the sum of NPmax and CR. GPmax was normalized for biomass by dividing by chlorophyll concentration. This biomass specific rate of production (PB_{max}) was termed the assimilation number. A summary of primary productivity and related data for each sampling date is presented in Appendix C.

Chlorophyll a

Samples of lake water (20-50 ml) were filtered on 47-mm Gelman AE glass fiber filters, ground, and extracted for 24 h (-20°C) in 90% acetone (APHA 1989). Chlorophyll a concentrations were determined spectrophotometrically and samples were acidified with 0.1 N HCl, so corrections could be made for phaeophytin. On December 5, 1990, the procedure was changed. Chlorophyll samples were extracted without grinding in acetone:DMSO (50:50) (Shoaf and Lium 1976). Chlorophyll a concentrations were determined fluorometrically with corrections for phaeophytin. Results using the two techniques were comparable. The acetone:DMSO extraction gave lower coefficients of variation than the acetone extraction with grinding.

RESULTS AND DISCUSSION

Lake Level

Data on lake stage obtained during 1989, 1990, and part of 1991 at the gauging station at Winter Garden are plotted in Fig. 2.2. During this period, the lake level varied nearly 2 ft (60 cm). During our study, the level ranged from a high of 66.96 ft to a low of 65.73 ft (Fig. 2.3), or only 1.22 ft (37 cm). It is clear from these data and other historical data (St Johns River Water Management District) that our study was conducted during a period of low water.

Water Temperature

Water temperature at 30 cm during the 18-month study ranged from 15-33°C (Fig. 2.4). Temperatures from mid April to mid October were generally greater than 28°C while temperatures during the remainder of the year were 24°C or lower. Temperatures during the sub-tropical winter which extended from late October to early April were generally <21°C.

Chlorophyll a

The concentration of chlorophyll a ranged from 40 to >200 mg m^{-3} (Fig 2.5). Although chlorophyll varied greatly between sampling periods, maximum standing crops of chlorophyll appeared to be seasonal. Maximum concentrations were generally greater than 100 mg m^{-3} from February through July. Concentrations peaked during the period of high water temperatures in 1990 but then declined from August to October while water temperatures remained high

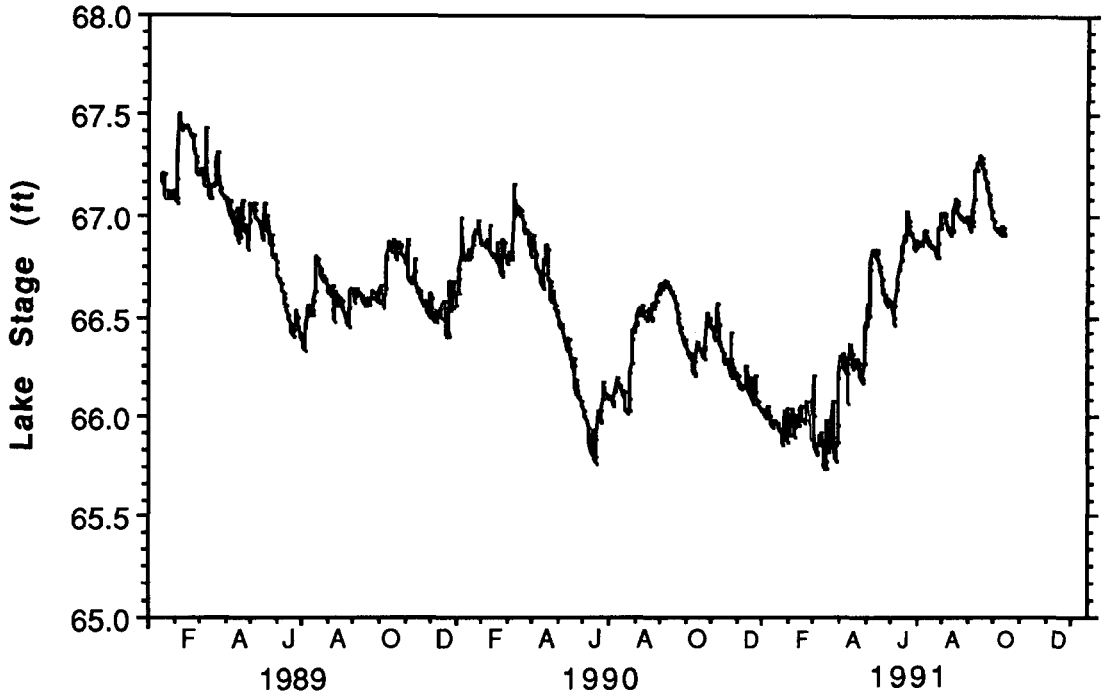

Figure 2.2. Lake stage (level) in feet at Winter Garden. Data are daily records beginning 1 January 1989. Data from guaging station at Winter Garden (St. Johns River Water Management District).

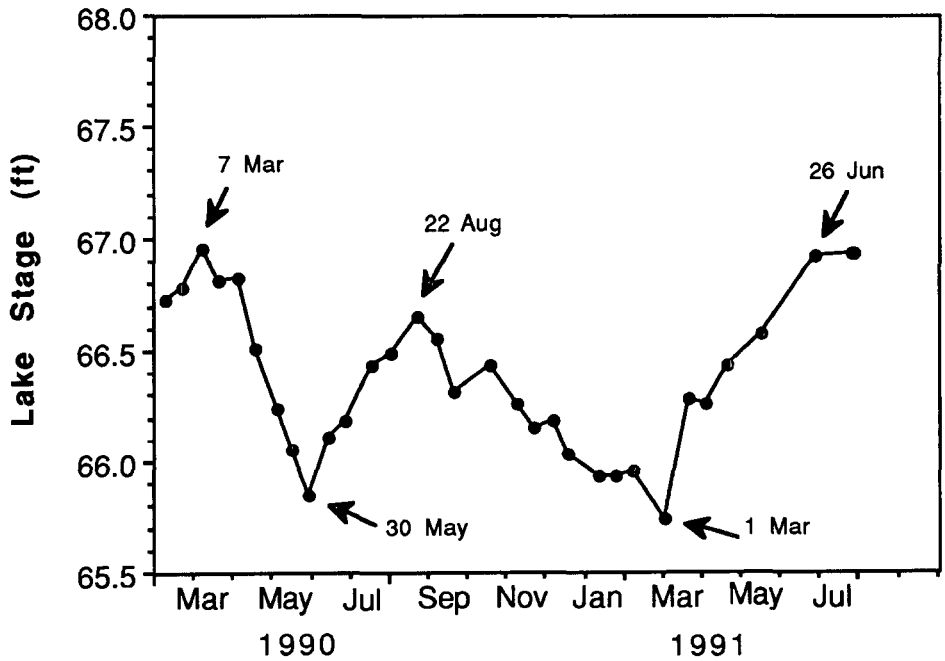

Figure 2.3. Lake stage (level) in feet at Winter Garden. Data are lake stage in feet on sampling dates used in the prsent investigation, Lake Apopka, February 1990 to July 1991. Data from guaging station at Winter Garden (St. Johns River Water Management District).

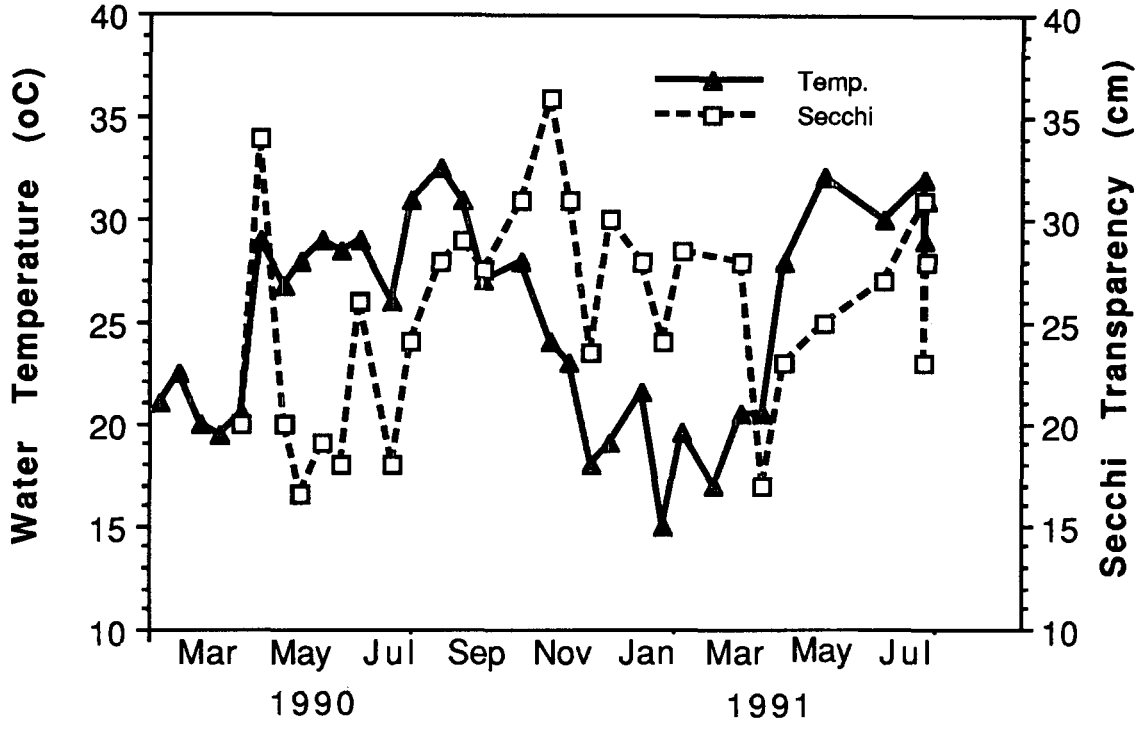

Figure 2.4. Water temperature (°C) at 30 cm and Secchi disc transparency, Lake Apopka, February 1990 to July 1991.

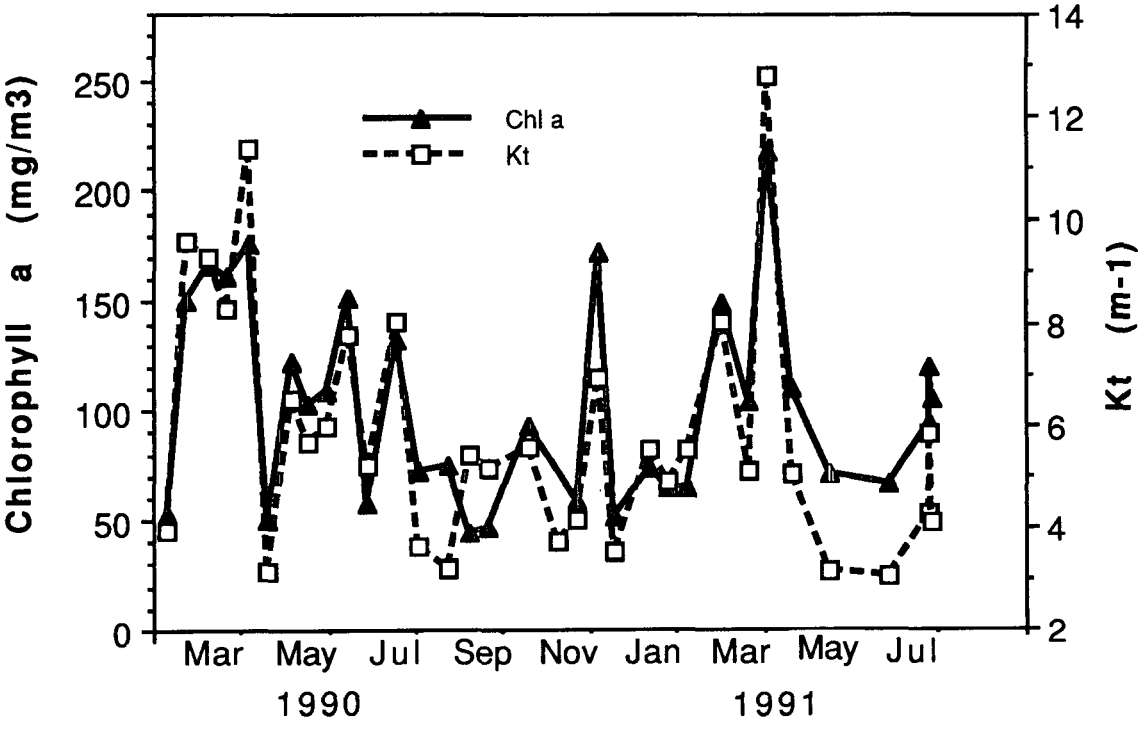

Figure 2.5. Chlorophyll a and extinction coefficient (k_t), Lake Apopka, February 1990 to July 1991.

(Fig. 2.4). Concentrations <80 mg m^{-3} were measured from August to February. The mean and standard deviation of chlorophyll a concentration obtained from our measurements was 104 ± 45.4 mg m^{-3} with a coefficient of variation of 43.6% (Appendix C). Lowe et al. (1992) reported a lower mean and standard deviation, 56.6 ± 11.3, with a lower coefficient of variation, 20.0%. Our mean and standard deviation for Secchi disc transparency was 25.5 ± 5.1 compared to 32 ± 10 reported by Lowe et al. (1992). This comparison indicates that chlorophyll a was higher during our study than during the previous period reported by Lowe et al. (1992).

The general seasonal pattern that can be inferred from our data is high concentrations (100-200 mg m^{-3}) beginning in February or March and continuing until July. During the remainder of the year, lower concentrations ranging from 50-100 mg m^{-3} would be expected. No clear causal mechanism for this pattern is evident at this time, but the increase in concentration is coincident with increasing seasonal water temperature and solar radiation. Short-term increases in chlorophyll a have been related to wind (see section on Short-Term Variability, Chapter 2.31).

Water Transparency

By any measure, Lake Apopka is turbid. Water transparency measured with a 20-cm black and white Secchi disc ranged from 16.5-36 cm during our 18-month study (Fig. 2.4). These data indicate that objects 6.5 inches (16.5 cm) below the water surface would not be visible when the lake is most turbid and objects below 14 inches (36 cm) would rarely be visible. Absorption coefficients of PAR (K_t)

measured with a LiCor Quantum Meter ranged from 3 to 13 (Fig. 2.5). The regression of chlorophyll on extinction coefficient yielded a significant linear regression (r^2=0.769, p<0.0001). The regression of chlorophyll on Secchi disc transparency was also significant (r^2=0.496, p<0.0001). Secchi disc transparency, however, only accounted for 50% of the variation in the data compared to 77% of the variance which could be explained by K_t (Fig. 2.6). This comparison indicates that K_t provides a more precise estimate than Secchi disc transparency of the relationship between chlorophyll and light attenuation in the water column. Precise measurements of transparency with the Secchi disc would not be expected in this turbid lake because a relatively small range in measurements was obtained as the result of limited visibility (Fig. 2.5) and because of the lack of precision in estimating Secchi depths <35 cm.

Although the statistical fit between chlorophyll a and K_t is very good, parameters in the regression of K_t on chlorophyll a can be used to estimate the fraction of the light attenuation due to chlorophyll a ($Y = 1.231 + .0458X$, $r^2 = 0.769$). The y-intercept for K_t of 1.23 m^{-1} is much greater than 0.033 m^{-1}, the K_w of pure water (Parsons et al. 1984), indicating a significant and relatively constant contribution to extinction from non-algal matter over the range of concentrations measured. In addition the slope of the line is 0.046 m^2 mg chl a^{-1}, an absorption coefficient for chlorophyll that is nearly 3-fold greater than the average value cited by Bannister (1974), and approximately twice as large as the maximum value that would be expected (Parsons et al. 1984). This indicates that some fraction of the contribution of non-algal matter is

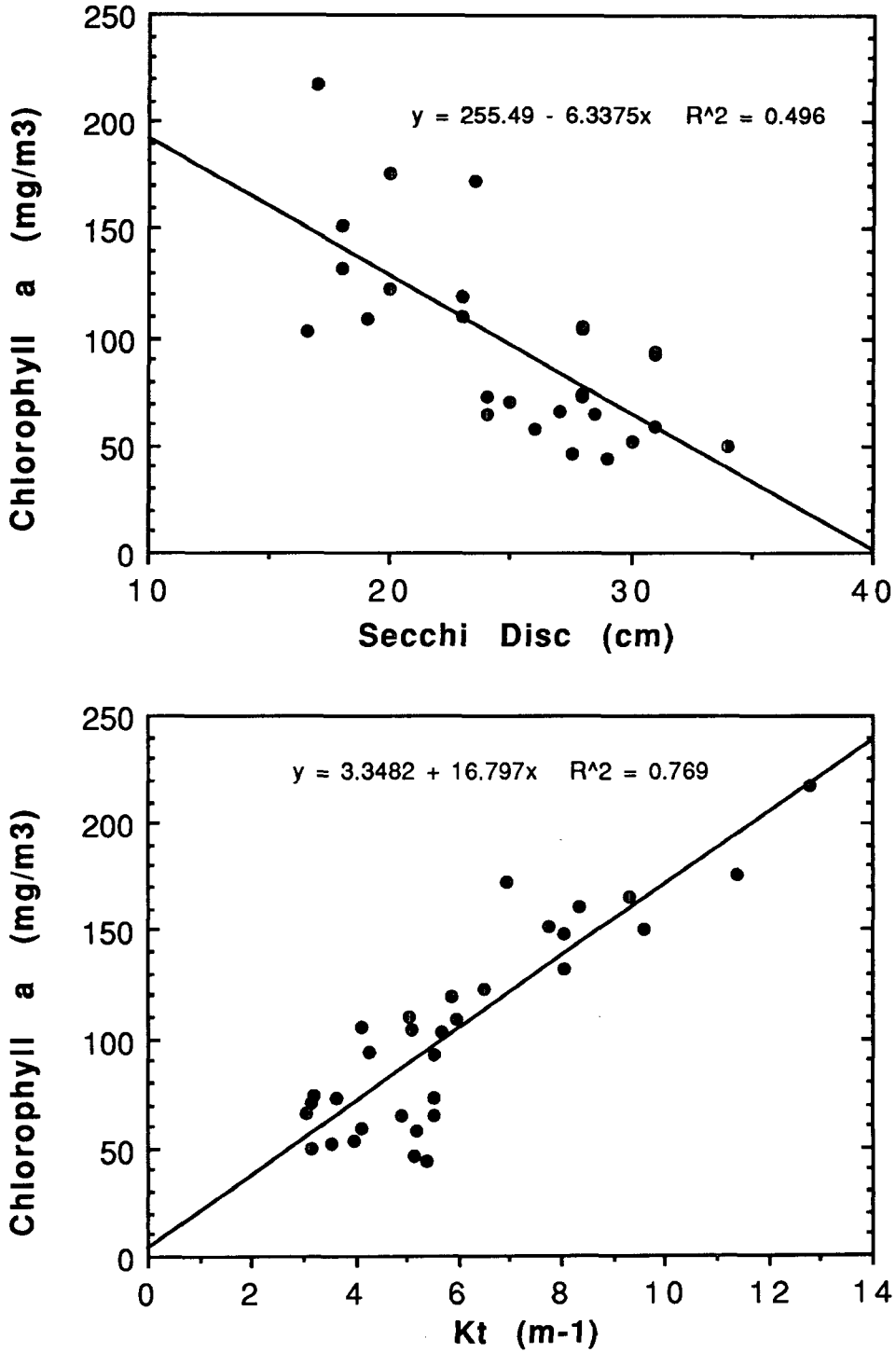

Figure 2.6. Regressions of chlorophyll a on Secchi disc transparency and quantum light extinction coefficient (K_t), Lake Apopka.

proportional to chlorophyll concentration. However, the absorption coefficient attributed to algal matter varies with phytoplankton species composition and other factors and, thus, could be at least half of the value obtained from our regression relationship (Bannister 1974, Parsons et al. 1984)

The good statistical relationship between K_t and chlorophyll *a* concentration shows that chlorophyll concentration could be estimated from K_t. In addition, we can infer that no more than 50% of incident light is absorbed by non-algal matter even after periods of wind resuspension of the nepheloid layer. By contrast, it is estimated that wind resuspension of particulate material in Lake Tämmaren (Sweden) reduced algal production to 15% of that during periods without wind (Hellström 1991). In that lake, turbidity is highly correlated with suspended inorganic matter. In Lake Apopka, we determined that inorganic material (determined from loss on ignition directly) in the near-bottom zone of unconsolidated sediments (a 5-10 cm nepheloid layer) ranged from 3500 to 17000 mg L^{-1} and represented 26% of the suspended solids in this nepheloid layer on three different dates in August and September 1991. On these dates, from 36-49% of the suspended solids in overlying waters was inorganic matter, but concentrations only ranged from 34-40 mg L^{-1}. Thus, contrary to some viewpoints, inorganic material derived from resuspended sediments is not the major component of turbidity in Lake Apopka. This is consistent with measurements by Reddy and Graetz (1991). Resuspension of the nepheloid layer, however, will affect water clarity because we found

at least a 100-fold greater concentration of suspended solids in this layer compared to overlying waters.

Primary Productivity

All of the variables related to primary productivity that were either measured or derived were characterized by high variability. Coefficients of variation for these variables ranged from 20-130%. This variability resulted not only from seasonal changes, but also from large differences that were found frequently from one sampling date to the next (see Appendix C).

Maximum Net Primary Productivity (NPmax) was highest from March to July when rates ranged from approximately 400 to nearly 1,000 mg C m^{-3} h^{-1} (Fig. 2.7). Rates were generally less than 200 mg C m^{-3} h^{-1} from August to February. It also appeared that maximum rates in 1990 were greater than those in 1991 when rates were >400 mg C m^{-3} h^{-1} on only two dates. Multiplying these rates by 12 to extrapolate hourly rates to daily rates gives values that range from 4.8 to 12 g C m^{-3} d^{-1}. By comparison high volumetric rates were obtained in temple ponds in India. Marzolf and Saunders (1984) reported values of 1.5 to 60 g C m^{-3} d^{-1} using diel oxygen changes to estimate primary production and Saunders et al. (1975) reported rates of primary production of 7.8 and 15.2 g C m^{-3} d^{-1} for two ponds using ^{14}C as a tracer. A comparable high value of 7.2 g C m^{-3} d^{-1} was reported for Sanctuary Lake, a hypereutrophic North American temperate lake (Saunders et al. 1975). These maximum volumetric rates of NP are very high; however, as discussed below,

these rates occurred only over a limited photic zone and are not a measure of areal primary productivity.

NPmax was negative on three dates and essentially zero on a fourth date (Fig. 2.7), indicating that the rate of oxygen production by primary producers was less than or only equalled the rate of oxygen consumption. Although this might at first indicate that dissolved oxygen (DO) deficits could cause environmental problems, none of the dissolved oxygen concentrations we measured was much less than 100% of saturation (Appendix B). Frequently, DO was supersaturated, indicating that photosynthetic activity had been high prior to the collection of samples. The fact that our data were obtained early in the morning when the diurnal variation in dissolved oxygen concentration would be near minimum indicates that dissolved oxygen concentrations were never dangerously low prior to any of our sampling periods.

Community Respiration (CR) was highly variable, particularly in 1990 (Fig. 2.8). Rates ranged from approximately 50-350 mg C m^{-3} h^{-1}. None of the rates measured in 1991 exceeded 300 mg C m^{-3} h^{-1}.

Maximum Gross Primary Productivity (GPmax), the sum of NPmax and CR, ranged from <200 to nearly 1200 mg C m^{-3} h^{-1} (Fig. 2.7). There was no obvious seasonal pattern, although the highest values were found from March to July in both years of sampling. GPmax was correlated with K_t (r^2 = 0.480), Secchi transparency (r^2 = 0.437), and chlorophyll *a* (r^2 = 0.262).

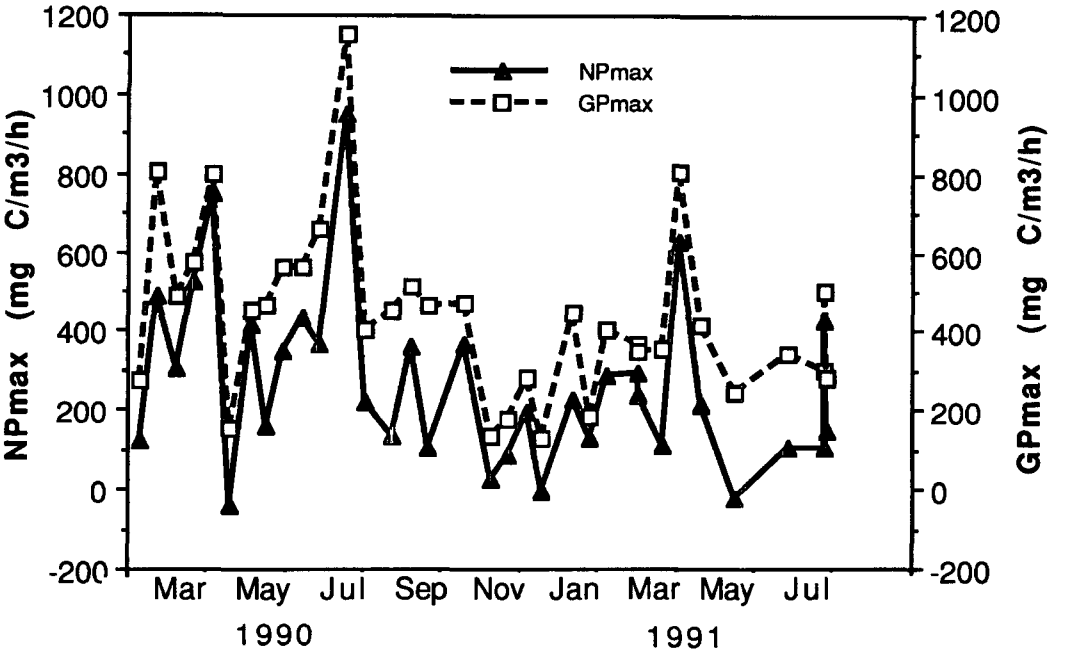

Figure 2.7. Maximum net primary production (NPmax) and maximum gross primary production (GPmax), Lake Apopka, February 1990 to July 1991.

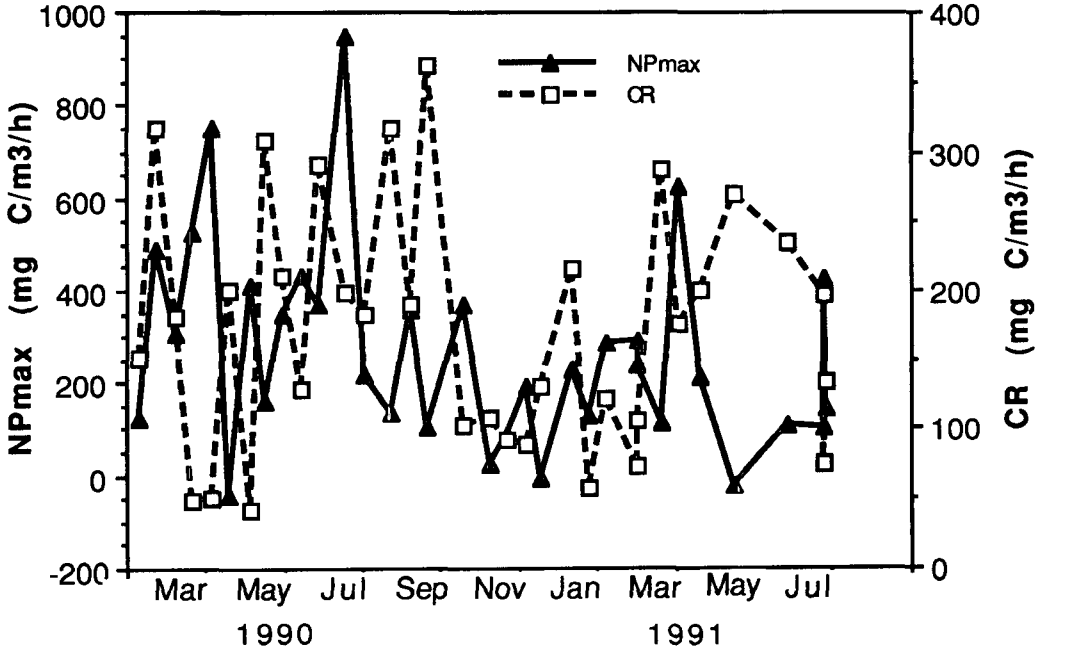

Figure 2.8. Maximum net primary production (NPmax) and community respiration (CR), Lake Apopka, February 1990 to July 1991.

GPmax was variable but had a lower coefficient of variation than NPmax (Appendix C). Less variability may have been found in GPmax because of a time lag between high rates of NPmax and CR (Figs. 2.7 and 2.8). This lag would be expected if CR were largely the result of decomposition of phytoplankton production. However, high CR may also result from wind-resuspended materials that are either chemically or biologically reduced during and after resuspension. In this case, there may not be a direct coupling between NPmax and dissolved oxygen demand in the water column. Thus, two possible explanations for the lower coefficient of variation are that there is either a lag in the community respiration associated with high NPmax or that CR and NPmax are decoupled.

NPmax/CR, the ratio of two measured variables, had the largest coefficient of variation (130%) of any of the primary productivity parameters (Appendix C). This high variability was mainly the result of three ratios that ranged from 10-15 that were obtained from March to May 1990 (Fig. 2.9). This ratio throughout the remainder of the study never exceeded 6.0 and was negative on three dates when NPmax was negative. Large short-term variability was evident also from data obtained on three consecutive days in July 1991. The ratio was 0.52 on July 24, increased to 5.73 on July 25, and then decreased to 1.09 on July 26 (Appendix C).

PB_{max} or assimilation number is a measure of maximum rate of gross photosynthesis per unit of chlorophyll. This biomass specific rate of production ranged from 2-12 (Fig. 2.9). Like most of the primary productivity related variables, this parameter tended not to

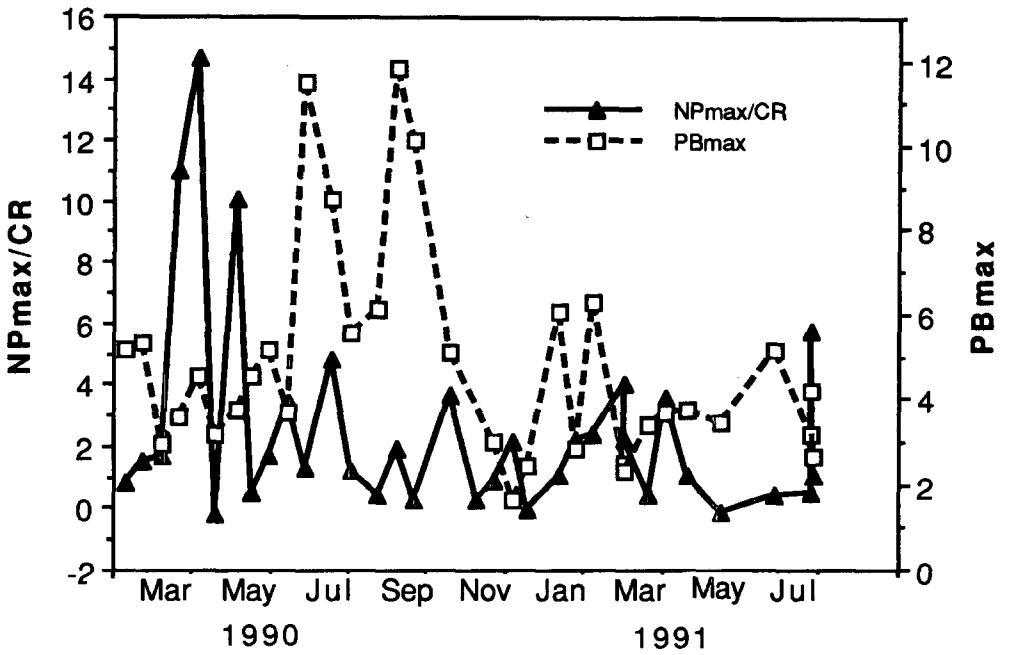

Figure 2.9. Assimilation number (PBmax/Chl a) and NPmax/CR, Lake Apopka, February 1990 to July 1991.

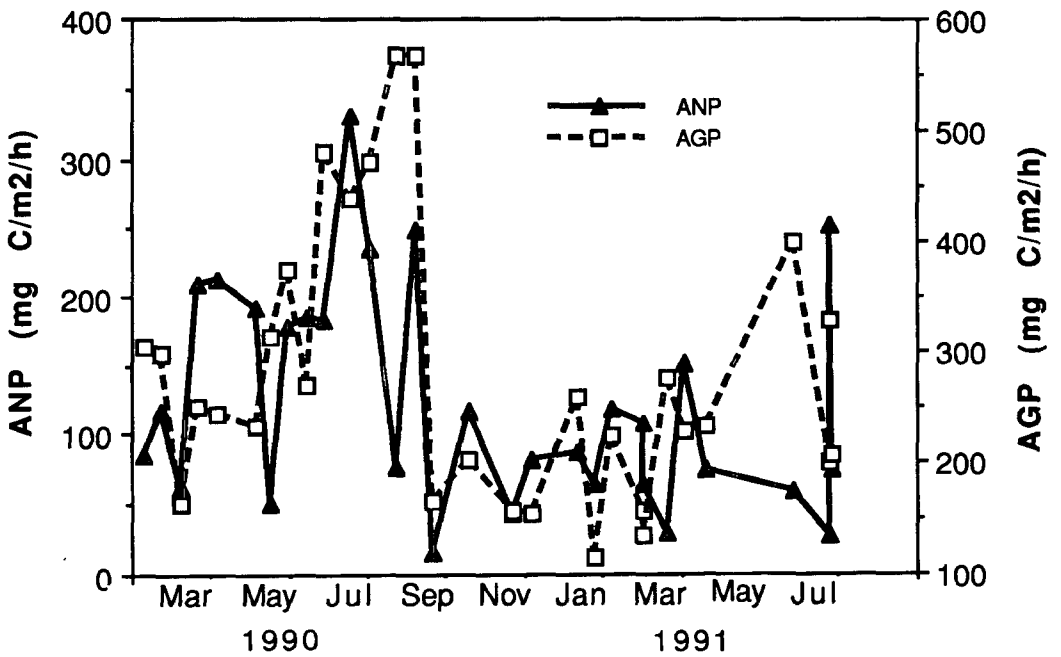

Figure 2.10. Areal Net Production (ANP) and Areal Gross Production (AGP), Lake Apopka, February 1990 to July 1991.

have a seasonal pattern. The temporal pattern, however, closely paralleled that of AGP (Fig. 2.10). The assimilation ratio has been used by some workers as an index of phytoplankton community nutrient status with higher numbers indicating more enriched waters and less nutrient limitation than lower numbers (see Chapter 5). Comparing assimilation numbers with the results of nutrient enrichment bioassays (Chapter 3) does not show any clear relationships. Thus, even though we found a 6-fold range in assimilation ratios, the utility of the ratio for assessing nutrient status of algal communities does not appear to be promising for Lake Apopka.

Areal Primary Production Estimates

Areal Net and Gross Primary Production (ANP and AGP) were estimated by integrating NP and GP to the estimated depth at which NP or GP was zero. The depth of the photic zone (compensation depth) (Figs. 2.11 and 2.12) was positively correlated with AGP (r^2 = 0.342, $p<0.001$), but not with ANP (r^2 = 0.053, $p>0.01$).

The largest values for ANP, >200 mg C m^{-2} h^{-1} (Fig. 2.10), were obtained from March to July 1990 while water temperatures were cool but also during the period of maximum increase in water temperature (Fig. 2.4). However, with one exception, values during the remainder of the study (from August 1990 to July 1991) were <200 mg C m^{-2} h^{-1} which indicate no seasonal relationship.

AGP was highest in spring and early summer in both years of sampling (Fig. 2.10). The high rates coincided with seasonal increases in water temperature (Fig. 2.4) and with periods of

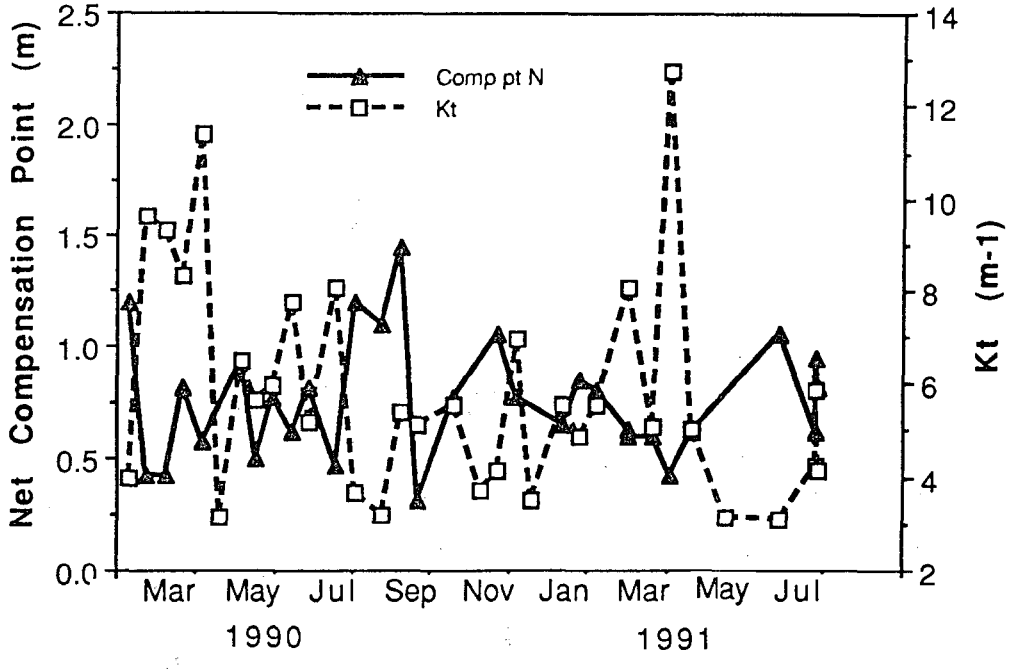

Figure 2.11. Extinction coefficient (K_t) and net compensation depth, Lake Apopka, February 1990 to July 1991.

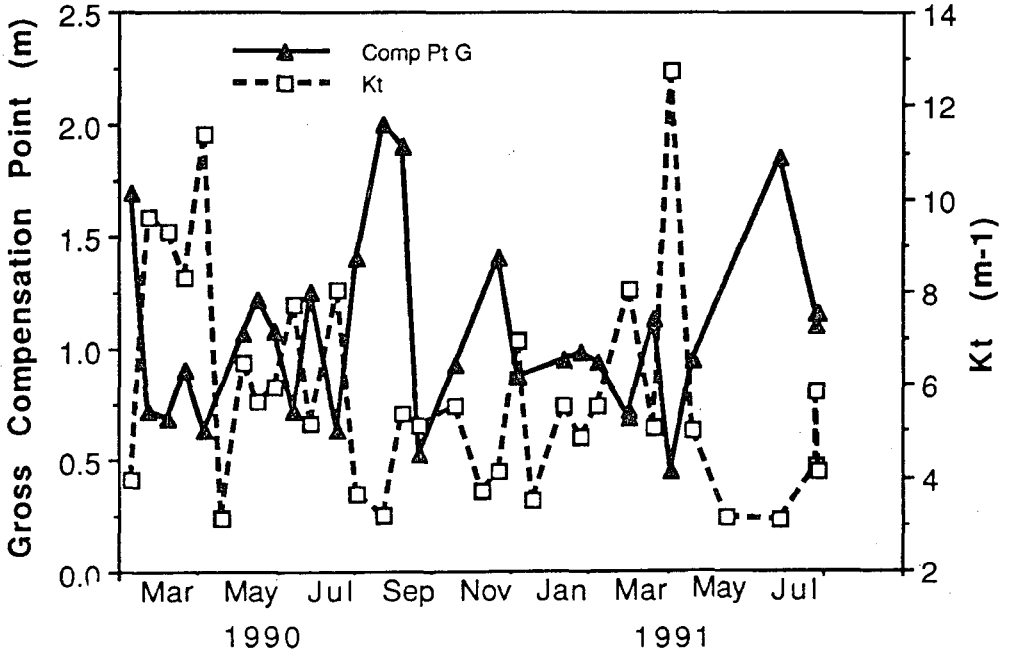

Figure 2.12. Extinction coefficient (K_t) and gross compensation depth, Lake Apopka, February 1990 to July 1991.

increasing lake level (Fig. 2.13). Rates ranged from 100 to 700 mg C m^{-2} h^{-1} with lowest rates occurring from September through April.

Our rates of ANP and GNP based on 2- to 3-h experiments represent a relatively small fraction of the day. We used short-term experiments to minimize effects of excess production of oxygen in light bottles. Rates of areal production are generally expressed in terms of daily rates. One convention used to extrapolate hourly rates to daily rates is to multiply hourly rates by 12. Daily rates for ANP calculated using this convention range as high as 4000 mg C m^{-2} d^{-1}. The high rates for ANP rank among those that characterize systems of maximum net primary production, including tropical and subtropical lakes (Wetzel 1983). We have also calculated AGP because of high and variable rates of CR during our study. The mean for AGP was 271 mg C m^{-2} h^{-1} (Appendix C). Rates during the periods of maximum production ranged from 350 to 700 mg C m^{-2} h^{-1}. Extrapolating these rates to a 12-h day yields daily rates that range from 4200 to 8400 mg C m^{-2} d^{-1}.

Extrapolating hourly rates to a 12-h day directly probably results in an overestimate of either ANP or AGP because our measurements were made at mid-day when incident irradiance is highest. To compensate for this overestimate, we compared the integrated daily PAR measured from sunrise to sunset to the maximum daily integrated PAR (assuming the the mid-day maximum occurred from sunrise to sunset). The ratio calculated from instantaneous rates for three dates averaged 0.65. Multiplying daily rates by this factor, therefore, is an appropriate correction to

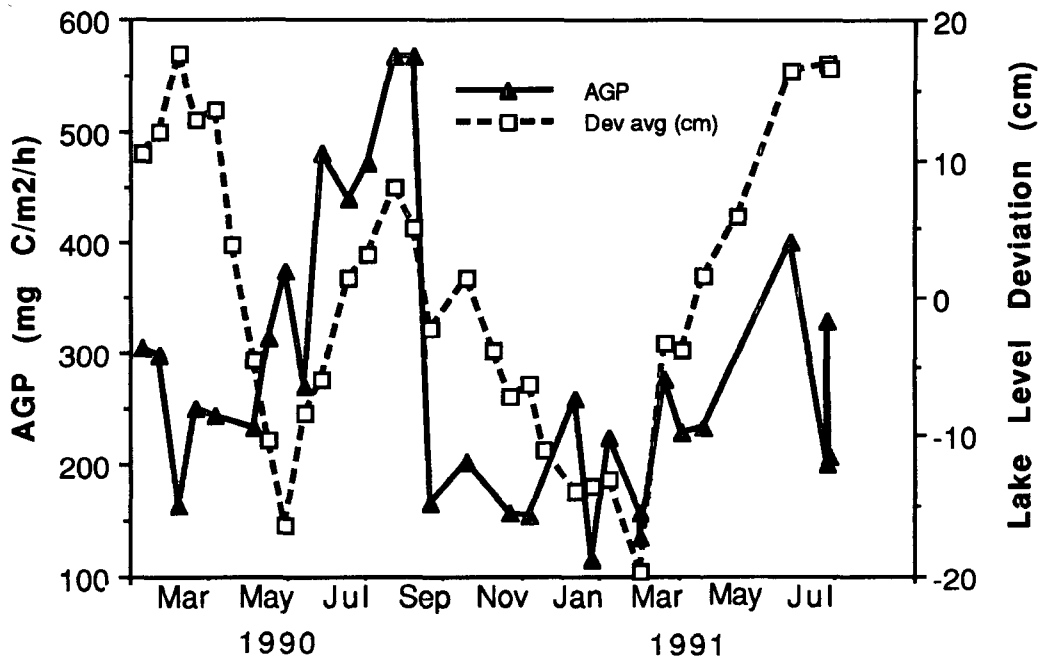

Figure 2.13. Areal Gross Production (AGP) and Deviation (Avg. Dev.) of lake level from 1989-91 mean, Lake Apopka, February 1990 to July 1991.

account for the daily variation in incident PAR if it is assumed that photosynthesis is dependent on irradiance.

Primary productivity was measured over 12 h on 25 July 1991 from 0700 to 1900 h. On this date, primary productivity was measured as usual at mid-day, but in addition was measured also in the early morning and late afternoon. The results of this experiment includes primary productivity measured for 10.35 of the 12.0 h from 0700 to 1900 h (Table 2.1).

Data from the experiment on measuring primary production directly over 12 h (Table 2.1) can be used to check the validity of the extrapolation of mid-day hourly rates to daily rates. In this experiment the hourly rate for AGP during the mid-day experiment was 328 mg C m^{-2} h^{-1}, or 3940 mg C m^{-2} d^{-1} if this rate was maintained for a 12-hour day. Multiplying this rate by 0.65 gives a daily rate of 2560 mg C m^{-2} d^{-1}. This compares with a value of 1900 mg C m^{-2} d^{-1} that is obtained from the sum of the 3 short-term experiments. However, the total time of incubation for the three experiments was only 10.35 h, and an incubation was not conducted from 1017-1112 h which undoubtedly would have been during the period of maximum photosynthesis. If we assume that carbon was fixed at the mid-day rate during the missing morning period (355 mg C m^{-2}) and at half the mid-day rate for the missing afternoon period (175 mg C m^{-2}), the corrected total for AGP is 2430 mg C m^{-2} d^{-1}, a value that is in good agreement with the extrapolated rate of 2560 mg C m^{-2} d^{-1}. These data indicate that our extrapolation using a factor of 0.65 to correct for daily variation

Table 2.1. Comparison of primary productivity data from three *in situ* incubations on 25 July 1991 at successive times to simulate a 12-h day. NPmax and GPmax are maximum net and gross primary productivity (mg C m^{-3} h^{-1}), ANP and AGP are areal net and gross primary production (mg C m^{-2} h^{-1}), Total AGP and ANP are areal gross production (mg C m^{-2}) for a time period, and Light is total PAR flux (mol m^{-2}) for the time of incubation.

Time	NPmax	GPmax	ANP	AGP	Total ANP	Total AGP	Light
0712-1017	342	383	143	190	441	586	11.54
1112-1415	424	486	252	328	769	1000	18.72
1447-1900	54	174	16	74	67	312	3.81
	Total				1277	1898	

in irradiance for mid-day experiments provides a good approximation of *in situ* areal production rates.

To convert estimates of AGP to true values of ANP requires that they be corrected for respiration by phytoplankton. Our estimates of community respiration (CR) include respiration from all sources (phytoplankton, heterotrophs, and possibly oxygen consumption for chemical oxidation). CR during our study averaged 172 mg C m^{-3} h^{-1}, or 39% of the average rate of GPmax, but as indicated this is obviously an overestimate of phytoplankton respiration. On 25 July 1991 (Table 2.1), Total ANP was 67.3% of Total AGP, indicating that CR was 33% of AGP. The highest hourly rate for AGP in Lake Apopka was 700 mg C m^{-2} h^{-1} (Fig 2.13). Multiplying this rate by 12 and 0.65 to convert to a daily rate and by 0.67 to convert to ANP yields a rate of 3660 mg C m^{-2} d^{-1}. This net rate would be higher if

phytoplankton respiration could be estimated. Although our data provide no good means of estimating phytoplankton respiration, it is clear that Lake Apopka is highly productive because the highest daily rates of areal net primary production reported from subtropical and tropical lakes range to as high as 3800 mg C m^{-2} d^{-1} (Wetzel 1983).

P vs. I Relationships

Our data on P vs. I relationships were obtained from *in situ* incubations using neutral density filters to simulate depth. Because only five light levels (0, 1, 2, 4, and 6 screens) were used, a limited set of irradiance levels for each experiment was available to determine photosynthetic parameters of interest, particularly, a, the initial slope of the P vs. I curve; P_{max}, the maximum rate of photosynthesis; and I_k, the irradiance at which P is half of P_{max}. Different estimates of P_{max} have been discussed above when we presented data on NPmax and GPmax and on PB_{max} (biomass specific GPmax normalized to chlorophyll a concentration). Data on these estimates of P_{max} are given in Appendix C.

It is clear that the photosynthetic parameters varied widely during the course of our study. Two examples can be used to illustrate the wide range in values (Fig. 2.14). P_{max} was found at a very low irradiance, approximately 130 µmol m^{-2} s^{-1} on 7 February 1990, compared to about 500 µmol m^{-2} s^{-1} on 6 September 1991. These data on P_{max} indicate that phytoplankton in Lake Apopka can be adapted to a range of light intensities. The community on 7 February was adapted to relatively high range in irradiance as evidenced by the relatively small effect of high levels of irradiance

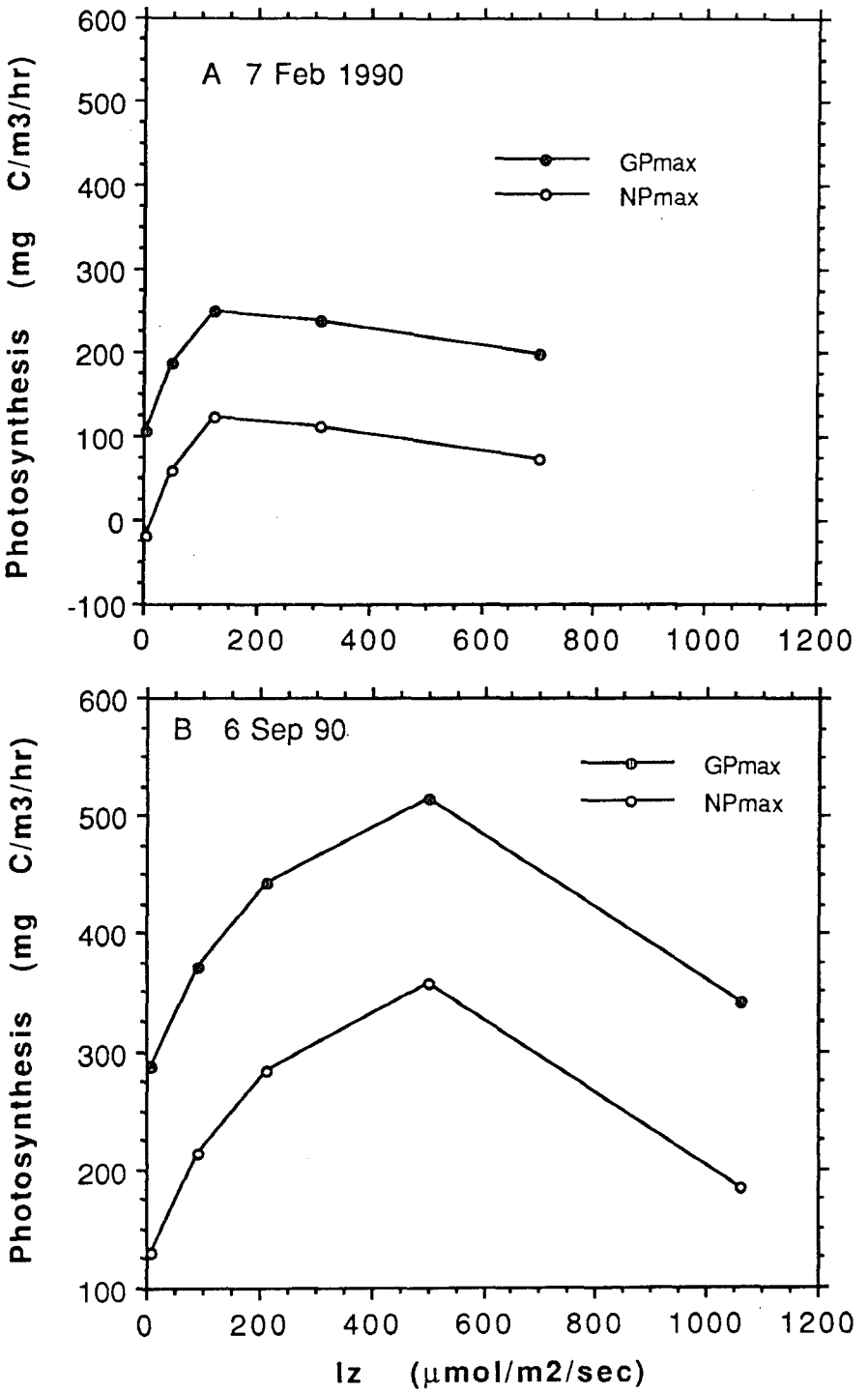

Figure 2.14. Photosynthesis vs irradiance (P vs I) relationships of Lake Apopka phytoplankton on 7 February 1990 (A) and 6 September 1990 (B).

on photosynthesis. This community can be described as being shade adapted because the highest rate of photosynthesis was at approximately 130 μmol m^{-2} s^{-1}. By contrast, the community on 6 September appeared to be adapted to high irradiance because the photosynthetic maximum was at approximately 600 μmol m^{-2} s^{-1}. The much lower rate of photosynthesis at the one irradiance level >600 μmol m^{-2} s^{-1} suggests inhibition of photosynthesis at the irradiance level of 1100 μmol m^{-2} s^{-1}. The degree of adaptation to high levels of irradiance cannot be described from this experiment because no data were obtained for irradiances between 600 and 1100 μmol m^{-2} s^{-1}.

Phytoplankton in Lake Apopka must survive under widely varying conditions of irradiance. During calm periods, some phytoplankton actually sink to aphotic depths where there is essentially no light for photosynthesis. The algae survive in a resting stage and are capable of photosynthesis soon after they are returned to the photic zone by periodic resuspension events (Carrick et al. in prep). By contrast, algae, in a turbulent environment (periods in which cells are maintained in suspension and do not sink), would not be exposed to high, inhibitory irradiances for long periods of time. Adaptation of phytoplankton to the very low average light intensities that are found over most of the water column in Lake Apopka, therefore, would enable phytoplankton photosynthesis to proceed at high rates in this shallow, turbid environment. Thus, it seems clear that phytoplankton in this lake must be adapted to the short-term variation in the physical environment that characterizes this shallow, turbid lake.

Short-Term Variability

Results for many of the parameters we measured during 18 months of study are characterized by large ranges in values. It seems clear from the data presented above that this variability can not be attributed to seasonal variation because of the large variability within seasons. This pattern in the results and data we have collected suggests that variability results from events occurring on time scales of a few days.

An estimate of temporal variability in both physical and biological conditions in terms of per cent coefficient of variation (CV) was obtained by dividing the standard deviation for the mean for each variable by that mean and then multiplying by 100. Values for CV of variables measured during the study are shown in Fig. 2.15.

Water temperature and absorption coefficient, measured at a single site at two-week intervals, exhibited large variances with temperatures changing as much as 8°C between samples (Appendix C). GPmax and chlorophyll a also showed extreme temporal variability (coefficients of variation >50%) and did not correlate significantly with temperature or lake level (all r^2 <0.15), two variables which are a measure of seasonal change. High variability in Lake Apopka, therefore, is pronounced and indicates the dynamic nature of the system.

Short-term variation in Lake Apopka (1-3 d) can also be considerable as shown by data collected from 27 February to 3 March 1991 (Fig. 2.16). We measured a nearly two-fold increase in phytoplankton chlorophyll a and carbon, as well as a 50% reduction in

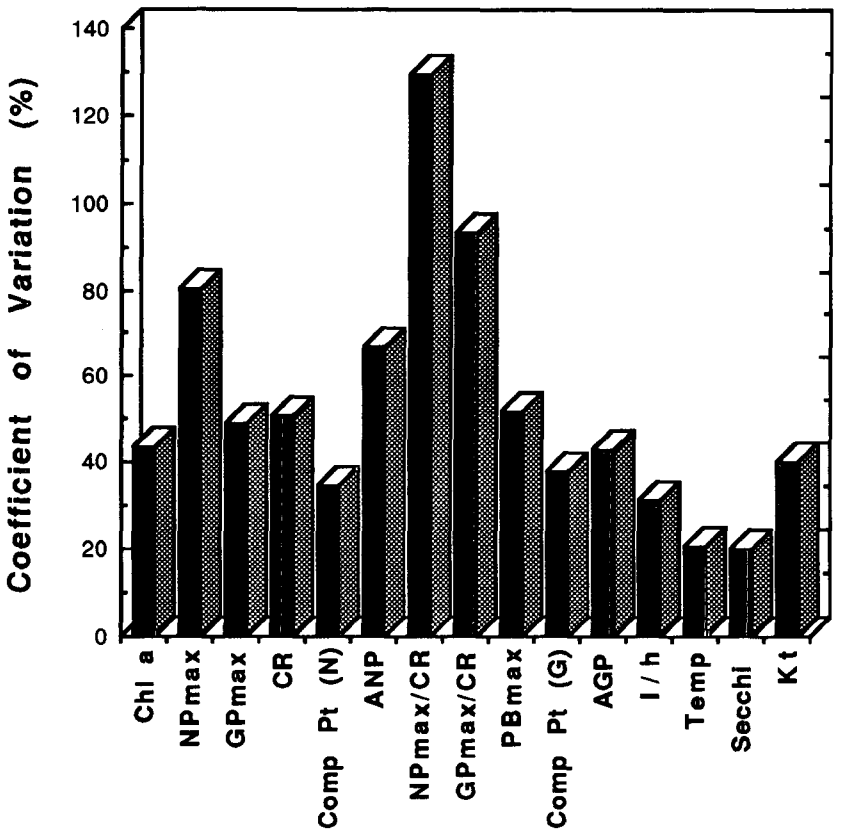

Figure 2.15. Coefficient of variation (%) in parameters measured on 33 sampling dates, Lake Apopka, February 1990 to July 1991.

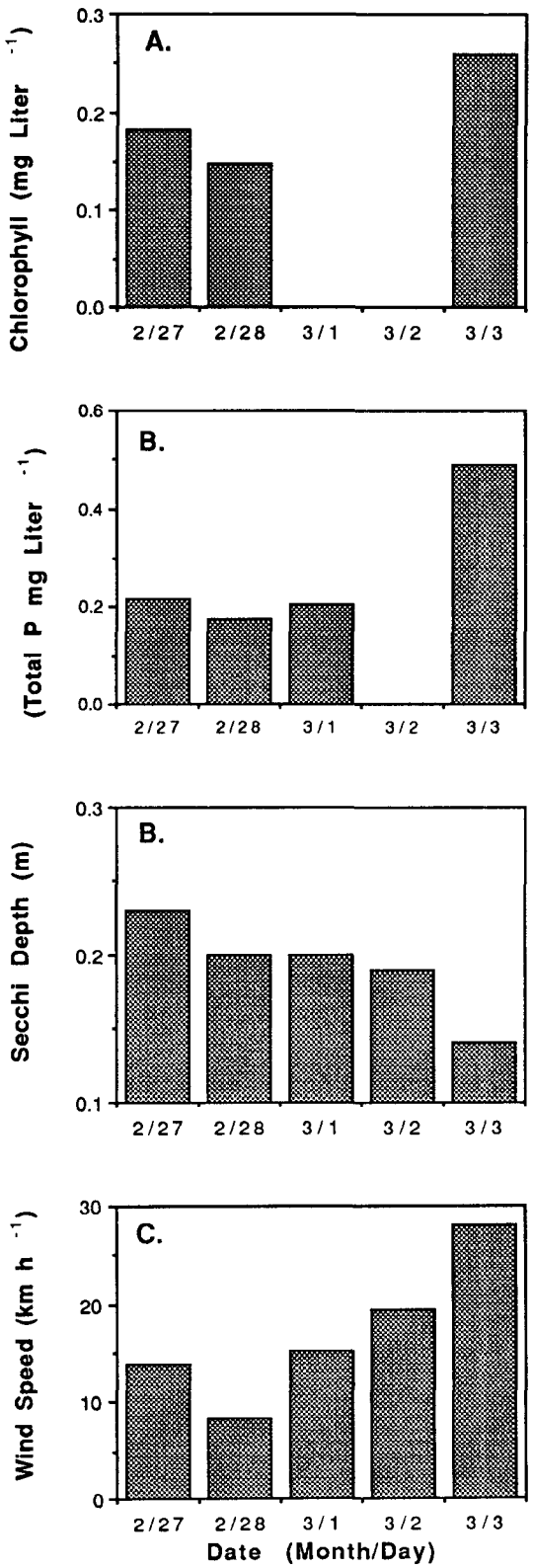

Figure 2.16. Estimates of (A) chlorophyll a, (B) total phosphorus (C) Secchi depth, and (D) wind speed measured on five consecutive days (27 February to 3 March 1991) in Lake Apopka, Florida. Wind speed was obtained from National Oceanic and Atmospheric Administration data base for Orlando International Airport.

light penetration (as Secchi depth) over a 3-d period. Water column nutrient concentrations also increased during this period. Total phosphorus increased nearly two-fold and total nitrogen increased by 20%. These changes were associated with a storm that increased wind speed over a period of two days (9.4 to 17.4 mph, daily averages) and resuspended unconsolidated lake sediments. In contrast, under calm conditions the water column stabilizes and surface blooms of *Botryococcus braunii* commonly develop in a matter of hours (Shireman and Opuszynski 1992; personal observation).

Variation in phytoplankton productivity on short time scales also was found in data collected from 24-26 July 1991. During this time primary productivity, measured as NPmax, GPmax, or ANP, was highest on July 25 (Fig. 2.17). NPmax increased by a factor of four from July 24 to July 25 and GPmax and ANP were 60% greater on July 25 than on July 24. The highest variability in parameters measured during the 18 months of study was also found in NPmax and the ratio NPmax/CR (Fig. 2.15). PB_{max} was also highest on July 25, indicating that nutrient conditions for phytoplankton growth had improved on that date. It should also be noted that irradiance was higher on July 25 than on July 24 indicating that the variation may have been a direct response to changes in irradiance.

Some of the variation we have found may be the result of variation in nutrient loading which we have not studied. It was determined from an analysis of correlation coefficients that change in lake level was not significantly related to any of the variables measured during this study. However, episodic events, such as runoff of nutrients, that also affect lake level may be important in

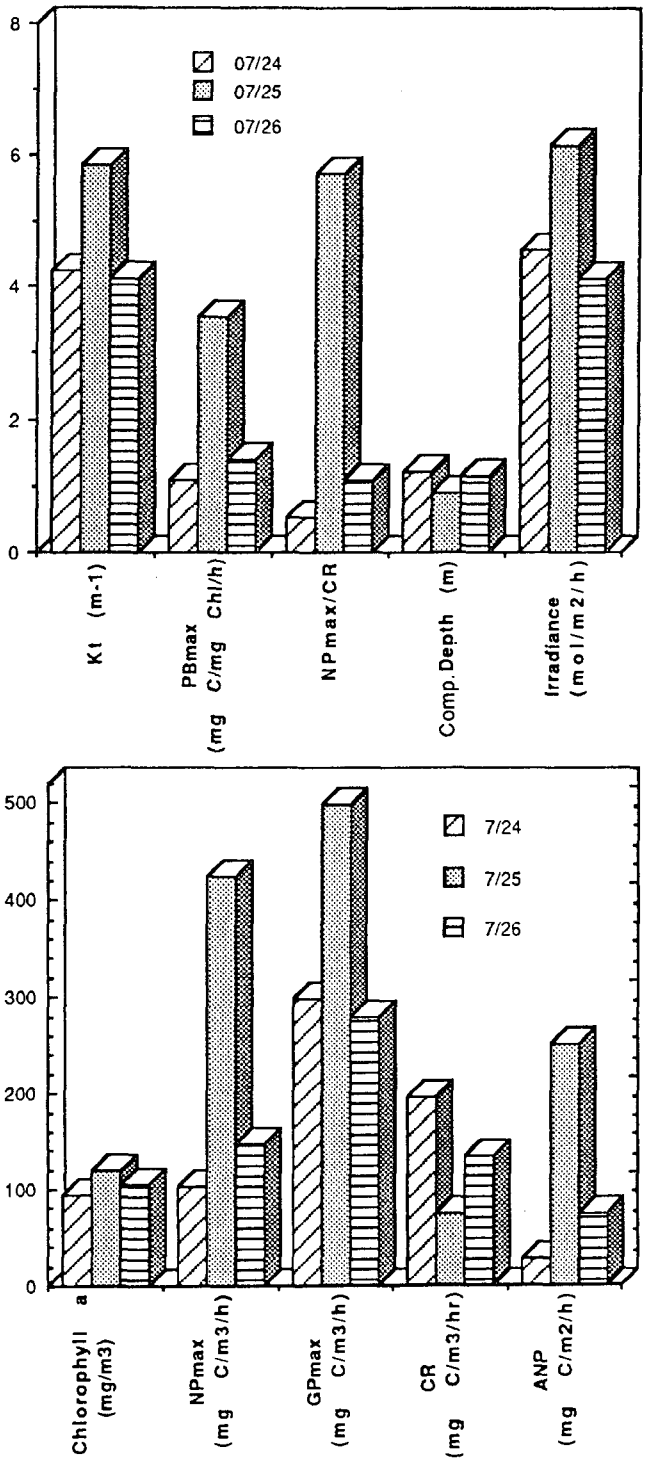

Figure 2.17. Variability in parameters related to primary productivity, Lake Apopka, 24-26 July 1991.

short-term variation. We believe that much of the variability in data presented in this chapter is the result of short-term changes in dynamics of this shallow system. We believe the major factors that control short-term variability are associated with meteorological events. These events include wind-induced resuspension of phytoplankton and nutrients in unconsolidated particulate materials that are present in high concentrations near the bottom of the lake (in a nepheloid layer) and short-term changes in incident irradiation that would be associated with storms and frontal systems. Resuspension events bring sedimented algae from the aphotic zone into the euphotic zone of the lake and short-term changes in solar irradiance influence light-dependent rates of primary production (Carrick et al. in prep.).

SUMMARY AND CONCLUSIONS

The visual perception that Lake Apopka is a shallow, turbid environment is substantiated by data on light extinction measured with a quantum meter and by data on water transparency measured with a Secchi disc. K_t (ln base) ranged from approximately 3-13 m^{-1}. From regression analysis, it was estimated that 1.2 m^{-1} of these extinction units were attributable to some background level of suspended non-algal matter and dissolved stains and that at least half of the remaining extinction could be attributed to chlorophyll *a* in algae. From a practical viewpoint, suspended materials in the water during times of our measurements limited visibility (Secchi depth) to depths ranging from 16.5 cm (approximately 6 inches) to 36 cm (approximately 15 inches).

High levels of nutrient enrichment clearly place Lake Apopka in the classification of hypereutrophic lakes. Although not reported in this Chapter because data were not collected routinely, we measured total phosphorus levels as large as 200 μg P L^{-1}. One of the undesirable manifestations of increasing nutrient enrichment is an increase in algal standing crops. The mean chlorophyll *a* concentration obtained from our study was 100 μg L^{-1} with a number of values that ranged from approximately 150-200 μg L^{-1}.

Nutrient enrichment has also caused high rates of primary production which would be expected in this hypereutrophic lake. The rates we obtained are comparable in magnitude to the highest rates reported in the literature for lakes. We believe that our estimates may be conservative because measurements were made with the oxygen method under conditions of oxygen supersaturation. Under these conditions some of the oxygen produced may form bubbles in the water. Thus, oxygen produced during photosynthesis may escape from the bottles used for measurements.

We conclude that the large variability in variables we measured during this study is the result of short-term variation (2-5 days) in environmental processes. We believe the major factors that control short-term variability are associated with meteorological events. These events include wind-induced resuspension of phytoplankton and nutrients in unconsolidated particulate materials that are present in high concentrations near the bottom of the lake (in a nepheloid layer) and short-term changes in incident irradiation that would be associated with storms and frontal systems. Resuspension events bring sedimented algae from the near-bottom

layer, an aphotic zone, into the euphotic zone of the lake and short-term changes in solar irradiance influence light-dependent rates of primary production (Carrick et al. in prep.).

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Chapter 3

NUTRIENT ENRICHMENT BIOASSAYS

INTRODUCTION

An understanding of the relationship of phytoplankton dynamics to nutrient supplies requires knowledge of nutrient stoichiometry and phytoplankton physiology. Redfield et al. (1963) studied carbon (C), nitrogen (N) and phosphorus (P) stoichiometry of pelagic marine plankton and determined a C:N:P atomic ratio of 106:16:1 respectively. Researchers have since applied this ratio to determine the status of phytoplankton nutrient limitation. Generally, an N:P ratio near 16:1 is considered to be optimal while less than 10:1 would indicate an N-deficient state and a ratio greater than 20:1 would indicate a P-deficient state. Historical total N and total P concentrations for Lake Apopka are 4.03 mg N L^{-1} and 0.192 mg P L^{-1} (Huber et al. 1982) which converts to a 45:1 N:P atomic ratio. This would suggest that the phytoplankton in Lake Apopka are strongly P-limited. Reddy and Gratz (1991) reported that measured concentrations of ammonium N and soluble reactive P indicated neither nutrient should limit primary production. Sommer (1989), in work on a shallow hypereutrophic lake, concluded that nutrient stoichiometry alone is not adequate to explain spatial and temporal variability in phytoplankton biomass. The missing component that ties nutrient stoichiometry to phytoplankton biomass is the physiological adaptation of the phytoplankton to a dynamic environment.

Phytoplankton nutrient physiology has been studied by using physiological indicators such as alkaline phosphatase activity, NH^4+

stimulated carbon fixation in the dark, nutrient uptake kinetics, and nutrient enrichment bioassays (Van Donk et al. 1989, Dodds and Priscu 1990). Each of these methods has useful applications as well as inherent limitations (see Chapter 5).

The approach employed in this chapter is nutrient enrichment bioassay (NEB). NEB can be categorized in two forms; those that assay the nutrient content of filtered lake water by growth response of laboratory cultured species and those that assay the growth response of the natural phytoplankton community in lake water. We used the latter for the same reasons given by Schelske et al. (1978): the natural phytoplankton community is representative of the lake ecosystem and it has a physiological history due to the environmental conditions in the lake. Nutrient stoichiometry and NEB together provide complementary insight to the interactions of the phytoplankton community and its environment.

Researchers have evaluated the scale and design of NEB, large versus small volume, and the environment, laboratory or *in situ*, where they are conducted (see Schelske 1984). In any NEB design, be it *in situ* or laboratory based, the phytoplankton community is isolated from the complex of natural factors such as turbulent mixing, nutrient fluxes, and light regimes. The experimental conditions must be considered in the interpretation of the results. The utility of NEB is the ability to partition and control some of the confounding environmental factors so that specific nutrient conditions can be assessed. NEB is the only method that can be used to assess which nutrient is potentially the first to limit phytoplankton biomass and to rank nutrients in the order of potential

limitation (Maestrini et al. 1984). Laboratory-based NEB can be used to provide a secondary inference on the potential of light as a limiting factor because light becomes a controlled variable (Sommer 1989).

The nutrient status of the phytoplankton community in Lake Apopka is influenced by several factors which include total phytoplankton biomass, grazing pressure, available nutrient pools, turnover rates for internal nutrient pools, and external nutrient loading. Static measurements of the primary nutrients, nitrogen and phosphorus, are strongly affected by episodic events such as wind mixing in this shallow temperate to subtropic lake. Because of these episodic effects, concentration of the different forms of nitrogen and phosphorus can vary widely. This large short-term variation is one factor which confounds the modeling of phytoplankton biomass as a function of these nutrient concentrations alone. It is necessary, therefore, to define the response of the phytoplankton community in terms of the quantity and form of available nutrients.

Nutrient enrichment bioassay was used in this study to measure the dynamic response of phytoplankton to nutrient enrichment under controlled conditions. An increase in phytoplankton biomass stimulated by nutrient enrichment when compared to a non-enriched control indicates that nutrient to be limiting under the experimental conditions and thus to be potentially limiting in the environment. It must be recognized that the information obtained from NEB is only valid for the time and place the phytoplankton community was sampled. The extent of temporal or spatial sampling that may be

required depends on the objectives of each specific study of nutrient limitation.

The objectives of this series of nutrient enrichment bioassays were to: 1) determine what nitrogen form is utilized and stimulates Lake Apopka phytoplankton growth, 2) evaluate concurrent *in situ* and laboratory-based nutrient-enrichment bioassays for determination of appropriate assay protocol, 3) qualitatively rank the limiting status of N and P on the phytoplankton communities, and 4) determine if co-limitation by N and P occurs, i.e. whether either N or P is a secondary limiting nutrient.

MATERIALS AND METHODS

Sample Collection

Monthly water samples were collected from a fixed station approximately 500 m southwest and offshore of Magnolia Park on the northeast shore of Lake Apopka (see Fig. 2.1). Water was pumped from 30 cm below the water surface using a low-pressure open-impeller 12-volt pump and transported to the laboratory in light-shaded 20-L acid-washed polycarbonate carboys. Water samples were protected from extreme temperature changes during transport and stored for no more than 24 h prior to the start of the experiments.

Experimental Design

A 2^2 factorial design with qualitative fixed effects was used for all experiments. The independent variables were nutrient type and the response variable was phytoplankton biomass. Each nutrient

was tested at two levels, enriched and ambient. All treatments were run in duplicate or triplicate to assess within treatment variability. Elser et al. (1990) recommended that replication is needed to test primary nutrient effects and nutrient interaction. Analysis of variance, SAS Institute Version 6.04 GLM Procedure (SAS 1989), was employed to determine significant treatment and interaction effects on the maximum response for each treatment for the duration of the assay excluding the initial biomass value. Duncan's multiple-range test was used to do a pairwise comparison between means of the maximum response for treatments. A $P<0.05$ was used to determine significance.

Nutrient treatments

Nutrient treatments are given in Table 3.1. The selection of nutrient treatments was based on the recommendations of Schelske (1984) that the amount of nutrient enrichment should be "realistic" and, as a rule of thumb, no more than twice the total ambient concentration for that nutrient. The N:P ratio was 10:1 by weight and 22:1 by atomic ratio. This ratio was based on preliminary results from assays that indicated N was the primary limiting nutrient in Lake Apopka.

Table 3.1. Treatments used for nutrient enrichment bioassays.

TREATMENT	ADDITION
Control (C)	none
Nitrogen (N) *	400 µg N L^{-1}
Phosphorus (P) **	40 µg P L^{-1}
Nitrogen + phosphorus (N+P)	400 µg N L^{-1} + 40 µg P L^{-1}

* = N as KNO_3
** = P as K_2HPO4

In Laboratory Assays

All laboratory assays were conducted in 500-ml Erlenmeyer flasks that had been previously acid washed. Lake water in a polycarbonate carboy was continuously mixed on a stirring plate while 300-ml aliquots were drawn off into each flask. All treatments were run in triplicate. Flasks were incubated in a temperature-controlled water bath with the temperature set to mimic ambient lake temperature as closely as possible. Temperatures utilized ranged from 20°C to 30°C. Light intensity was fixed at 120 µmol m^{-2} s^{-1}. The photoperiod was set for 12/12 dark to light hours respectively during the months from October through March and 11/13 dark to light hours during the months from April through September. Samples were taken at the start of the assay and once every 24 h. The duration of the assays ranged from 72 to 240 h.

The first two assays (December 7, 1989 and January 25, 1990) were setup using a 1:1 dilution of whole lake water with lake water that had been filtered through a 0.45 µm pore filter. The third assay (February 22, 1990) was set up to determine the effect dilution had

on phytoplankton growth response. One complete set of treatments with replicates (C, N, P, N+P) was setup with a 1:1 dilution as per the prior assay dates while a second set of treatments was setup using undiluted whole lake water. All subsequent assays were run on undiluted whole lake water with two exceptions. The assays on April 19 and August 23, 1990 were run using the 1:1 dilution. This modification was made to reduce the planktonic biomass while maintaining the same dissolved constituent concentrations. The purpose was to allow greater potential growth of the phytoplankton while running more detailed nutrient uptake and physiological indicator studies referred to in Chapters 4 and 5 of this report.

Comparison of Nitrogen Sources

The form of available nitrogen to use for the nitrogen enrichment treatment was determined by running two bioassay series (December 7, 1989 and January 22, 1990) on Lake Apopka water comparing the phytoplankton growth response to nitrate nitrogen and ammonium nitrogen. These assays were run using the laboratory assay method and the 1:1 dilution of whole lake water with filtered lake water as previously described. Treatments included all those listed in Table 3.1 with the addition of three treatments: NH_4 = 400 µg N L^{-1} (as NH_4Cl), NH_4+P, NO_3+NH_4+P.

Comparison of Laboratory and *in situ* Assays

To determine whether results from laboratory experiments were applicable to natural conditions, two attempts were made to run laboratory and *in situ* nutrient enrichment bioassays

concurrently. The first attempt, in March 1991, was not successful due to operational difficulties. Experiments conducted in July 1991 were successful. The *in situ* assays were conducted in 4-L transparent collapsible polyethylene containers. Water was collected at the Magnolia Park station in the evening and transported to a temporary field station located near the lake. Lake water in a polycarbonate carboy was continuously mixed on a stirring plate while 3 L of water were added per container. Treatments used were C, N, P and N+P (see Table 3.1). Two containers were used per treatment for a total of eight containers. The containers were attached to floating rack and submerged at 30-cm depth. Samples were collected every 24 h for 96 h; all sampling was conducted at dusk to prevent photo-shock. The laboratory part of the comparison was executed using the laboratory assay methods described previously.

Response Determination

Biomass response was measured as chlorophyll a (chl a) *in vivo* fluorescence (IVF) using a Turner Designs Fluorometer (Model 10). The use of IVF as a biomass indicator has been shown to be a rapid and reliable method of accessing relative differences between treatment groups originating from the same original population (Schelske et al. 1978, Elser and Kimmel 1986, Peeters and Peperzak 1990). Further confirmation of the strong relationship of IVF to extracted chl a and optically measured biovolume are presented in Chapter 4 of this report. Spectrophotometric analysis of 90% acetone extracted samples was done to determine initial and final

chl *a* (APHA 1989). Phytoplankton samples from the initial and the maximum response treatment for each assay were preserved in Lugol's solution for future analysis.

RESULTS AND DISCUSSION

Comparison of Nitrogen Sources

Two nutrient enrichment bioassays comparing phytoplankton community response to ammonium and nitrate nitrogen enrichments were run in December 1989 and January 1990. The results are presented as time plots in Fig. 3.1. The statistical ranking and grouping of treatments by assay date is presented in Table 3.2. Nitrate nitrogen stimulated significant increases in phytoplankton biomass, as indicated by increased IVF, over ammonium nitrogen for both assays.

In the December 1989 assay, ammonium nitrogen did not stimulate any increase in phytoplankton biomass from the initial level and produced significantly lower response compared to the nitrate treatment. The ammonium response was significantly greater than the phosphorus or control treatments in the December 1989 assay but not different from the control and phosphorus treatments in the January assay. There was no significant difference between the control and phosphorus treatments, both resulted in significant declines in phytoplankton biomass for both assays. There was no significant phosphorus interaction with either nitrogen form indicating no phosphorus deficiency in the phytoplankton community throughout either assay. There was a significant interaction of nitrate and ammonium in the December

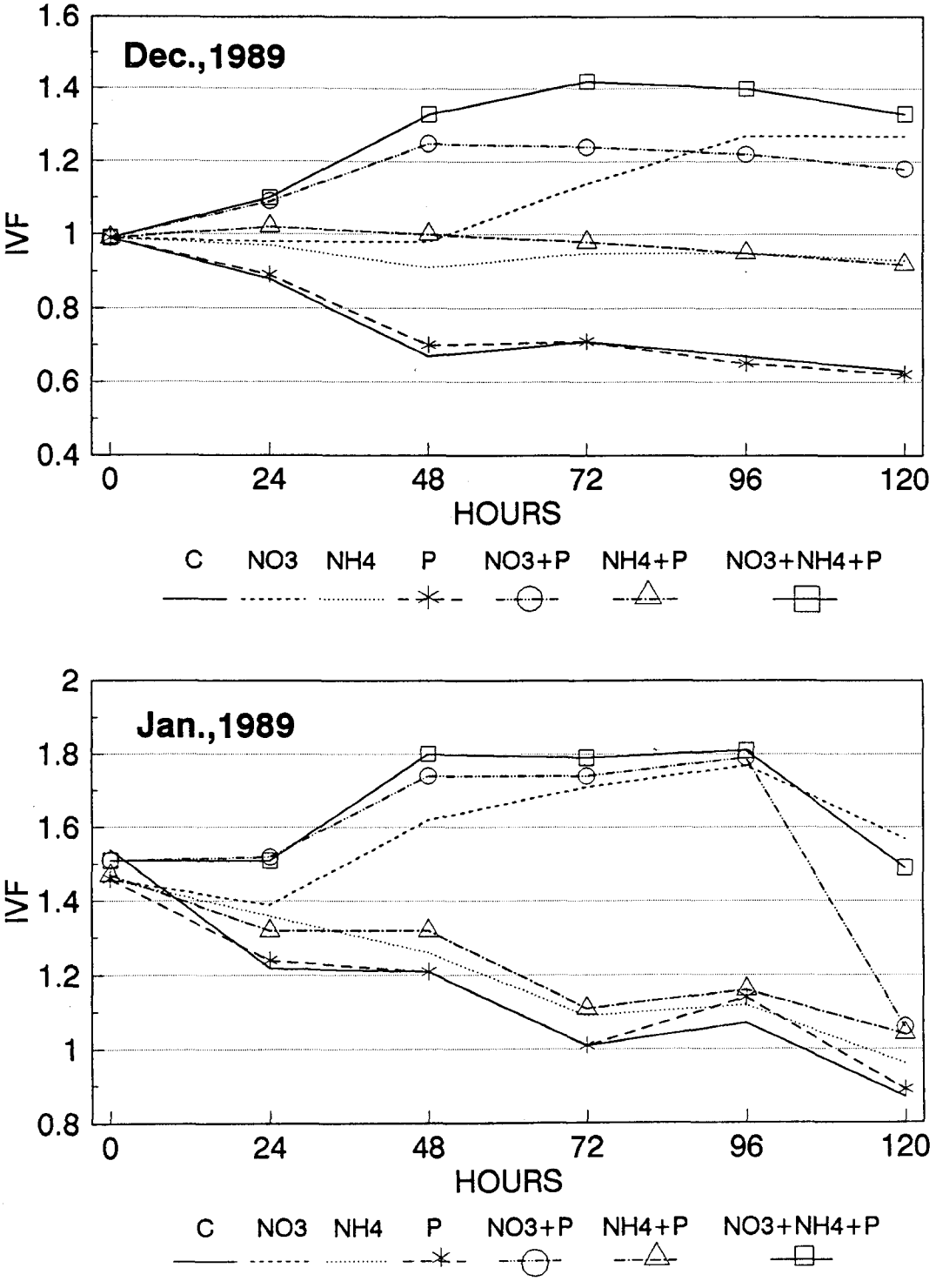

Figure 3.1. Comparison of nitrate and ammonium as nitrogen sources for nutrient enrichment bioassays. C=control. NO_3=400 µg N L^{-1}. NH_4=400 µg N L^{-1}. P=40 µg P L^{-1}. IVF=*in vivo* fluorescence used as relative biomass indicator for phyto-plankton. Hours = time in hours from start of assay. Assays were run as 1:1 dilution of whole lake water with filtered lake water.

Table. 3.2. Grouping and ranking of maximum treatment responses for each assay date. Treatments underlined together are not statistically different at $P<0.05$. N = 400 µg N L^{-1} as nitrate. C = control. NH_4 = 400 µg N L^{-1} as ammonium. P = 40 µg P L^{-1} as PO_4.

MONTH	TREATMENT RESPONSE
* 7 Dec 1989	$\underline{N+NH_4+P} > \underline{N+P\ N} > \underline{NH_4+P\ NH_4} > \underline{C\ P}$
* 25 Jan 1990	$\underline{N+NH_4+P\ N+P\ N} > \underline{NH_4+P\ NH_4\ P\ C}$
22 Feb 1990	$\underline{N+P} > \underline{N} > \underline{C\ P}$
* 22 Feb 1990	$\underline{N+P\ N} > \underline{C\ P}$
22 Mar 1990	$\underline{N+P} > \underline{N} > \underline{C\ P}$
* 19 Apr 1990	$\underline{N+P} > \underline{N} > \underline{C} > \underline{P}$
17 May 1990	$\underline{N+P\ N} > \underline{C\ P}$
14 Jun 1990	$\underline{N+P} > \underline{N} > \underline{C\ P}$
19 Jul 1990	$\underline{N+P\ N} > \underline{C\ P}$
* 23 Aug 1990	$\underline{N+P\ P} > \underline{N\ C}$
21 Sep 1990	$\underline{N+P\ N} > \underline{C\ P}$
18 Oct 1990	$\underline{N+P\ N} > \underline{C\ P}$
21 Nov 1990	$\underline{N+P\ N} > \underline{C\ P}$
18 Dec 1990	$\underline{N+P} > \underline{N} > \underline{C\ P}$
23 Jan 1991	$\underline{N+P\ N} > \underline{C\ P}$
28 Feb 1991	$\underline{N} > \underline{N+P} > \underline{C\ P}$
3 Apr 1991	$\underline{N+P\ N} > \underline{C} > \underline{P}$
19 Apr 1991	$\underline{N} > \underline{N+P} > \underline{C\ P}$
16 May 1991	$\underline{N+P\ N} > \underline{P} > \underline{C}$
27 Jun 1991	$\underline{N+P\ N} > \underline{C}$

* assays run with 1:1 dilution of whole lake water to filtered lake water

assay which produced significantly greater response than nitrate alone. This was not the case for the January assay where no nitrate-ammonium interaction was observed. Based on these results, nitrate

nitrogen was used as the nitrogen source in all subsequent experiments for NEB.

Dilution effect on NEB response

The effect of dilution on the growth response of the phytoplankton in an NEB environment was tested in the February, 1990 assay. Dilution with filtered lake water reduces the phytoplankton biomass and other particulates while maintaining the same concentration of dissolved constituents. It can be seen in Figure 3.2 that a 1:1 dilution resulted in a 50% reduction in net IVF at time zero. Net IVF was obtained by subtracting the background fluorescence of filtered water from the gross fluorescence of undiluted lake water. The background fluorescence is derived from submicron sized particles which pass through the filtration process as well as the soluble fluorescent compounds in the water.

The response trends for treatments were similar between the whole water and 1:1 dilution assay with nitrogen being the primary limiting nutrient. Phosphorus alone produced responses no different than the control treatments. Apart from the net difference of IVF indicated biomass resulting from dilution there were three differences in IVF responses between the whole water and dilution assays. First, there was a significantly greater maximum response in the whole water N+P interaction than the N treatment alone while in the dilution assay there was no significant N+P interaction. Second, the rate of change in IVF in the first 24 hours differed between the dilution assay and the whole water assay. In the dilution assay N and N+P treatments stimulated greater percent

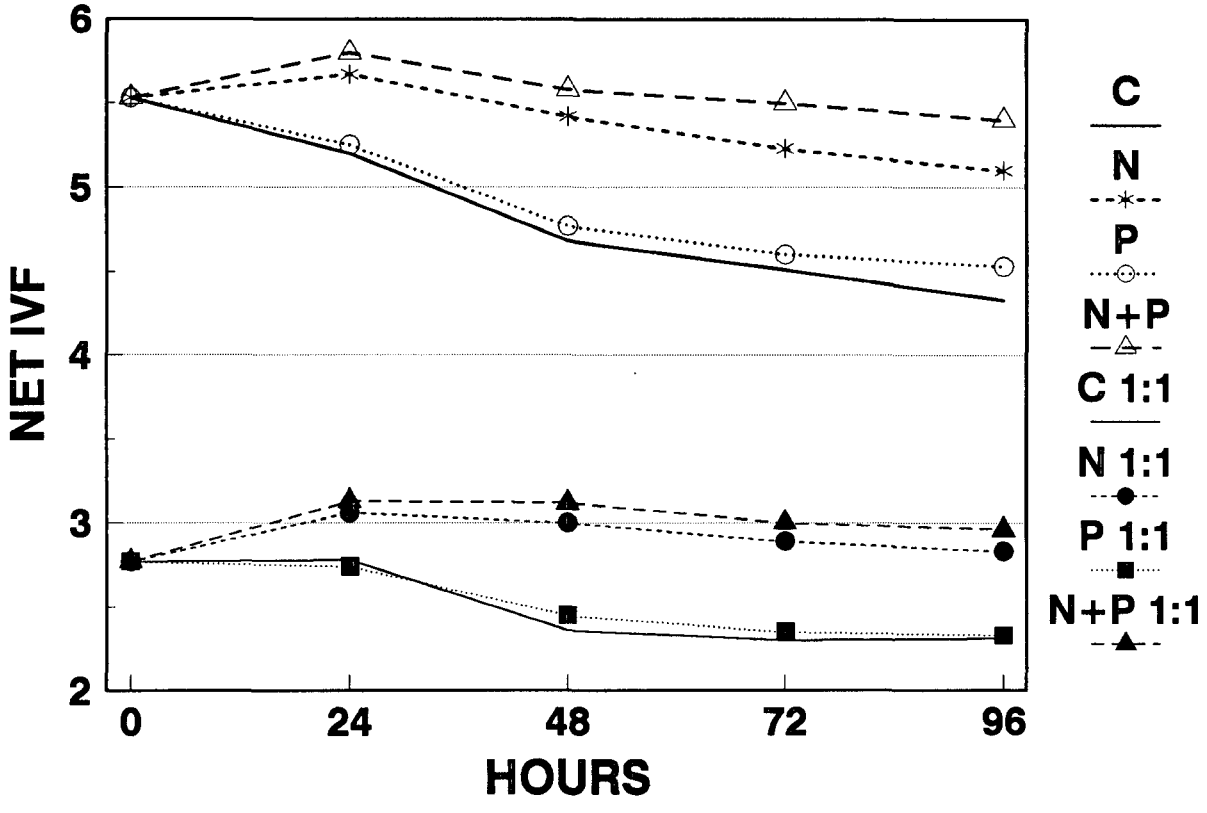

Figure 3.2. Comparison of dilution effect on phytoplankton biomass growth response in nutrient enrichment bioassay. Experiment conducted February 22-26, 1990. C=control. NO_3=400 µg N L^{-1}. P=40 µg P L^{-1}. Hours = time in hours from start of assay. Treatments with 1:1 notation indicate phytoplankton biomass was reduced 50% by diluting whole lake water 1:1 with 0.45 µm filtered lake water. Treatments without 1:1 notation indicate whole lake water treatments. IVF=*in vivo* fluorescence used as relative biomass indicator for phytoplankton.

increase than the same treatments in the whole water assay. Third, the percent decline of IVF in the control and P treatments was much lower for the dilution assay as compared to the whole water assay. The results of this comparison indicated that reduction of planktonic biomass by a 1:1 dilution might stimulate greater growth but the overall general trend of nutrient limitation does not change (A more detailed study of dilution effects is presented in Chapter 4 of this report). Based on this conclusion, all assays were run on whole lake water except those where physiological assays were also conducted.

Laboratory Assays

Nutrient enrichment bioassays were run in the laboratory monthly from December 1989 through July 1991 with the exception of March 1991 when no assay was run. The March assay was run on 3 April 1991 to maintain a nominal monthly sampling of phytoplankton community response to nutrient enrichment. Results are presented as time plots in Figs. 3.1-3.7. Statistical ranking and grouping of treatments by assay date are presented in Table 3.2.

Nitrogen was either the primary or co-factor in limiting phytoplankton growth, as determined by stimulating the greatest response, for all but one assay date. The one exception was the August 1990 assay when phosphorus was the primary limiting factor. Nitrogen and phosphorus were co-factors in limiting algal growth in only 5 of the 20 experiments that were conducted.

The five assays in which the N+P treatment produced a significantly greater response than N alone was interpreted as a secondary P limitation brought on by an increase in phytoplankton biomass

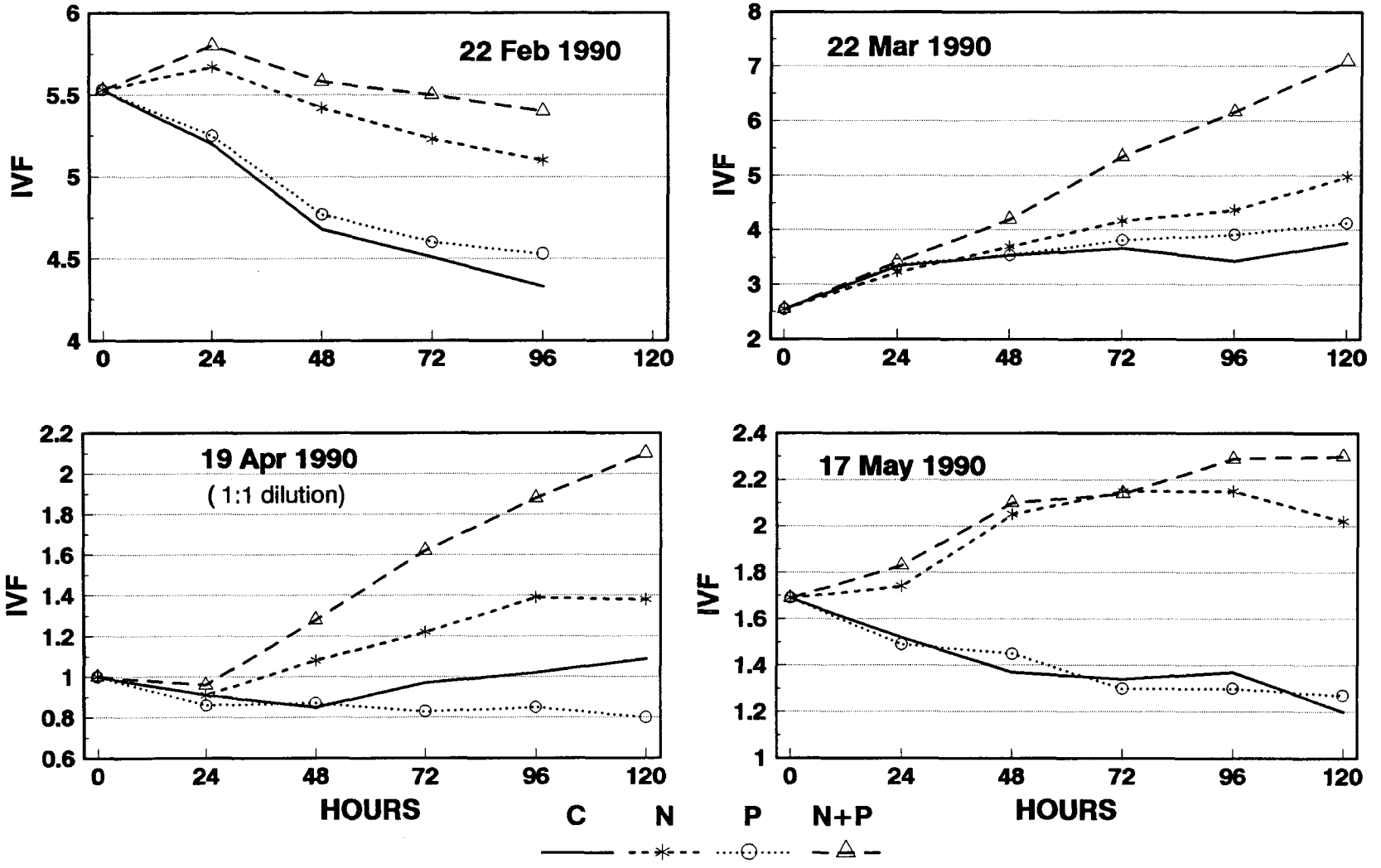

Figure 3.3. Results of nutrient enrichment bioassays run on water collected off Magnolia Park at Lake Apopka, Florida for the months of Feb.-May, 1990. C=control. NO_3=400 µg N L^{-1}. P=40 µg P L^{-1}. IVF=*in vivo* fluorescence used as relative biomass indicator for phytoplankton. Hours = time in hours from start of assay.

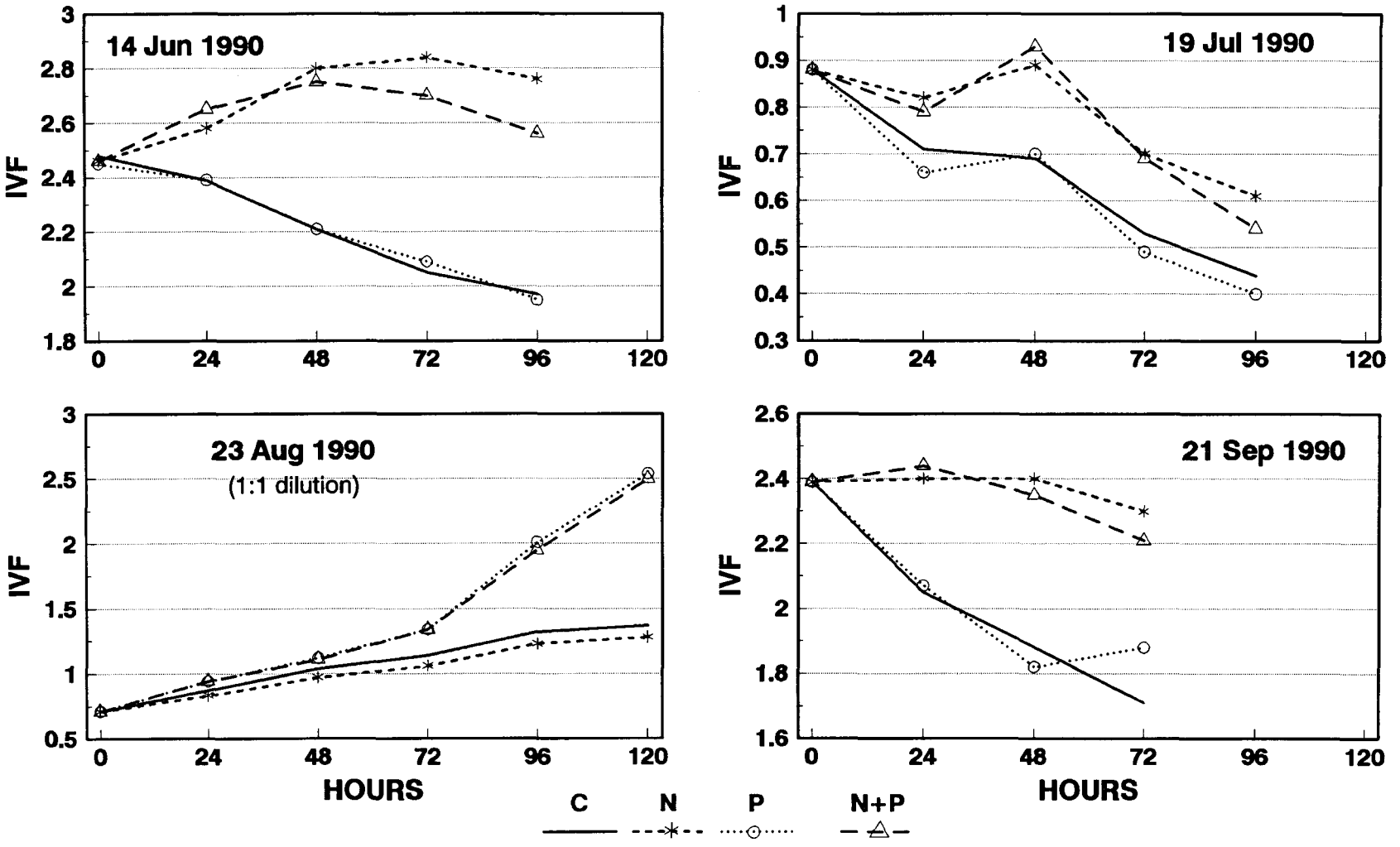

Figure 3.4. Results of nutrient enrichment bioassays run on water collected off Magnolia Park at Lake Apopka, Florida for the months of June-Sept., 1990. C=control. NO_3=400 µg N L^{-1}. P=40 µg P L^{-1}. IVF=*in vivo* fluorescence used as relative biomass indicator for phytoplankton. Hours = time in hours from start of assay.

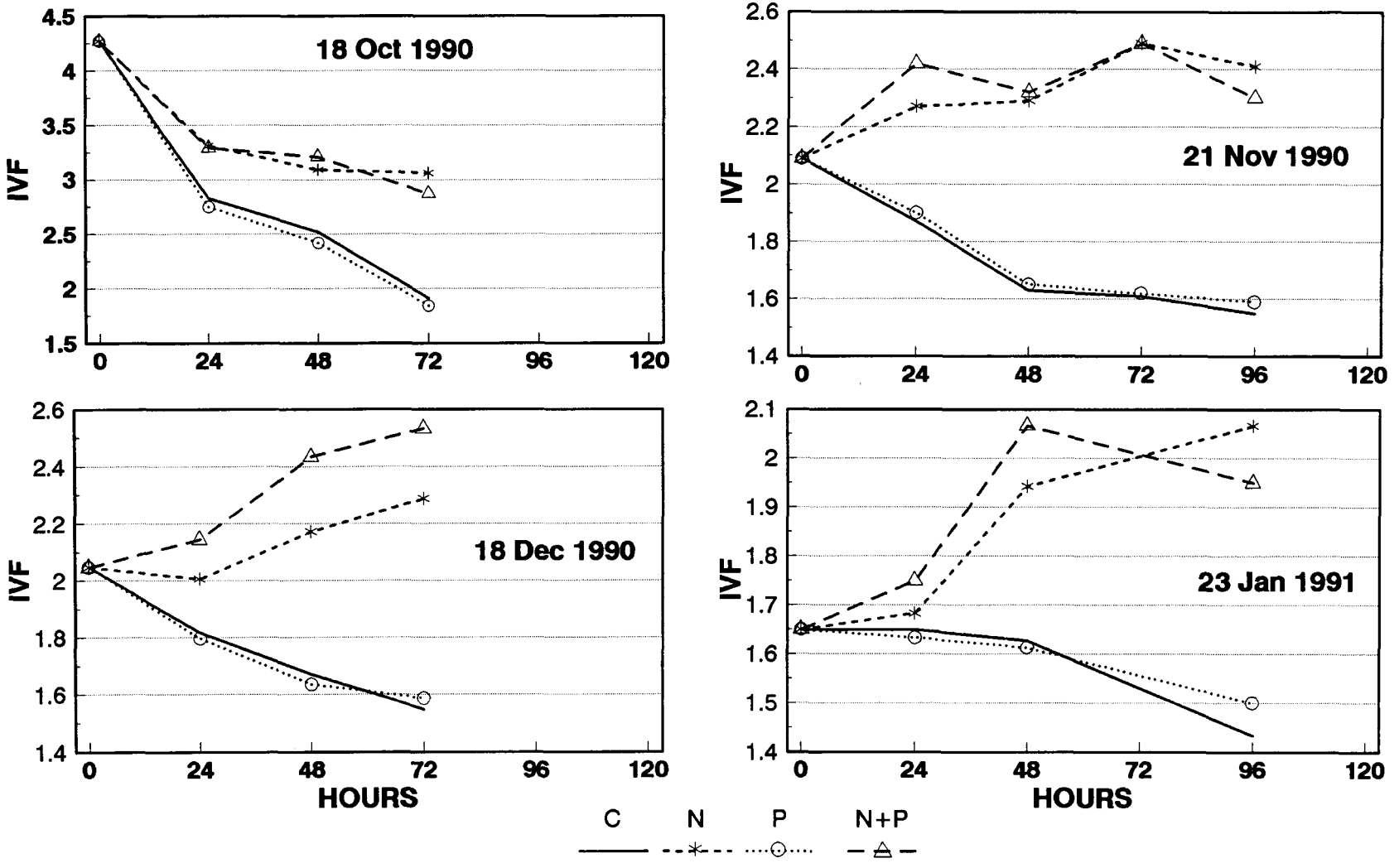

Figure 3.5. Results of nutrient enrichment bioassays run on water collected off Magnolia Park at Lake Apopka, Florida for the months of Oct. 1990-Jan. 1991. C=control. NO_3=400 µg N L^{-1}. P=40 µg P L^{-1}. IVF=*in vivo* fluorescence used as relative biomass indicator for phytoplankton. Hours = time in hours from start of assay.

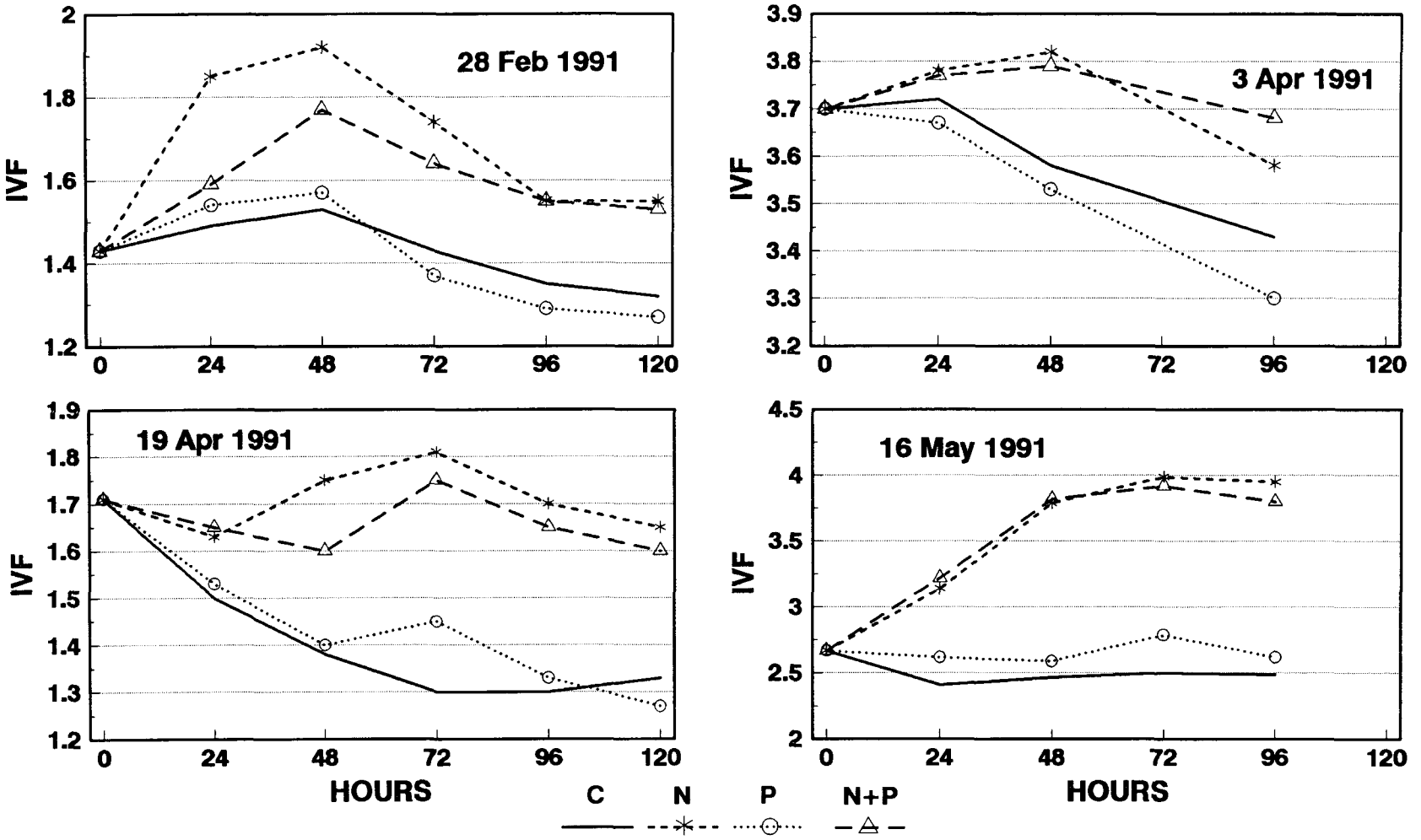

Figure 3.6. Results of nutrient enrichment bioassays run on water collected off Magnolia Park at Lake Apopka, Florida for the months of Feb.-May 1991. C=control. NO_3=400 µg N L^{-1}. P=40 µg P L^{-1}. IVF=*in vivo* fluorescence used as relative biomass indicator for phytoplankton. Hours = time in hours from start of assay.

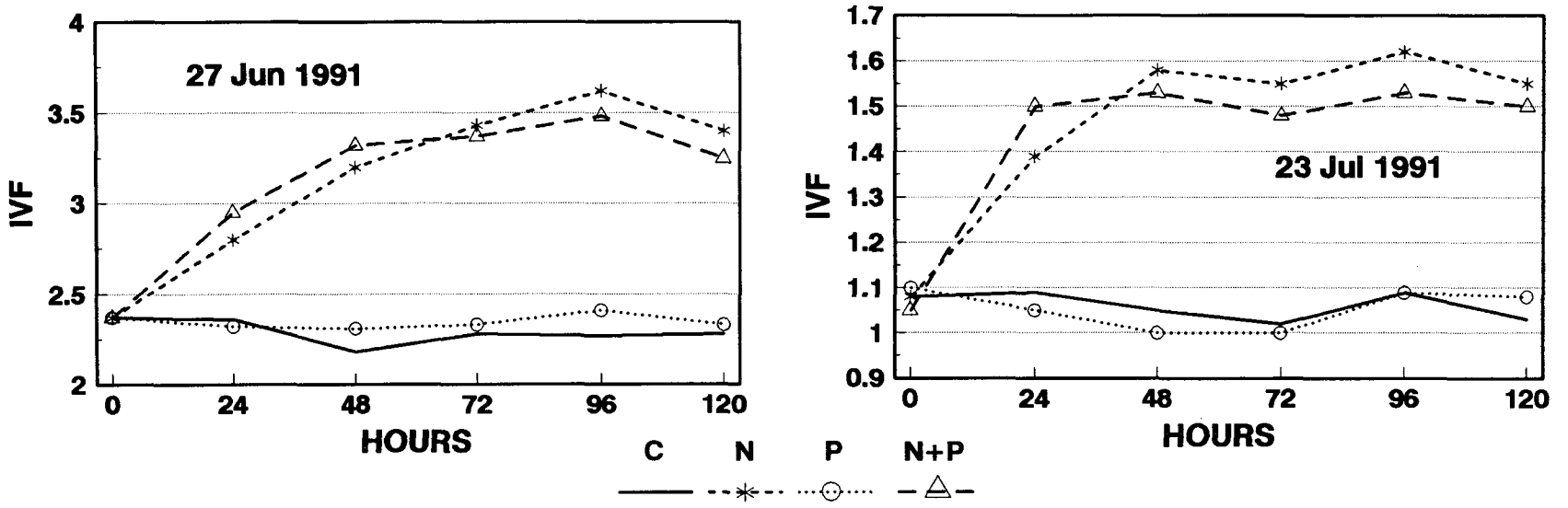

Figure 3.7. Results of nutrient enrichment bioassays run on water collected off Magnolia Park at Lake Apopka, Florida for the months of June-July 1991. C=control. NO_3=400 µg N L^{-1}. P=40 µg P L^{-1}. IVF=*in vivo* fluorescence used as relative biomass indicator for phytoplankton. Hours = time in hours from start of assay.

after elimination of the primary N limitation. N was determined the primary limiting nutrient since the P treatment response was not different from that of the control. These results indicate that P availability, either in the form of soluble reactive phosphorus or as polyphosphate storage products, was sufficiently low to become secondarily limiting when phytoplankton biomass was increased with nitrogen enrichment. The majority of assays, however, showed no significant difference between control and P treatments and no significant N+P interaction (no significant difference between the N and N+P treatments). These assay results show that increases in phytoplankton biomass were not limited by phosphorus availability and that supplies of phosphorus were in excess compared to the demands for phytoplankton growth, either in the lake or under conditions of nutrient-enhanced growth in the laboratory.

Besides the predominance of nitrogen limitation only a weak temporal pattern may be discerned. Four of five experiments in which there was a significant N+P interaction occurred from February to June 1990 (Table 3.2). During these five months, the N+P interaction was not significant only in May. The fifth experiment in which there was a significant N+P interaction was 18 December 1990. These results suggest that phosphorus availability was more limited during these experiments than during other experiments conducted during the study except for the August, 1990 assay. Since this pattern did not repeat in the same period the following year, 1991, there is not sufficient evidence from this study to conclude any relationship between phytoplankton nutrient status and month or season of the year.

There were five assay dates (March 22, April 19, August 23, 1990 and February 28, April 3, 1991) when the control treatment maximum IVF response was greater than the initial IVF value. It is concluded that the phytoplankton from these dates were not immediately nutrient limited at the time of sampling but, after some growth, became limited by the availability of nitrogen or phosphorus. The fact that four of these five dates occur in the early spring may or may not be indicative of a seasonal pattern. The August 23, 1990 assay is anomalous in that it is the only month in which no initial nutrient limitation was followed by a strong phosphorus stimulated growth. This is also the only assay in which the nitrogen supply was adequate to sustain phytoplankton growth after removal of phosphorus limitation.

The N and N+P treatments stimulated general increases in IVF compared to the initial IVF in 12 of 20 assays while in five of 20 assays N additions resulted in no change or generally negative IVF compared to the initial. In 14 of 20 assays, IVF in the C and P treatments declined significantly from the start to the end of the assay. Extracted chlorophyll values from the start and end of the assays confirmed the IVF results. It is not known if this decline in phytoplankton chlorophyll is indicative of actual phytoplankton biomass loss from cell death or grazing or if chlorophyll pigment loss is due to chlorosis or to other process associated with nutrient deficiency stress.

Comparison of Laboratory and *in situ* Assays

Results from the *in situ* and laboratory nutrient enrichment bioassay comparison are illustrated in Fig. 3.8. Responses to the nutrient enrichment treatments were similar for both locations. Nitrogen was the primary limiting nutrient tested with no significant N+P interaction. Control and P treatments were not statistically different for both locations and all times except for the 96-h IVF reading of the *in situ* assay in which the control treatment declined and the P treatment remained the same as the day before. The IVF response reached a maximum plateau by 48 h after the start of the assay for both locations and continued for the duration of the experiments. The most prominent difference between the two assay locations was the change in IVF response at 24 h. The IVF for the N and N+P treatments of the laboratory assay increased at twice the rate of the IVF response for the same treatments of the *in situ* assay. The IVF of the laboratory control and P treatments remained near the initial value throughout the assay while the *in situ* control and P treatments displayed a 16% decline in IVF by 24 h. The difference between the IVF means of the N containing treatments and the IVF means of the C and P treatments for all sample times after 24 h ranged from 30 to 36% for the laboratory assay and 29 to 33% for the *in situ* assay.

The primary differences between the two test conditions were container size and light. In the laboratory, 300 ml of lake water were added to 500-ml Erlenmeyer flasks whereas 4-L containers and 3 L of lake water were used in the *in situ* experiments. Comparison of the PAR flux for the two experimental conditions shows the *in*

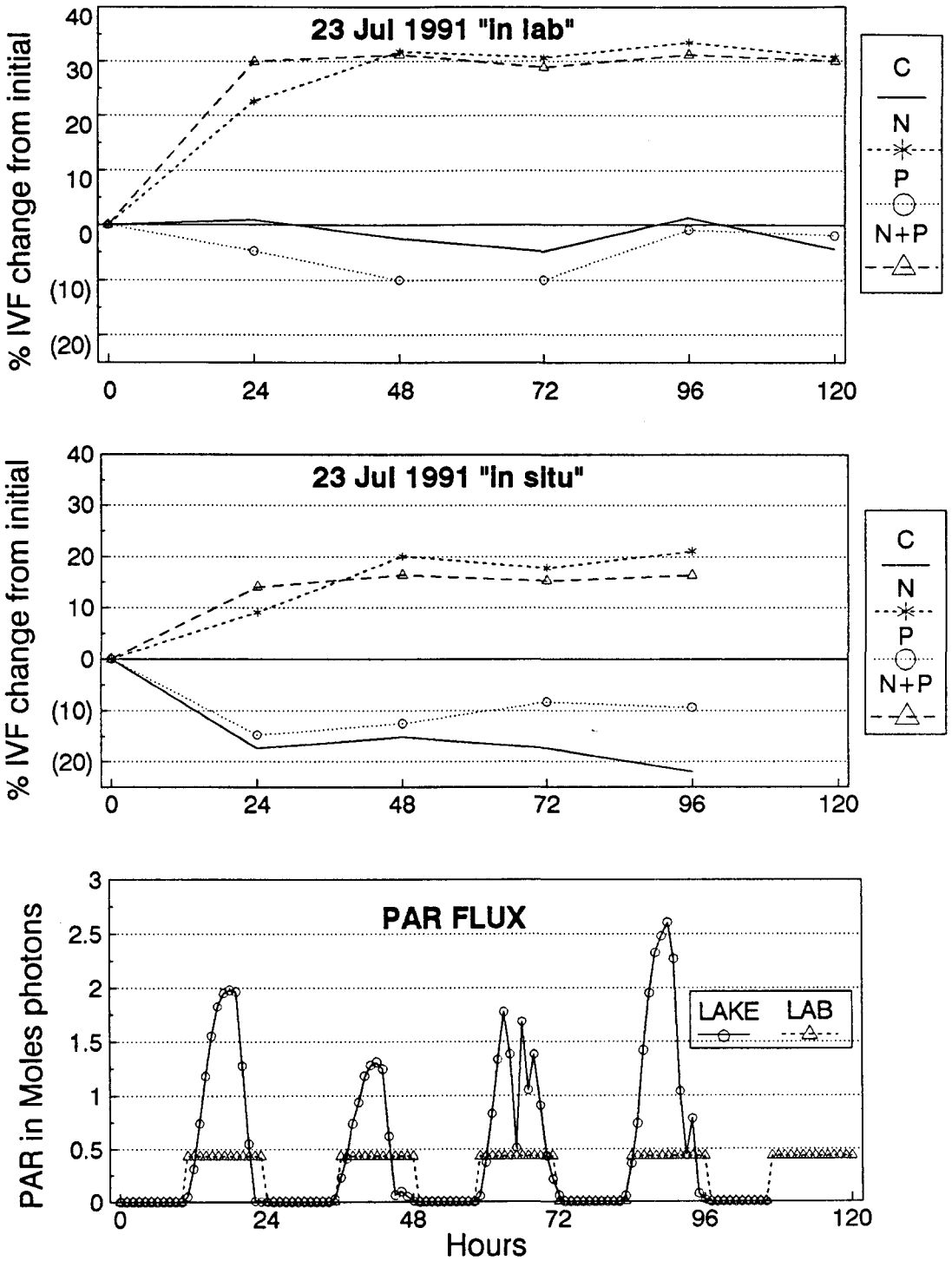

Figure 3.8. Comparison of results from nutrient enrichment bioassays run concurrently *in situ* and in laboratory. PAR=photosynthetic active radiation for 30 cm water depth in Lake Apopka, Florida and in a laboratory incubator. C=control treatment. N=400 µg N L^{-1} as NO_3. P=40 µg P L-1 as PO_4. IVF=*in vivo* fluorescence used as relative biomass indicator.

situ assay received approximately twice the PAR flux as the laboratory assay (Fig. 3.8). It should be noted that the *in situ* PAR flux does not take into account loss of light due to the containers which would reduce the actual PAR to the phytoplankton by approximately 20%. The comparison also illustrates the variability of *in situ* light as a function of time while laboratory light is either at a constant level or absent.

The relative difference of response between treatments was essentially the same for the two experimental conditions. The qualitative ranking of nutrient importance also did not differ. Based on these results, we conclude there are many logistical advantages to laboratory-based assays and no strongly compelling justification for the added logistical complexity and cost to do *in situ* NEB studies. This type of a conclusion has been found by other investigators who have conducted comparisons (see Schelske 1984).

CONCLUSIONS

The source of nitrogen is an important factor in determining the growth response of Lake Apopka phytoplankton communities. Nitrate nitrogen stimulated a greater growth response in nitrogen-limited conditions than ammonium nitrogen of equal nitrogen content.

Nitrogen was found to be the predominant limiting nutrient to growth in Lake Apopka phytoplankton. Nutrient enrichment bioassays showed primary nitrogen limitation or co-limitation in 19 of 20 monthly assays.

Conversely, indications of a surplus of available forms of phosphorus that could support phytoplankton growth were found in 19 of 20 monthly assays.

No general temporal patterns in phytoplankton nutrient status could be discerned.

Reduction of plankton biomass by a 1:1 dilution of whole lake water with filtered lake water enhanced phytoplankton growth but did not change the general pattern of nutrient limitation.

There was no relative difference between nutrient treatments for nutrient enrichment bioassays run *in situ* at Lake Apopka and the same assays run under laboratory conditions.

The experimental approach employed was based on measuring a direct response of phytoplankton to nutrient enrichment with either P or N or combined N+P. Results we obtained are based on responses to enrichment with two major nutrients. It is possible that limited supplies of other nutrients or other factors may also be important in controlling standing crops. Our experimental design does address two of the most important major nutrients and the only nutrient, P, whose supplies can be readily controlled.

We conclude that N is the primary limiting nutrient because the water of Lake Apopka generally contains large supplies of phosphorus that can be utilized for phytoplankton growth when the water is enriched with N. It also must be concluded that N limitation in Lake Apopka is the result of excessive P loading to the lake and that to reduce standing crops of algae P supplies must be reduced so that P becomes the primary limiting nutrient.

Results from nutrient enrichment bioassays and other evidence clearly identify excess supplies of P as the primary nutrient factor responsible for hypereutrophic conditions in Lake Apopka.

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Chapter 4

NUTRIENT LIMITATION OF LAKE APOPKA PHYTOPLANKTON: APPLICATION OF DILUTION EXPERIMENTS

INTRODUCTION

Although the relationship between water column nutrient concentrations (i.e., total phosphorus and total nitrogen) and phytoplankton biomass provides a useful framework for estimating the response of lakes to nutrient enrichment (e.g., Dillon and Rigler 1974; Jones and Bachmann 1976), this relationship is correlative and offers limited information on underlying causal factors. Ambient nutrient concentrations are not always the best predictors of nutrient status of the phytoplankton themselves, as deficient cells can take up nutrients in excess of their immediate growth requirements (e.g, Healy 1979; Lean and Pick 1981). However, several tests have been used to study the problem of nutrient limitation of phytoplankton, most notably are bioassays involving the enclosure of natural phytoplankton assemblages (e.g, Schelske 1984), whole lake fertilization (e.g., Schindler and Fee 1974), or the use of physiological indicators of nutrient deficiency (Vincent 1981a, 1981b, Lean et al. 1983; Dodds and Priscu 1990). Despite this, it can be difficult to identify the nutrients that limit phytoplankton growth in hypereutrophic waters due to persistent nutrient excesses (Paerl and Bowles 1987), thus alternative approaches may be required for assessing nutrient limitation.

Along these lines, there is debate as to which primary factors mediate phytoplankton dynamics in tropical and subtropical lakes (Kratzer and Brezonik 1981, 1982; Osgood 1982; Canfield et al. 1989), as comparatively little research has been conducted on these systems compared with temperate systems (Payne 1986; Lewis 1987). The evidence for nutrient limitation in subtropical lakes may be circumstantial, as few controlled experiments have actually been conducted to test this hypothesis (Canfield and Hoyer 1988; Schelske 1989). Relationships between ambient nutrient concentration (i.e., total phosphorus and total nitrogen) and phytoplankton biomass across a suite of Florida lakes support the idea that the phytoplankton biomass in these lakes is regulated by nutrients (Canfield 1983). However, additional work has spawned a debate as to whether phytoplankton abundance is under strict nutrient control, given the high levels of biomass supported in some of these lakes (Canfield and Hoyer 1988; Canfield et al. 1989; Duarte et al. 1990; Agusti et al. 1991).

In this report, we evaluated an approach for assessing the nutrient status of phytoplankton in the hypereutrophic, subtropical Lake Apopka located in central Florida. This approach involves using nutrient enrichment bioassay coupled with serial dilution of lake water (Paerl and Bowles 1987). The approach seems feasible, because most of the readily available N and P have been incorporated into phytoplankton biomass in Lake Apopka, and thus dilution of phytoplankton results correspondingly in the dilution of both N and P pools. While dilution experiments can suffer from experimental artifacts (e.g., Stockner et al. 1990; Li 1990), most of the critical

assumptions of the technique can be evaluated (e.g., Li 1986). Conducting nutrient enrichment bioassays on various dilutions of lake water is potentially useful because it can serve as experimental evaluation of the current management scheme being tested to improve water quality in Lake Apopka; particulate materials (phytoplankton and associated nutrients) in the water column of the lake are being removed via an artificial wetland (Lowe et al. 1992).

MATERIALS AND METHODS

Ambient Lake Conditions

Water samples for experimentation were collected with a 5-Liter PVC Niskin bottle or a submersible pump at a central station in the lake. Studies have shown that cells are not damaged when water is collected with a submersible pump. In addition, the growth of phytoplankton in water collected by pumping is not different compared with water collected with a Niskin bottle (H.J. Carrick, unpubl. data).

On each sampling date, temperature and oxygen profiles were measured with a YSI meter. Light penetration was determined with a Secchi disk (20 cm) and underwater scalar irradiance (PAR) was assessed with a LiCor LI-1000 meter fitted with upwelling and downwelling submersible sensors. Incident irradiance (PAR) was evaluated continuously with a second LiCor LI-1000 data logger (hourly integrated values, mol).

The occurrence of N and P in three major chemical phases was determined by filtration. Concentrations of dissolved, inorganic forms of P (SRP) and N (DIN, as NO_3-N + NO_2-N) were measured on water passing a 0.3 μm filter (0.3 μm Whatman 2000 EPM), while dissolved organic N and P were determined by measuring total N and P in this same filtered lake water fraction (<0.3 μm). N and P retained on the filters (>0.3 μm) was assumed to be in the particulate phase, while raw lake water was digested to determine concentrations of total N and P. All nutrient concentrations were measured on a Technicon II Autoanalyzer using standard colorimetric reactions (Davis and Simmons 1979).

Nutrient Dilution Experiments

The growth response of phytoplankton to nitrogen and phosphorus was evaluated with three types of laboratory nutrient-dilution experiments (Table 4.1). Two types involve the serial dilution of particulate material by mixing raw lake water with either distilled water (experiment on 9 January) or filtered lake water (24 January, 6 February, and 4 November) similar to experiments designed to estimate microzooplankton grazing (Landry and Hassett 1982). In the third type of experiment, raw lake water was diluted in a stepwise manner with a major ion solution (MIS) containing only major ions dissolved in distilled water and deficient of N and P (Table 4.2). The addition of MIS as a diluent reduces ambient soluble nutrients present in lake water while attempting to replicate the ionic composition of Lake Apopka water (Brezonik et al. 1978). To all dilution treatments, N and P were added both

Table 4.1. Experimental conditions for nutrient-dilution experiments conducted on five dates in 1991. Where DH_2O denotes deionized water, FLW denotes filtered lake water, and MIS denotes major ionic solution.

Date (1991)	Depth (m)	Diluent Used	% Diluent Additions	Nutrient Additions	Light ($\mu E\ m^{-2}\ s^{-1}$)	Temp. (°C)
09 Jan	0.3	DH_2O	60, 30, 0	N, P	150	21.5
23 Jan	0.3	FLW	60, 30, 0	N, P, N+P	150	15.0
06 Feb	0.3	FLW	60, 30, 0	N, P, N+P	150	19.5
26 Jun*	0.3	MIS+FLW	75, 50, 25, 0	N, P, N+P	150	30.0
04 Nov	0.3	FLW	95, 90, 80, 60, 0	N, P, N+P	150	19.0
	0.3	MIS	95, 90, 80, 60, 0	N, P, N+P	150	19.0

*Constant phytoplankton biomass maintained among treatments.

Table 4.2. Chemical composition and final concentration of constituent in major ionic solution (MIS) used in nutrient-dilution experiments.

Major Ion	Chemical Added	Final Concentration (as mg $Liter^{-1}$ of ion)
Ca^{2+}	$CaCO_3$	35.0
Cl^-	HCl	35.0
CO_3^-	$CaCO_3$, $MgCO_3$	121.2
K^+	K_2SO_4	7.3
Mg^{2+}	$MgCO_3$	20.0
Na^+	$NaHCO_3$	15.2
SO_4^{2-}	K_2SO_4	9.0

singly and in combination to duplicate bottles (Paerl and Bowles 1987).

In all experiments, acid washed bottles (500-ml Erlenmeyer flasks) received varying mixtures of raw lake water and one of three diluents (final volume 350 ml). Filtered lake water (FLW) was prepared by passing raw lake water through a 0.2 μm filter capsule (Gelman maxi-cap) under low pressure (< 10 mm Hg), while distilled water used on 9 January was prepared with a reverse osmosis system.

The experiment conducted on 26 June was constructed to maintain constant phytoplankton biomass among treatments. This was achieved by placing varying amounts of MIS, filtered lake water (FLW), and raw lake water (RLW) such that RLW constituted 25% of the mixture in all treatments (constant biomass). Of the 32 bottles used in this experiment, 8 bottles contained 25% MIS as a percent of the final volume in the flask, 8 had 50% MIS added, 8 had 75% MIS added, and eight contained no MIS. The other five experiments were constructed similarly; however, phytoplankton biomass was allowed to vary as a function of the amount of diluent added to raw lake water.

Duplicate bottles from each of the dilution treatments received factorial enrichment with N and P, whereby two bottles were augmented with PO_4^{3-} (40 μg P $Liter^{-1}$), two were enriched with NO_3^- (400 μg N $Liter^{-1}$), two bottles received both N+P, and two bottles did not receive nutrients and served as controls. This was true for all experiments except that conducted on 9 January in which N and P were added singly without an N+P treatment.

Experimental bottles were incubated for 5 d in a walk-in growth chamber or in Percival incubators, where light and temperature conditions were maintained at levels similar to those in the lake at the time of collection (Table 4.1).

In all experiments, bottles were sampled daily (every 24 h) at approximately the same time for *in vivo* fluorescence. Once collected, samples were allowed to dark adapt for 20 minutes to provide a better measure of phytoplankton biomass, and were then read with a Turner Designs Fluorometer (Vincent 1979). Extracted chlorophyll a concentrations at the start and termination of the experiments conducted in June were determined fluorometrically using an acetone:DMSO (50:50) extraction procedure that does not employ grinding (Shoaf and Lium 1976).

The time-dependent change in *in vivo* fluorescence readings for the phytoplankton in each of the bottles was determined by calculating exponential growth rates (r) using the following:

$$r = \ln (N_t / N_o) / t$$

where N_t is the maximum *in vivo* fluorescence achieved in the experiment, N_o is the initial *in vivo* fluorescence allowing 24 h for adaptation if necessary, and t is the duration of time between initial and final samplings. In addition, hot-water extractable phosphorus (HEP) was measured in experiments on 4 November. HEP was determined by filtering water onto membranes (0.45µm, Gelman Supor); the filters were placed in tubes and autoclaved for 60 min which hydrolyzes polyphosphate stored in cells (Fitzgerald and Nelson 1966). The soluble reactive P hydrolyzed from cells was then

measured on the samples using the methods outlined above. Values referred to hereafter as P-storage rates (d^{-1}) were derived using an exponential growth equation:

$$\text{P-storage} = \ln (HEP_t / HEP_o)/ t$$

where HEP_t is the cellular HEP concentration at time t, HEP_o is the initial HEP concentration, and t is the duration of time between initial and final samplings.

Estimates of phytoplankton growth were then analyzed using a two-way analysis of variance (ANOVA), with nutrient treatments and the level of dilution considered fixed factors. When two-way ANOVA yielded significant interaction terms, one-way ANOVA was utilized where dilution was considered a blocked factor and nutrient treatments were fixed. All the data met assumptions of homoscedasticity and Tukey's test ($P < 0.05$) was used to test for pair-wise differences among the fixed factors. All statistical analyses were performed using SYSTAT (Version 4.0, 1991).

Evaluating Assumptions of Dilution Bioassays

The critical assumptions of dilution techniques have recently come into serious question (Li 1990). While many of these assumptions can be readily tested, potential problems concerning passage of cells through leaky filters and contamination of diluents resulting from cell breakage require careful checks (Stockner et al. 1990). The major criticisms of this technique are four; (1) artifacts due to containing phytoplankton in bottles, (2) filter-induced enrichment of nutrients, (3) occurrence of cells in the diluent due to

passage through leaky filters, and (4) contamination of the diluent after preparation.

We designed experiments to test these four assumptions, thus aiding in the interpretation of results. To evaluate the effects of containment on Lake Apopka phytoplankton, water was collected from 0.3 m depth, mixed, and dispensed into replicate vessels of varying volume (500, 700, 1000, and 2000 ml containers). All eight vessels were incubated at ambient light and temperature and subsampled at 0, 24, 48, 120, 140, and 160 h for extracted chlorophyll a. Second, nutrient contamination of diluents was assessed by determining concentrations of N and P in the three diluents (distilled H_2O, MIS, and FLW) as described above. Third, the occurrence of cells in the diluents and contamination related to sampling (air-borne or inadvertent inoculation of flasks) was evaluated on 4 November by incubating replicate flasks containing MIS, FLW, and FLW with added N and P. *In vivo* fluorescence was measured daily and initial and final subsamples were prepared for epifluorescence microscopy to determine if autotrophic cells were present and grew in the diluents (Caron 1983). Fourth, on 4 November the grazing pressure on phytoplankton by organisms <80 µm in size was evaluated by screening lake water through an 80 µm plankton net; this inoculum was diluted with FLW at five levels (0, 60, 80, 90, 95 % added FLW) and incubated as described above. *In vivo* fluorescence was determined daily from each flask and growth rates were calculated. These rates were regressed onto the increasing percent of <80 µm lake water.

RESULTS

Ambient Nutrient Pools

Total nitrogen (N) and phosphorus (P) are present in high concentrations in the surface waters of Lake Apopka (Table 4.3). A large percentage of both nutrients occurs in the particulate phase (48.0 and 85.6%, respectively). Dissolved inorganic N and P occur in very low (marginally detectable) concentrations and constitute very little of respective N and P pools. The remainder of N and P is present in a dissolved organic fraction (51.9 and 12.6%, respectively).

Nutrient-Dilution Experiments

Changes in phytoplankton growth rates among nutrient treatments were fairly consistent (Figs. 4.1, 4.2, 4.3, 4.4, 4.5 and 4.6). In five of six experiments, growth was enhanced by N addition alone while P addition had little or no effect. However, both experiments conducted on 4 November produced different results (Figs. 4.5 and 4.6); phytoplankton growth in N+P treatments was significantly higher than that following N addition, while both of these treatments yielded higher growth than P and control treatments.

Treatment-level differences in phytoplankton growth among dilution levels were observed in five of the six experiments (Two-way ANOVA, Table 4.4). Pairwise comparisons ($p < 0.05$) among dilution treatments indicated that bottles receiving the greatest dilution of MIS used in two of the six experiments yielded significantly higher growth. It appears, however, that pairwise

Table 4.3. Distribution of phosphorus and nitrogen into soluble reactive P (SRP) or dissolved inorganic N (DIN), dissolved organic P (DOP) or dissolved organic N (DON), particulate P (PP) or particulate N (PN) components for surface water collected from Lake Apopka on six dates in 1991. Values for total N and P are concentrations (µg Liter^{-1}).

	Phosphorus				Nitrogen			
Date	SRP (%)	DOP (%)	PP (%)	Total (µg L^{-1})	DIN (%)	DON (%)	PN (%)	Total (µg L^{-1})
23 Jul	4.4	19.6	76.0	109.5	0.1	58.7	41.2	3237
24 Jul	3.5	10.4	87.6	137.7	0.1	49.1	50.9	3682
25 Jul	2.6	8.2	90.5	187.7	0.1	48.9	51.1	3682
26 Jul	3.5	11.9	86.6	139.3	0.1	51.8	48.1	3627
27 Jul	4.3	12.6	85.7	111.9	0.1	51.1	48.9	3439
16 Sep	0.1	12.8	87.1	110.7	0.1	.	.	2968
Average	3.1	12.6	85.6	132.8	0.1	51.9	48.0	3439.2
SD	1.6	3.8	5.0	30.2	0.0	4.0	4.0	288.5

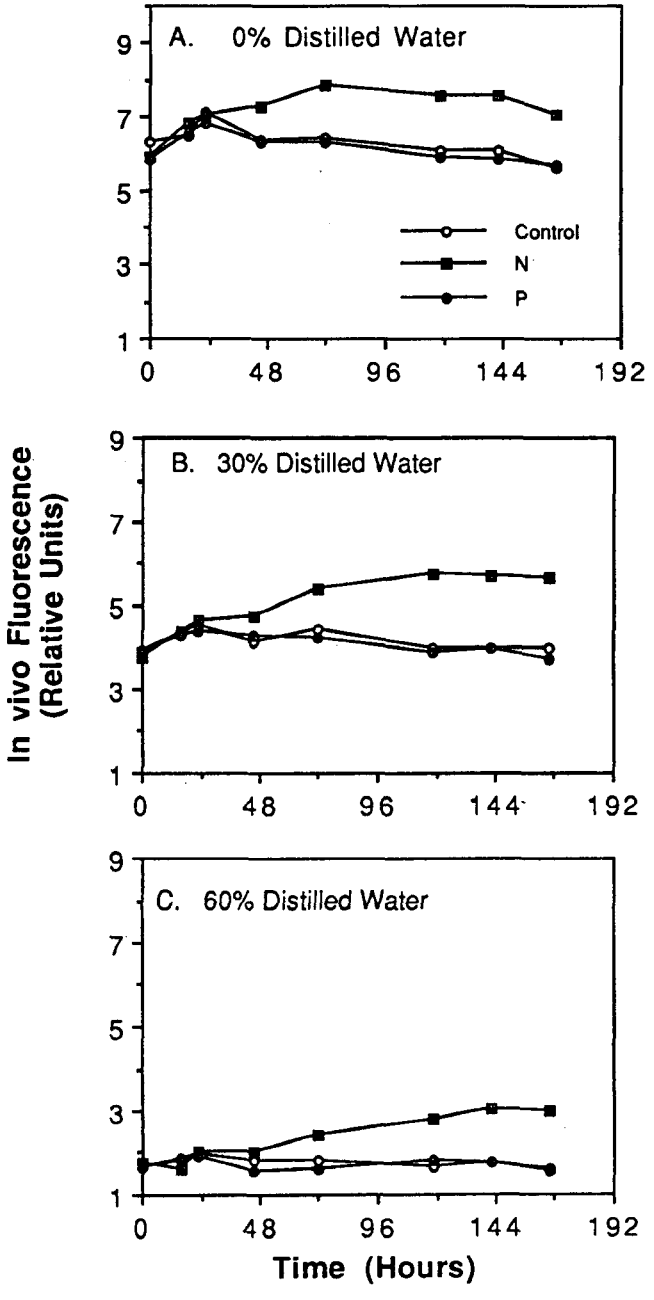

Figure 4.1. Average *in vivo* fluorescence values for Lake Apopka phytoplankton incubated on 9 Jan among three nutrient treatments and three levels of dilution.

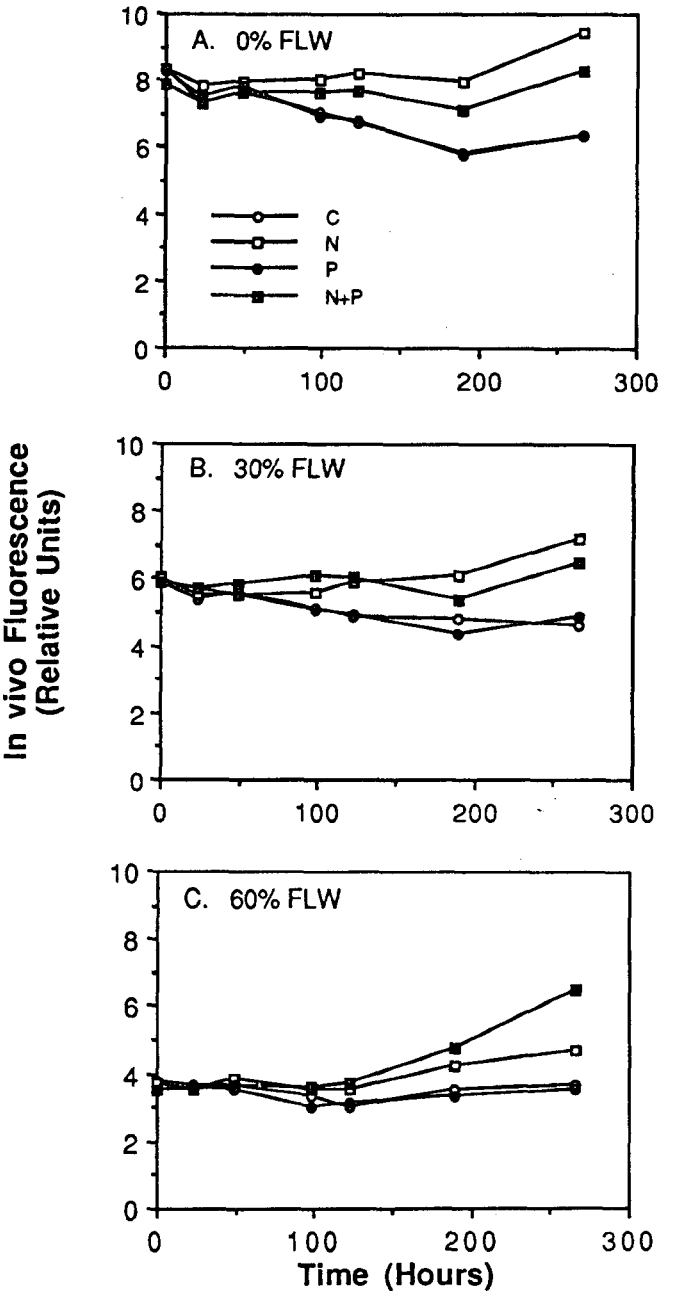

Figure 4.2. Average *in vivo* fluorescence values for Lake Apopka phytoplankton incubated on 24 Jan among four nutrient treatments and three levels of dilution. FLW = Filtered lakewater.

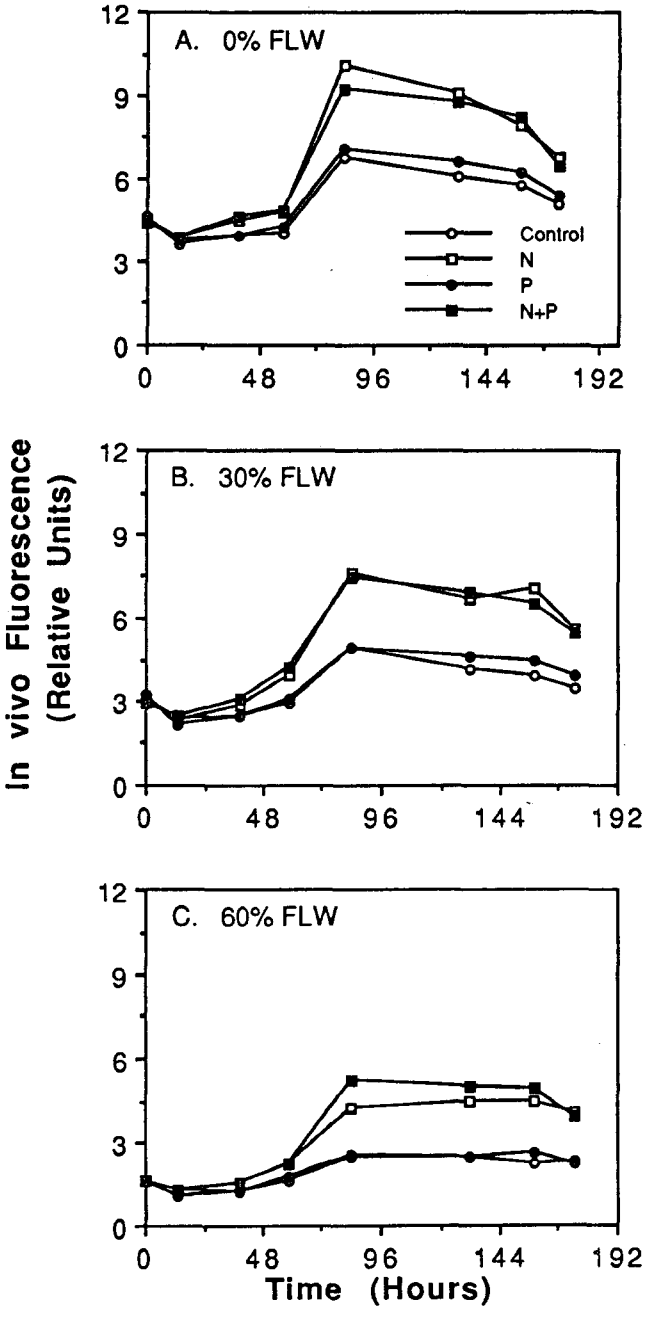

Figure 4.3. Average *in vivo* fluorescence values for Lake Apopka phytoplankton incubated on 6 Feb among four nutrient treatments and three dilution levels. FLW = Filtered lakewater.

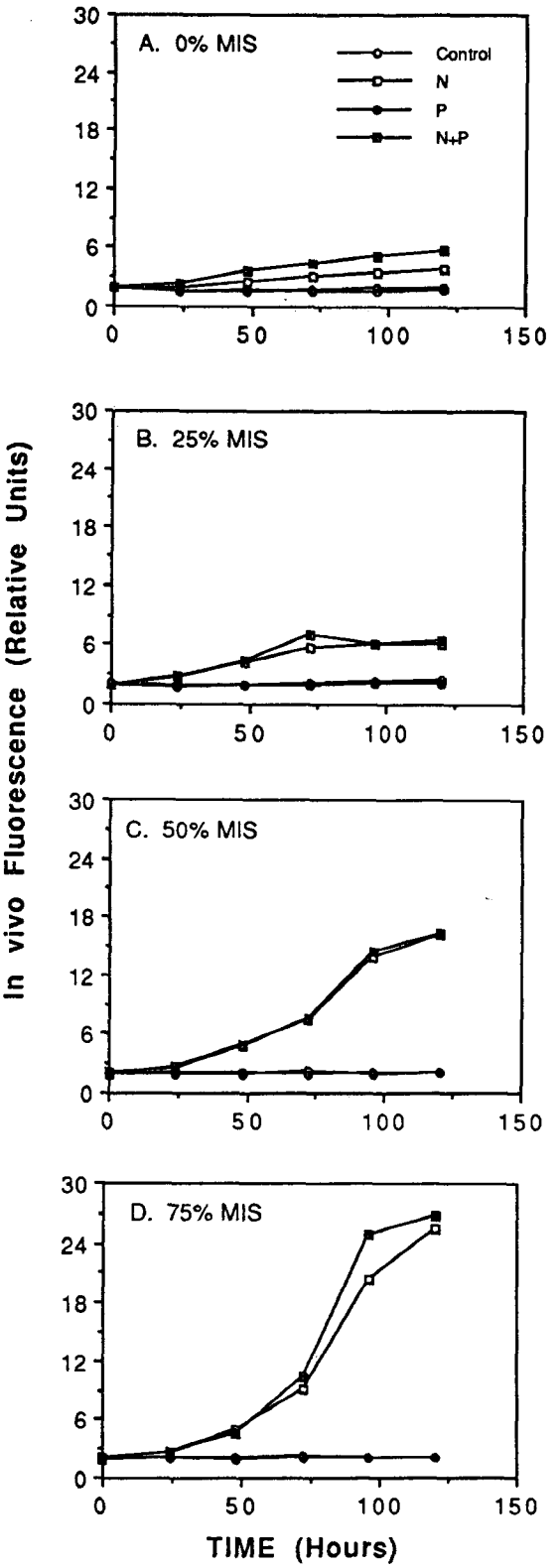

Figure 4.4. Average *in vivo* fluorescence values for Lake Apopka phytoplankton incubated on 26 June among four nutrient treatments and four dilution levels. Biomass was constant among treatments. MIS = Major ion solution.

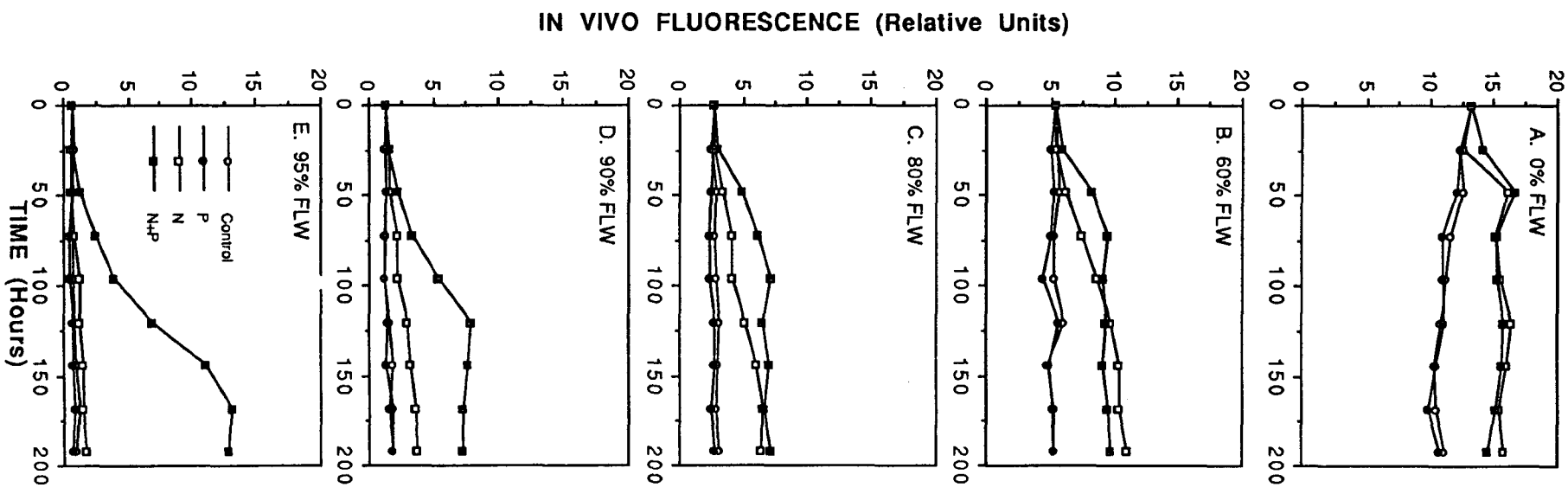

Figure 4.5 Average *in vivo* fluorescence values for Lake Apopka phytoplankton incubated on 4 Nov. among four nutrient treatments and five dilution levels. FLW = Filtered lakewater.

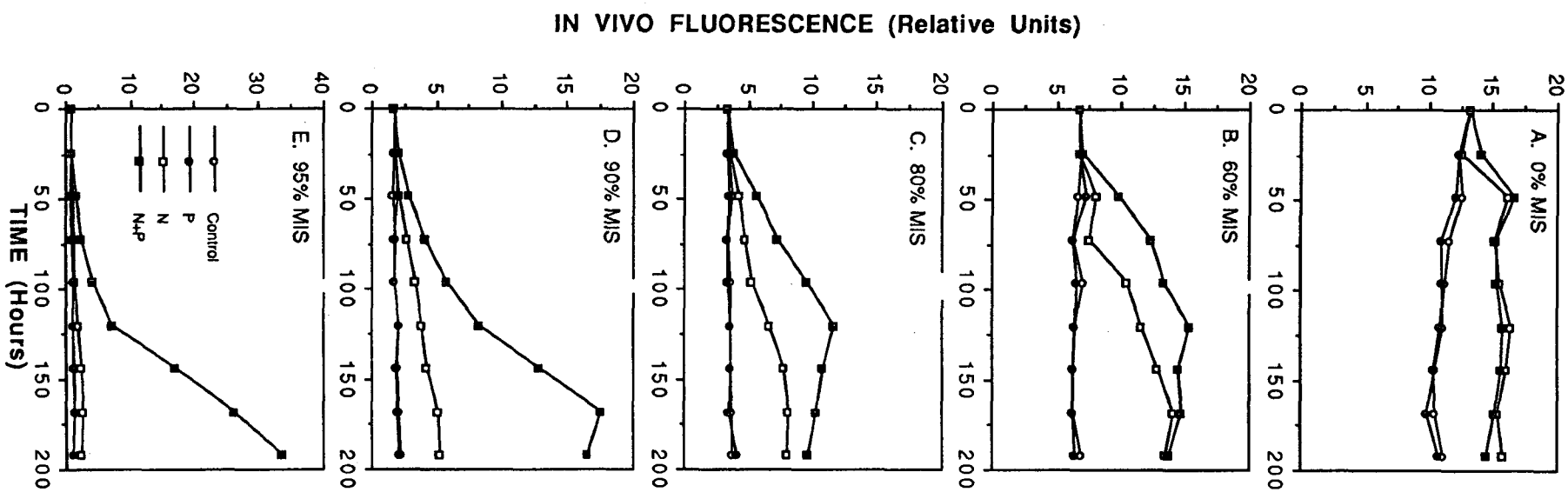

Figure 4.6 Average *in vivo* fluorescence values for Lake Apopka phytoplankton incubated on 4 Nov. among four nutrient treatments and five dilution levels. MIS = Major ion solution.

Table 4.4. Results from two-way analysis of variance comparing phytoplankton growth rates among dilution and nutrient treatments, whereby initial biomass varied as a function of dilution. Studies were conducted on surface lake water collected from Lake Apopka on five dates in 1991 and one of three diluents were used (DH_2O=deionized water, FLW=filtered lake water, MIS=major ionic solution). Treatments joined by underlining were not significantly different (Tukey's comparisons ($p<0.05$), $^{***}p<0.00001$, $^{*}p<0.05$).

Date (Diluent)	Two-way Factor	F-stat	Tukey's Pair-wise Comparison (Least to Greatest)
09 Jan	Dilution	0.56	0 30 60
(DH_2O)	Nutrients	34.34***	C P N
	Interaction	0.19	
24 Jan	Dilution	26.34***	0 30 60
(FLW)	Nutrients	63.39***	C P N N+P
	Interaction	5.05*	
07 Feb	Dilution	21.90***	0 30 60
(FLW)	Nutrients	52.73***	C P N N+P
	Interaction	3.88*	
26 Jun	Dilution	242.4***	0 25 50 75
(MIS/FLW)	Nutrients	419.8***	C P N N+P
	Interaction	81.5***	
04 Nov	Dilution	51.20***	0 60 80 90 95
(FLW)	Nutrients	320.47***	C P N N+P
	Interaction	14.04***	
04 Nov	Dilution	99.63***	0 60 80 90 95
(MIS)	Nutrients	372.91***	C P N N+P
	Interaction	17.96***	

differences in these experiments were complex, as indicated by significant interactions obtained in nearly all experiments and the lack of consistency between the two-way ANOVA and pairwise test results. This point is further illustrated, as growth among the four nutrient fields differed markedly at various levels of dilution (Fig. 4.7A). In addition, when growth at each level of dilution is plotted for the four nutrient treatments it is clear that the degree of overlap among dilution treatments is dependent on the nutrients added (Fig. 4.7B).

Given this, one-way ANOVA was used to test for differences among nutrient at each level of dilution in order to investigate interactions between nutrients and dilution on algal growth (Table 4.5). The general pattern of switching from N-limitation to co-limitation by both N and P among treatments was observed consistently in treatments receiving 60% or more diluent, although these differences were not always statistically significant. Viewed another way, dilution appears to relax the degree to which phytoplankton are limited by N.

Algal Yield and Total Phosphorus Concentration

Algal yield (maximum *in vivo* fluorescence minus the initial fluorescence yield) measured in the N-addition treatments was related with ambient total phosphorus concentrations (or percent raw lake water) in nearly all experiments (Figs. 4.8, 4.9). Maximum yields were achieved following N-addition in all experiments except

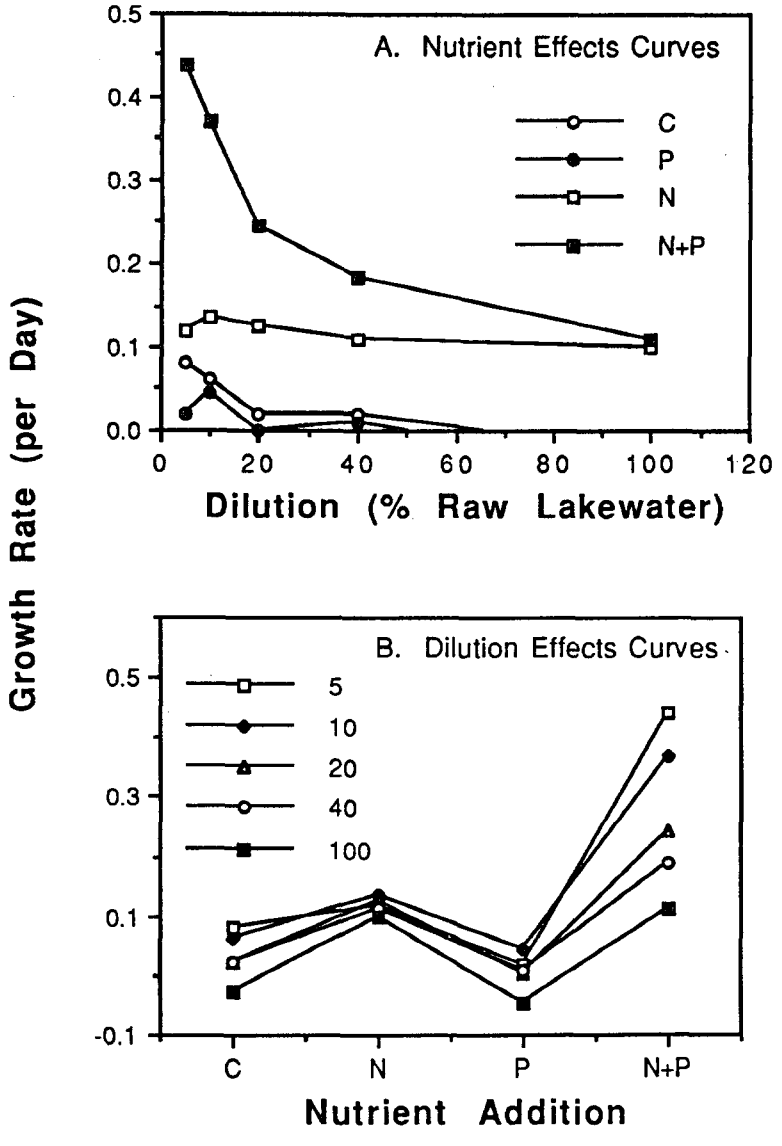

Figure 4.7. Phytoplankton growth response on 4 Nov. among nutrient treatments (A) and for five levels of dilution with filtered lakewater (B).

Table 4.5. Results from one-way analysis of variance comparing increasing phytoplankton growth rates among nutrient treatments at each level of dilution (blocked factor). Studies were conducted on surface (0.3 m) lake water collected from Lake Apopka in 1991. Treatments joined by underlining were not significantly different from one another when assessed with Tukey's pair-wise comparisons ($p < 0.05$). Abbreviations for diluents conform to those defined in Table 4.4. Where $^{***}p<0.00001$, and $^{**}p<0.01$.

Date (Diluent)	Blocked Factor		F-stat	Tukey's Pair-wise Comparison (Least to Greatest)			
7 Feb (FLW)	Dilution	0%	76.4***	C	P	N+P	N
		30%	51.7***	C	P	N+P	N
		60%	13.8**	C	P	N	N+P
24 Jan (FLW)	Dilution	0%	18.5***	C	P	N+P	N
		30%	91.3***	C	P	N+P	N
		60%	19.2***	C	P	N	N+P
26 Jun (MIS/FLW)	Dilution	0%	421.6***	C	P	N	N+P
		25%	153.0***	C	P	N	N+P
		50%	261.3***	C	P	N+P	N
		75%	546.1***	C	P	N	N+P
4 Nov (FLW)	Dilution	0%	121.8***	C	P	N	N+P
		60%	72.9**	C	P	N	N+P
		80%	169.0***	C	P	N	N+P
		90%	2294.5***	C	P	N	N+P
		95%	40.3**	C	N	P	N+P
4 Nov (MIS)	Dilution	0%	121.8***	C	P	N	N+P
		60%	169.0***	C	P	N	N+P
		80%	18.0**	C	P	N	N+P
		90%	212.0***	C	P	N	N+P
		95%	202.9***	C	P	N	N+P

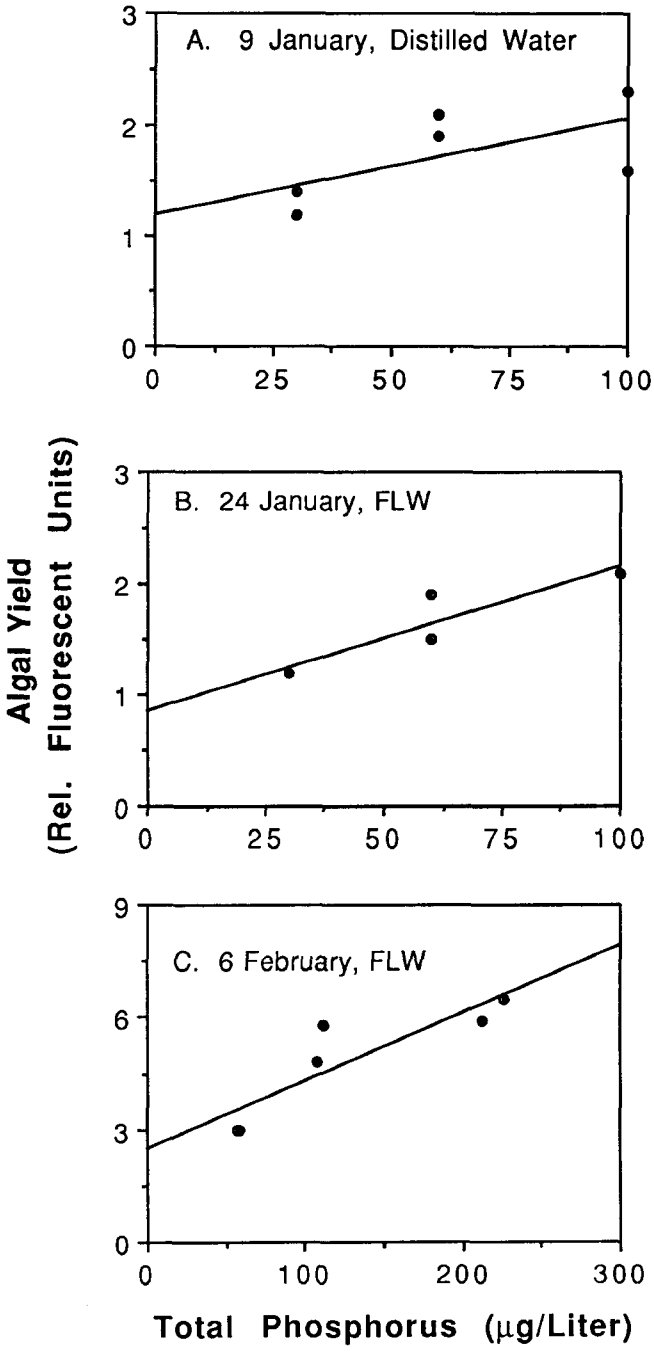

Figure 4.8. Maximum algal yield regressed or total phosphorus for three nutrient-dilution experiments conducted on surface phytoplankton communities in Lake Apopka.

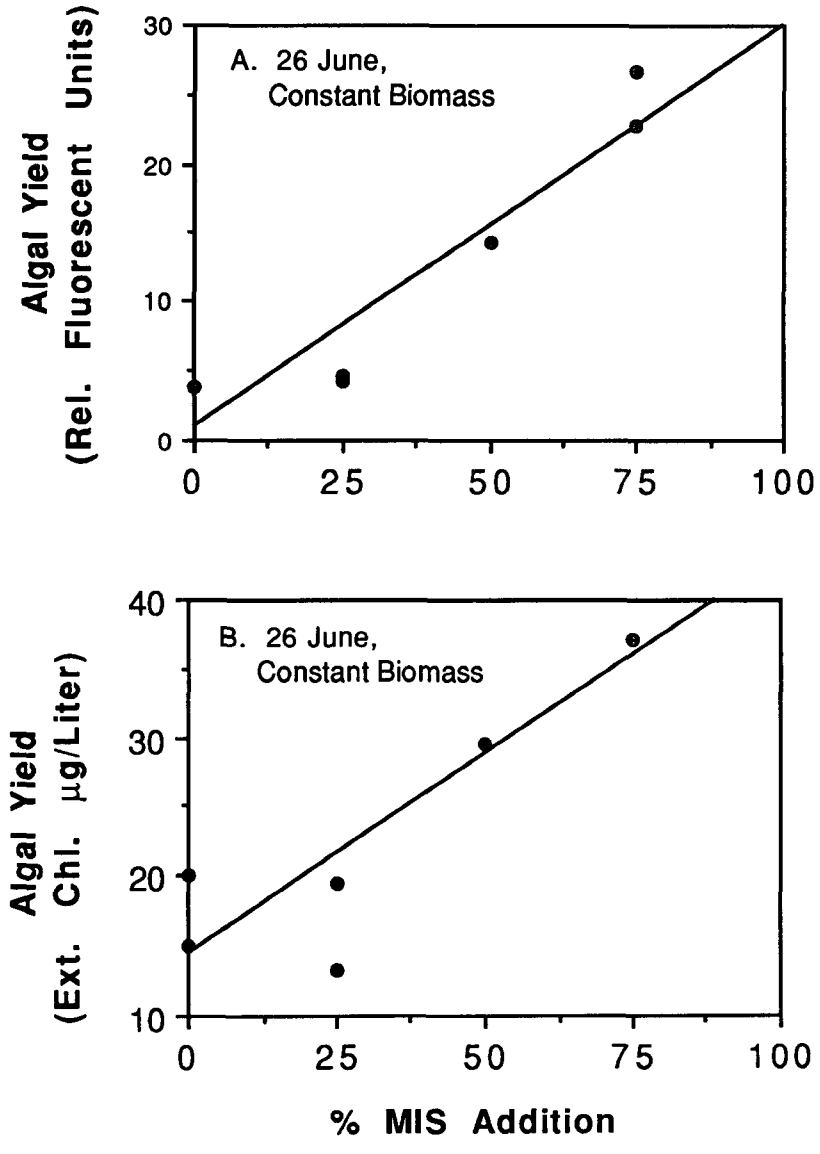

Figure 4.9. Maximum algal yield regressed onto percent MIS (major ion solution) added to lakewater for surface phytoplankton communities in Lake Apopka. Varying percentages of FLW (filtered lake water) were added to bottles in addition to the amount of MIS indicated such that algal biomass constitute 25% of the total mixture.

those on 4 November when N+P addition prompted maximum yield (see below). Total phosphorus accounted for 78-89% of the variation in algal yield (Table 4.6) although the nature of this relationship did vary between experiments. On 9 January, a marginally significant relationship was observed ($p < 0.20$). Also, this relationship with total phosphorus was not relevant in the experiments on 26 June where algal biomass and total P concentrations were constant among bottles. Instead, we measured changes in algal yield with the increasing percent of MIS added and found that algal yield was augmented by greater additions of MIS (Fig. 4.9).

Relationships between algal yield and total phosphorus were different among nutrient treatments following dilution with both FLW and MIS on 4 November. A negative relationship between maximum yield and total P (Figs. 4.10 and 4.11) was obtained for the N+P treatment. However, yields in the N treatment increased linearly, excluding data for the 100% raw lake water dilution treatment (0% dilution, Fig. 4.12). These results are important, because the degree of N-limitation of the phytoplankton was relaxed by dilution, such that phytoplankton in the N-treated flasks became P-limited and experienced higher yields with increasing total P, as predicted. Conversely, enrichment with both N and P promoted the greatest phytoplankton yields, and these were inversely related to total P concentrations. In addition, algal yields increased with the percent MIS added in the experiments conducted on 26 June where biomass (and total P) was constant among treatments. These results were surprising because total P did not vary among dilution treatments, thus suggesting that MIS treatment probably promotes higher algal yields.

Table 4.6. Summary of linear regression analyses assessing the relationship between the maximum yield measured with *in vivo* fluorescence (relative units from the N-addition treatment) and total phosphorus (μg Liter^{-1}) for five nutrient-dilution experiments (variable biomass). In addition, regression analyses evaluating the relationship between maximum algal yield and percent MIS added were conducted on one date (constant biomass). All experiments were performed on surface phytoplankton communities collected from Lake Apopka.

Diluent	Date	Y-intercept	Slope	n	r^2	Prob
			Variable Biomass:			
FLW	09 Jan	1.20	0.087	6	0.42	0.200
	24 Jan	0.85	0.131	6	0.86	0.005
	06 Feb	2.49	0.182	5	0.78	0.050
	04 Nov	0.42	0.097	10	0.79	0.002
MIS	04 Nov	1.33	0.150	10	0.86	0.001
			Constant Biomass:			
MIS	26 Jun	0.98	0.2900	8	0.89	0.001
	26 Jun[a]	14.37	0.2880	8	0.83	0.005

[a]Relationship based upon extracted chlorophyll a (μg Liter^{-1}).

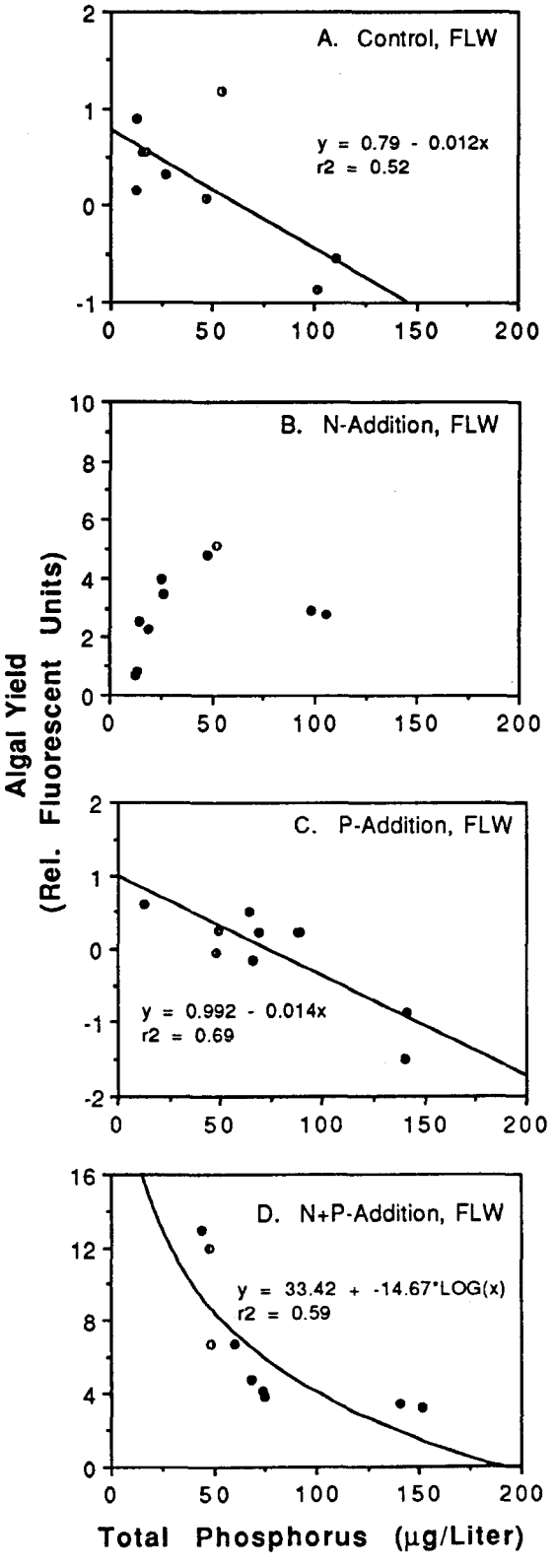

Figure 4.10. Maximum algal yield among four nutrient treatments regressed onto total phosphorus for a nutrient-dilution experiment conducted on 4 Nov. FLW = Filtered lakewater.

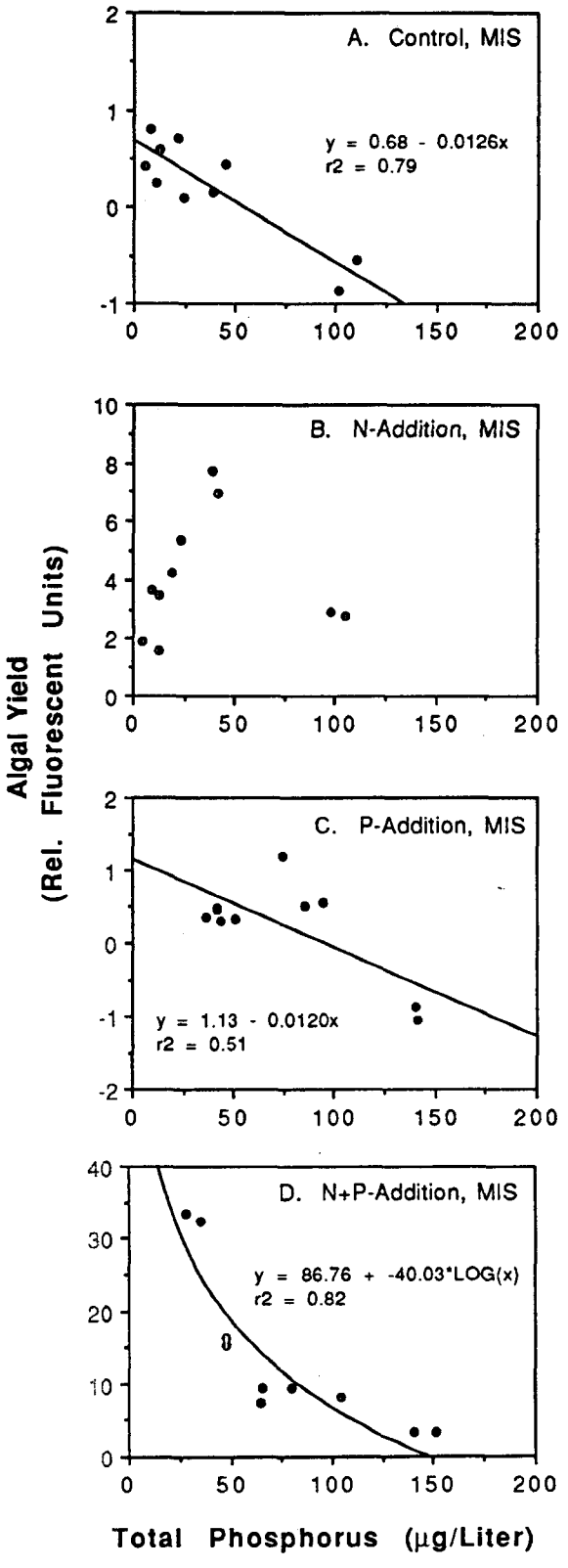

Figure 4.11. Maximum algal yield among four nutrient treatments regressed onto total phosphorus for a nutrient-dilution experiment conducted on 4 Nov. MIS = Major ion solution.

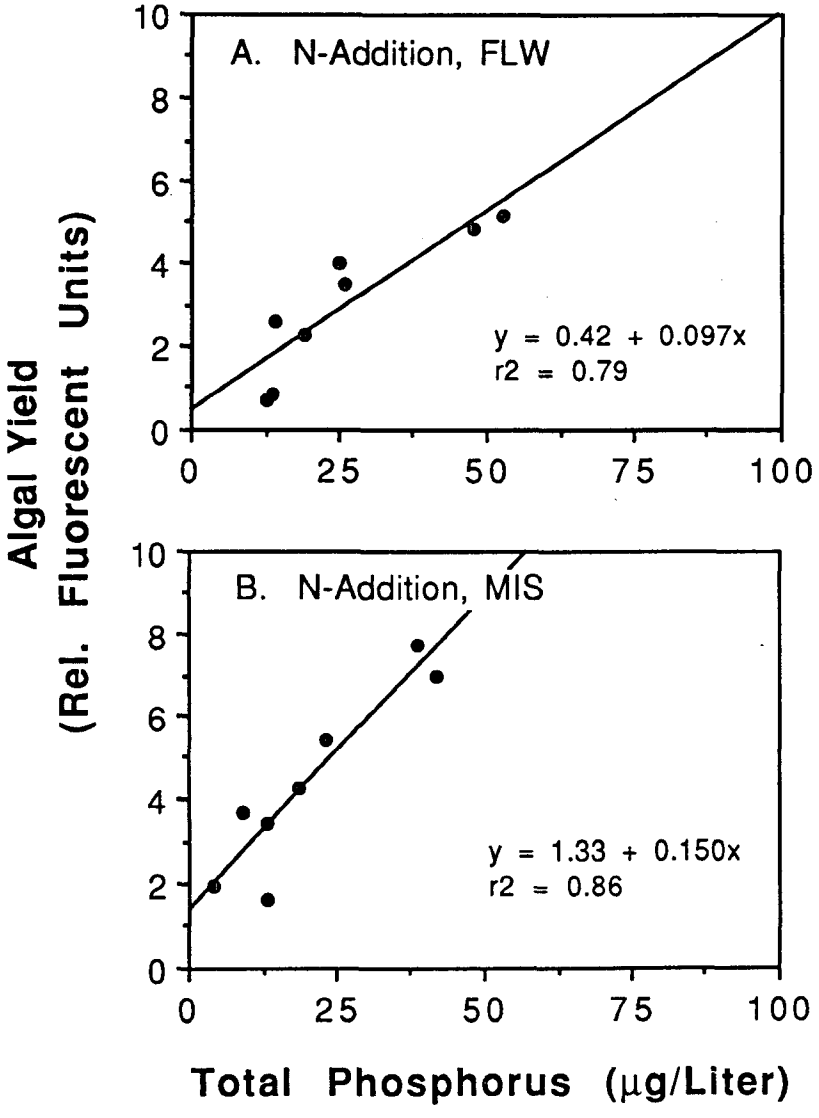

Figure 4.12. Maximum algal yield following nitrogen addition treatments to both FLW and MIS diluents regressed onto total phosphorus (4 Nov.) excluding values for 0% FLW and 0% MIS.

Phosphorus Storage Rates

Phosphorus storage rates varied greatly with the dilution of lake water in experiments conducted on 4 November using FLW (Fig. 4.13). After 24 h of incubation, P-storage rates increased logarithmically with increasing dilution of phytoplankton biomass in both P and N+P nutrient treatments. Conversely, rates in the control and N treatments plotted against the percent raw lake water showed no discernible pattern, presumably due to the fact there was very little P available for uptake.

Critical Assumptions of Lake Water Dilution Experiments

Experiments were conducted to test the major assumptions of the nutrient-dilution experiments (Fig. 4.14, Table 4.7). Preliminary tests indicate that there were no differences observed in the growth of phytoplankton incubated in bottles of varying size, such that containment artifacts in the 300-ml bottles utilized in our experiments were probably small (Fig. 4.14A). Second, N and P (both soluble and total) concentrations in all three of the diluents used in our experiments were lower than concentrations of N and P present in lake water (Table 4.7). Third, microscopic inspection of the diluents showed that very few cells passed through the filtration system used here. In addition, the growth of cells which might have passed through filter capsules did not manifest during the course of the experiments, even when we added nutrients to promote their growth (Fig. 4.14B). This indicates that no detectable contamination of nutrients or cells occurred during the course of our experiments. Fifth, grazing pressure by organisms <80 μm in size was capable of

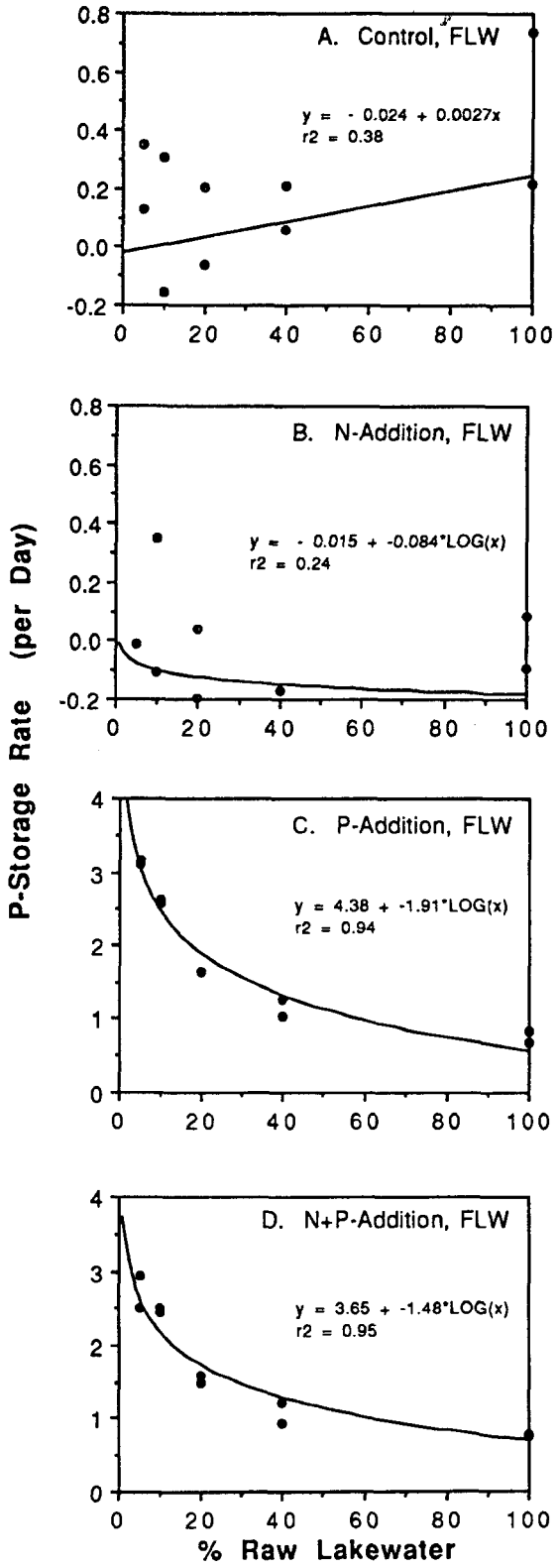

Figure 4.13. Phosphorus storage rates for Lake Apopka phytoplankton among four nutrient treatments and five levels of dilution with filtered lakewater. Experiments were conducted on 4 Nov.

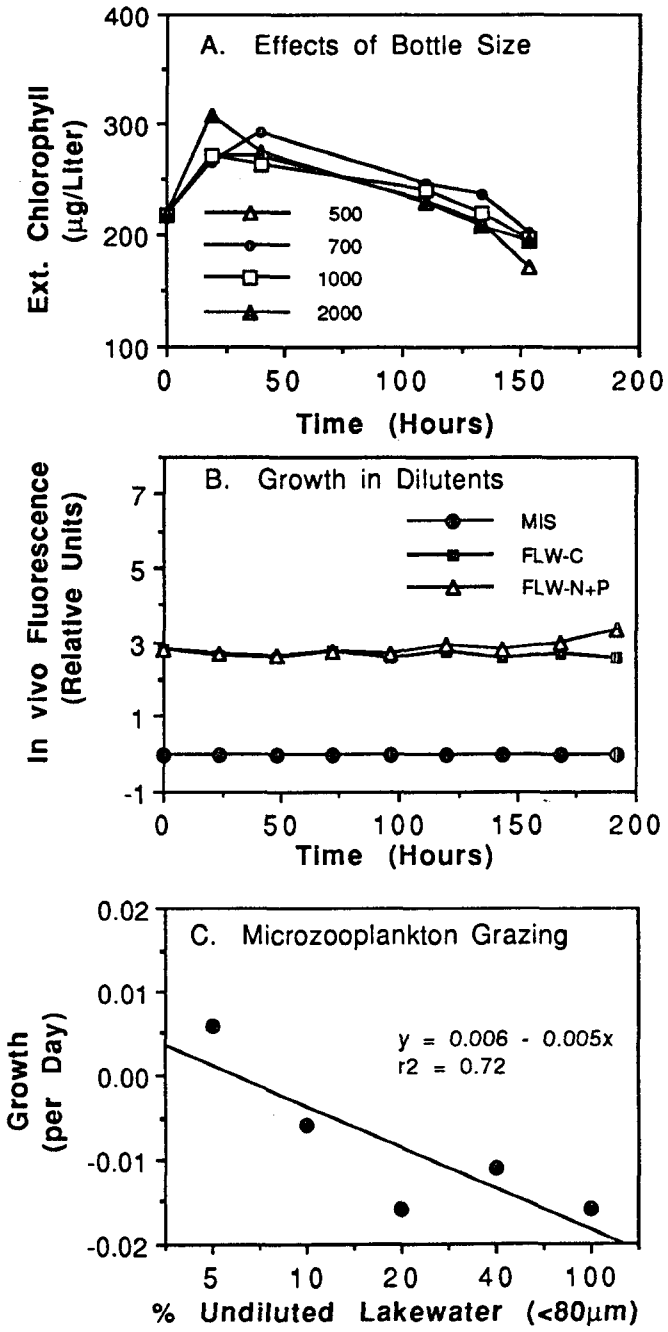

Figure 4.14. Experiments evaluating the effects of bottle size (A), algal growth in diluents used in experiments (B), and the influence on microzooplankton grazing on algal growth.

Table 4.7. Average (standard deviation) concentrations of phosphorus and nitrogen (in μg $Liter^{-1}$) for soluble reactive P (SRP), dissolved inorganic N (DIN), total P (TP), and total N (TN) for three diluents used in nutrient-dilution experiments. These values are compared with values for raw lake water collected (0.3 m) on 4 November from Lake Apopka. Estimates were derived from duplicate water samples.

	Phosphorus		Nitrogen	
Diluent	SRP	TP	DIN	TN
DH_2O	0.0	0.0	0.0	0
FLW	1.8 (0.7)	7.2 (0.0)	0.6 (0.1)	1295 (29.7)
MIS	1.4 (0.2)	5.9 (0.6)	0.9 (0.9)	23 (26.9)
Lake	2.0 (0.4)	105.8 (3.4)	0.8 (0.7)	3136 (128.7)

inparting losses that balance phytoplankton growth in the absence of added nutrients (Fig. 4.14C).

DISCUSSION

Effects of Dilution on Nutrient Limitation

The addition of nitrogen in our experiments clearly increased the growth of phytoplankton in undiluted Lake Apopka water; however, reduction of particulates in lake water can alter the response of the phytoplankton to nutrients. This is particularly true at dilution factors of 60% or greater, whereby phytoplankton growth appears to become limited by P. This factor was a consistent feature in nearly all of our experiments independent of the diluent utilized, although additional growth stimulation was achieved in treatments receiving the greatest dilution with major ionic solution (see below).

The occurrence of phosphorus limitation to freshwater phytoplankton has been demonstrated in a variety of environments, suggesting that N-limitation is less common and more prevalent in marine systems (Hecky and Kilham 1988; Vitousek and Howarth 1991). However, recent work in subtropical and tropical lakes indicates that N-limitation of phytoplankton may be more frequent than previously thought, particularly in productive lakes like Lake Apopka (Agusti et al. 1991). Moreover, reanalysis of experiments on temperate lakes suggests that N and N+P limitation are more prevalent than P-limitation (Elser et al. 1990) and nitrogen utilization can be an indicator of the severity of eutrophication

(Schelske 1975). These results support the notion that N-limitation is a consistent feature in some freshwater systems, particularly eutrophic lakes.

Predicted Changes in Algal Yield with P-Loading

The relationship between algal yield (relative *in vivo* fluorescence units, FU) and total phosphorus concentrations (μg Liter^{-1}) might be useful in assessing the influence decreasing P-loads have on phytoplankton chlorophyll yields. Our experiments were designed to determine the change in *in vivo* fluorescence (dependent variable) expected per unit increase in total P (independent variable), as expressed by the slope of this regression. The average slope from our N addition experiments was 0.129 FU μg P Liter^{-1} (coefficient of variation 30.2%). Our estimates predict that a 50% change in chlorophyll (FU) would require an equal change in total P, given the range of concentrations tested (e.g., Fig. 4.12). However, it is important to remember that these estimates were derived from experiments in which we induced P-limitation of the phytoplankton by (1) increasing dilution, and (2) nitrogen enrichment. Thus, extreme caution should be exercised in interpreting these results because it is not clear how this relationship in nature may be complicated by a number of factors (see below).

Factors Influencing Algal Yield-Total Phosphorus Relationships

Associated changes in algal yield with total P along our experimental dilution series may be complex, because manipulated

total P concentrations covary with other parameters. This is best illustrated in the experiments conducted on 4 November (see Fig. 4.5), where N, P, N+P and no additional nutrients were added to bottles at five levels of dilution (0, 60, 80, 90, and 95 % FLW). This experimental design was repeated substituting MIS for FLW as the diluent. The phytoplankton response to nutrient enrichment was similar between diluents; however, MIS promoted higher phytoplankton yields. We attribute these differences to substances inadvertently added (as chemical impurities) with the diluent. Trace elements that can be present as impurities in some manufactured chemicals (e.g., Carrick and Lowe 1988) can enhance algal growth (Lin and Schelske 1981). The possibility also exists that an inhibitory substance in Lake Apopka water was diluted with the addition of MIS, in that allelopathy can be important in Cyanobacteria dominated systems like Lake Apopka (Keating 1978).

Despite this, the relationship between algal yield and total P is similar to that utilized to estimate microzooplankton grazing on microplankton (Landry and Hassett 1982). Because increasing lake water dilution relieves grazing pressure, the slope of the growth rate of the prey (dependent variable) across the dilution series (independent variable) is an estimate of their grazing loss while the intercept is an estimate of prey growth. Maximum algal yields in control, P, and N+P treatments were inversely related to total phosphorus concentrations, while this relationship was positive when phytoplankton were enriched with N. To assess the effects of grazing in these experiments, algal yield values were converted to growth rates (yield divided by the incubation time); these values

were then regressed onto increasing percent raw lake water. Regressions using yield and growth rate as the dependent variables were not different for C, P, and N+P treatments (Figs. 4.10 and 4.15); however, the growth rate of N-enriched phytoplankton was inversely related with percent raw lake water (total P), whereas yield was positively related with total P.

Changes in algal yield with total P in the C and P treatments can most likely be attributed to microzooplankton grazing, as these results are akin to those commonly derived from experiments design to measure grazing rates (e.g., Landry and Hassett 1982; Fahnenstiel et al. 1991). Growth and grazing losses were similar between these two treatments, whereby grazing accounted for 17-25% of growth. Interestingly, the magnitude of these grazing losses correspond with the relative contribution microzooplankton make to the total planktonic grazer community (25% by biomass) in Lake Apopka (Crisman and Beaver 1988; H.J. Carrick, unpubl. data). The fact that grazing organisms <80 μm in size balance phytoplankton growth in one experiment (Fig. 4.14C), suggests even tighter couplings occur among smaller size organisms.

The phytoplankton yield versus total P relationship was positive following fertilization with N; however, when yields were standardized for the time it took to achieve maximum biomass (converted to growth rates) the relationship between growth and percent RLW was inverse. Viewed another way, higher algal yields generally took a longer period of time to accumulate. This might be explained by changes in the nutrient status of the phytoplankton with dilution. For instance, phytoplankton enriched with both P and

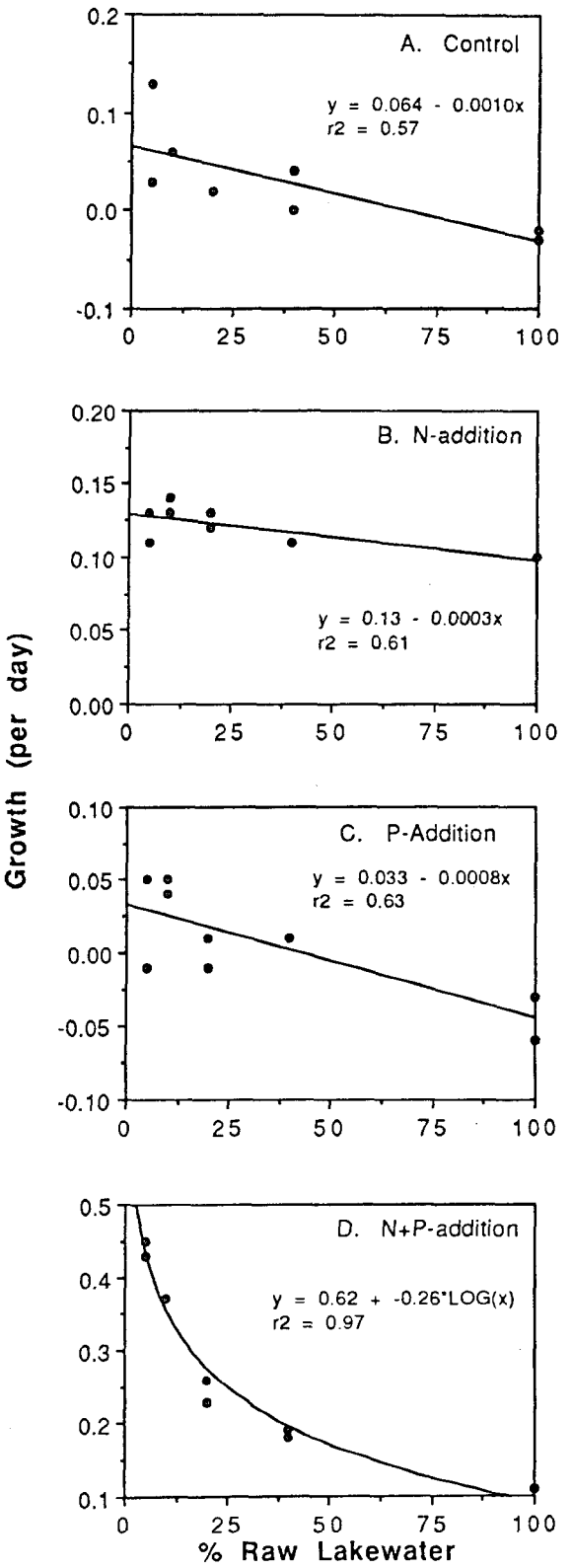

Figure 4.15. Algal growth rates among four nutrient treatments regressed onto percent raw lake water for a nutrient dilution experiment conducted on 4 Nov. The diluent used was filtered lakewater.

N+P exhibited a logarithmic increase in their P-storage capacity with dilution (decreasing total P, Fig. 4.13); this type of response is usually associated with phytoplankton that are P-limited (Lean and Pick 1981). Given the differences in growth response with dilution between N and N+P treatments, we conclude that phytoplankton enriched with N at high dilution had an initial burst of growth (under N-limited conditions) which slowed as they became more P-limited. The opposite appears to be true for N enriched phytoplankton at low dilution, whereby these more P-sufficient cells grew more slowly but ultimately produced greater absolute yields. Therefore, addition of N alone appeared to augment phytoplankton growth beyond the grazing rate exerted by microzooplankton and ultimately promotes an increase in algal yields with total P because phytoplankton become more P-limited when lake water particulates are diluted.

The question of why phytoplankton become more P-limited with dilution (or the degree of N-limitation is relaxed with dilution) is difficult to assess. The possibility exists that increased dilution also increased the light climate in the bottles. This might be important because augmented growth attributable to light-nutrient interactions has been observed for phytoplankton occurring in low light environments (e.g., Fahnenstiel et al. 1984), like those in Lake Apopka. While we agree that this factor needs to be reviewed in more detail, we feel light did not vary significantly among dilution treatments, because the bottles possessed a relatively small path length (3 cm) through which to attenuate light given average Secchi depths in Lake Apopka (25 cm). On the other hand, the concentration of nutrient per unit phytoplankton biomass changes along our

dilution series, because we added the same dose of nutrients to bottles. This means that the quantity of N or P at 90% FLW dilution is approximately 10-fold higher than that available to undiluted phytoplankton. The potential importance of this factor cannot be ruled out, in that phytoplankton growth can be related to nutrient supply (e.g., Schelske et al. 1986).

CONCLUSIONS

The application of nutrient-dilution experiments to the study of phytoplankton nutrient limitation in Lake Apopka is important for several reasons. First, the large magnitude of nutrients incorporated into algal biomass is indicative of the degree of nutrient "overloading" that exists in Lake Apopka. Such a condition is difficult to evaluate using classical approaches which assess relationships between phytoplankton biomass and phosphorus loading (Jones and Bachmann 1976). Second, the high proportion of N and P present in particulate matter (and low concentration of soluble reactive forms) makes it possible to apply nutrient enrichment along a series of dilutions of raw lake water in order to create a gradient of nutrient conditions to be utilized for predictive purposes (Paerl and Bowles 1987). Third, these experiments, in a sense, mimic the workings of the experimental marsh being applied to the current water quality problem in Lake Apopka (Lowe et al. 1992). Like the marsh, in these experiments nutrients that are bound in the particulate phase were removed. In addition, changes in the nutrient status of the phytoplankton were evaluated.

While the results from nutrient-dilution experiments were not confounded by containment problems or the other experimental artifacts we evaluated (Table 4.7; Fig. 4.13), some factors need to be considered prior to the applications of these results. First, even though the impact of microzooplankton grazing losses in our experiments were minor relative to nutrient-augmented phytoplankton growth, such losses constituted a significant portion of growth with no nutrient addition and P addition. Because microzooplankton grazing is directly related to increasing percent of raw lake water, this factor is an inherent feature of nutrient-dilution experiments that has been previously overlooked (Paerl and Bowles 1987). Second, MIS added to lake water samples promoted higher phytoplankton growth relative to samples receiving FLW, presumably due to the existence of impurities and/or complex chemical alterations resulting from the chemicals used in its construction. Thus, caution must be exercised when deriving estimates of algal yields from samples diluted with major ionic solution (MIS). This result also indicates that other chemical substances, in addition to N and P, could limit phytoplankton growth in Lake Apopka and further downplays the notion that phytoplankton growth was light-dependent in these experiments. Third, the concentration of nutrients available to phytoplankton varied with dilution, such that the ratio of nutrients to phytoplankton in the most dilute treatments was higher by an order of magnitude. This might account for observed changes in algal nutritional state; phytoplankton became more P-limited with dilution (or the degree of N-limitation was relaxed).

Increased P-storage by the phytoplankton with increasing dilution may have utility as a management tool. The relative capacity of phytoplankton to take-up and store P is a good indicator of their nutrient status (Healy 1979) and the affinity of phytoplankton for P increases under more oligotrophic conditions (Lean and Pick 1981). Thus, it makes inherent sense that as Lake Apopka phytoplankton are diluted with filtered lake water, their affinity for P-storage increases, as demonstrated in our experiments. These cells also contain more P per unit biomass (more than 3-fold higher in 95% compared with 0% FLW) and may remove P from the lake more efficiently than undiluted phytoplankton. Therefore, increased P-storage capacity with dilution may aid in the removal of P from Lake Apopka by: (1) removing available P more swiftly from the water column, and (2) creating a higher net decrease in the internal P load by producing more P-rich particles to be deposited in the wetland. The relative effectiveness of this internal "P-pump", in concert with P removal by the wetland, requires further experimentation in order to assess its role in the net export of P from Lake Apopka.

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Chapter 5

INDICATORS OF PHYTOPLANKTON NUTRIENT LIMITATION: AN EVALUATION OF METHODS

INTRODUCTION

Knowledge concerning the factors that govern lake productivity and trophic state are paramount to both our appreciation and ability to manage lake ecosystems. External nutrient-loading can explain much of the variation in phytoplankton biomass among lakes and can serve as a useful predictive tool (Jones and Bachmann 1976). However, the predictability of this relationship is limited because deficient cells can take up nutrients in excess of their immediate growth requirements (e.g., Droop 1968; Healey 1979). In addition, difficulties in measuring external nutrient concentrations (Rigler 1966; Tarapchak and Nalewajko 1986) and internal nutrient-loading (Rigler 1956; Lean 1973) hinders further the utility of the relationship. Lastly, the "nutrient-chlorophyll" relationship is correlative and other tests are required to truly elucidate the role nutrients play in influencing phytoplankton biomass.

Physiological indices of nutrient limitation and nutrient enrichment bioassays (NEB) can provide direct information on the current nutrient status of the phytoplankton themselves and their affinity for the nutrients in question (e.g., Lean et al. 1983; Dodds and Priscu 1990; Elser et al. 1990). Various indices have been

developed to assay for both nitrogen (N) and phosphorus (P) limitation. The attractiveness of these techniques lies in the fact that phytoplankton nutrient limitation can be assayed without enclosing phytoplankton for long periods of time (usually < 2 h) and many of these methods do not require the addition of nutrients. It is the focus of this portion of the report to: (1) give a brief literature review of physiological indices used to measure phytoplankton nutrient limitation, (2) present a case study in which we compared several indices in order to evaluate nutrient limitation of natural Lake Apopka phytoplankton, and (3) indicate (at present) what we feel are the most promising indices to study limitation of phytoplankton communities in Lake Apopka.

Physiological Indices of Nitrogen Deficiency

Four techniques are commonly used to determine nitrogen limitation of phytoplankton. The first and most common is measured using ammonium enhanced dark ^{14}C uptake (Yentsch et al. 1977; Vincent 1981a). Lakewater is dispensed into darkened vessels containing freshly collected lakewater, to which various concentrations of ammonium as NH_4Cl is added, while simultaneously inoculating each flask with $H^{14}CO_3^-$. Vessels are sampled through time to determine rates of carbon uptake. Carbon-14 uptake in the <1 μm size class should be evaluated to determine the portion of fixation attributable to chemoautotrophic NH_4-oxidizing bacteria (Dodds and Priscu 1991). Second, uptake of N can be determined directly by adding NO_3^- labelled with ^{15}N as a tracer and added to lakewater (Murphy 1980). This can be performed at a

number of $^{15}NO_3^-$ concentrations to determine specific uptake kinetics (Lean et al. 1982, 1983; Timberley and Priscu 1986). Third, the activity of nitrate reductase can be measured simply by extraction in toluene and appears to reflect the N-status of phytoplankton cells (Hochman et al. 1986; Wynne and Berman 1990). Fourth, uptake of N (as NH_4^+ or NO_3^-) can be estimated by measuring the disappearance of added soluble N from lakewater by serial sampling and analysis using common wet chemistry methods (e.g., Dortch et al. 1982).

Physiological Indices of Phosphorus Limitation

Four methods are commonly used to evaluate phosphorus limitation of phytoplankton. The first method evaluates P-deficiency using ^{32}P-turnover (Vincent 1981b; Lean et al. 1983), whereby collected lakewater is incubated in vessels with carrier-free ^{32}P-PO_4^{3-}. Subsamples are removed from the vessel and at various time intervals (up to 2 h) and processed for liquid scintillation counting. When ^{32}P-turnover is corrected for chlorophyll (or some measure of phytoplankton biomass) it can more accurately reflect the P-status of phytoplankton cells (Lean et al. 1983). Low numbers (short times) indicate P-deficient cells and long times are characteristic of P-sufficient cells. The uptake kinetics of P by phytoplankton can be measured by determining ^{32}P-turnover at increasing concentrations of cold PO_4^{3-} (Lean and Pick 1981). Second, a simple hot-water extraction procedure (HEP) to determine surplus-stored P (luxury consumption) can be conducted by concentrating phytoplankton onto filters followed by autoclaving

(60 min) to hydrolyze polyphosphates as SRP (Fitzgerald and Nelson 1966). HEP concentration appears to be a good indicator of nutrient status, as it is inversely related to P-limitation of phytoplankton (Wynne 1981; Schelske and Sicko-Goad 1990). Third, high alkaline phosphatase activity (APA) is indicative of increasing P-limitation (e.g., Smith and Kalff 1981; Wetzel 1981). APA can be measured by enzymatic hydrolysis of an artificial substrate added to lakewater samples yielding a fluorescent product which can be quantified with a fluorometer (Hill et al. 1968). Fourth, uptake of P (as SRP) can be estimated by measuring the disappearance of added soluble P from lakewater by serial sampling and analysis using common wet chemistry methods (e.g., Schelske et al. 1986).

RESULTS AND DISCUSSION

Evaluation of Methods: A Case Study

On two occasions (18 April and 21 August 1990) the nutrient status of Lake Apopka phytoplankton and their affinity for nutrients (N and P) were evaluated using both nutrient enrichment bioassays (NEB) and two nutrient deficiency indices (Alkaline phosphatase activity, APA and hot water extractable phosphorus, HEP). The design of the nutrient enrichment bioassays was simple and is described in detail in Chapter 3. Briefly, a factorial design was employed, whereby two bottles (500-ml flasks) containing freshly collected lake water were augmented with PO_4^{3-} (40 µg P $Liter^{-1}$), NO_3^- (400 µg N $Liter^{-1}$), N+P, and two did not receive nutrients and served as experimental controls. All bottles were incubated for 5 d at ambient light and temperature in a walk-in incubator. Daily

samples were taken to determine in vivo fluorescence and various chemical measurements (APA and HEP). Experiments on 18 April and 21 August 1990 were performed in collaboration with Dr. Susan Newman; the details of which are discussed in her dissertation (Newman 1991).

These data demonstrate that indices of P-limitation were consistent with results from the bioassays when P was limiting phytoplankton growth (21 August) but were otherwise difficult to interpret (Table 5.1). For static, point in time field measurements of HEP and APA to be useful in predicting N and/or P limitation in Lake Apopka, phytoplankton must be tested under a variety of physiological conditions in parallel with standard NEB's. Such data could provide quantitative limits in HEP and APA associated with phytoplankton nutrient status, similar to that which exists for ^{32}P-kinetics (Lean and Pick 1981).

Additional results from nutrient-dilution experiments indicate the promise in using estimates such as the rate of HEP storage as an indicator of P-deficiency. In experiments conducted on 4 November, 1991 phytoplankton demonstrated varying degrees of N and P limitation along a gradient of increasing lake water dilution as determined from NEB; undiluted phytoplankton were N-limited and become co-limited by N and P with increasing dilution (Chapter 4; Figs. 4.5 and 4.6). Correspondingly, the overall rate of HEP-storage for the phytoplankton in the P and N+P treatments increased logarithmically with dilution (Chapter 4; Fig. 4.13). These results indicate that increasing HEP-storage by phytoplankton appears to be good indicator of P-deficiency. Moreover, measurements of HEP-

Table 5.1. A comparison of various methods evaluating the nutrient limitation of Lake Apopka phytoplankton on two dates in 1990. Where bioassay=final/initial (*in vivo* fluoresence units), APA=alkaline phosphatase activity (nmol APA μg Chl min^{-1}), and HEP=hot water extractable phosphorus (μg P μg Chl^{-1}).

		Treatment			
Date	Method	C	N	P	N+P
18 April	Bioassay	1.00	1.40	1.00	2.10
	APA	2.20	2.50	0.50	0.50
	HEP	0.086	0.080	0.206	0.091
21 August	Bioassay	2.10	2.10	3.60	3.60
	APA	0.70	0.70	0.15	0.15
	HEP	0.68	1.34	0.67	1.25

storage by phytoplankton in differing nutritional states, such as the assemblages existing along our dilution gradients, might eventually be used to predict the current nutrient status of Lake Apopka phytoplankton with this relatively simple measurement. These P-storage measurements differ from the HEP values presented above (Table 5.1), in that they are exponential uptake rates, that take into account changes associated with cellular division (which is an exponential function).

CONCLUSIONS

Under most circumstances it appears to be necessary to utilize nutrient enrichment bioassays in order to make definitive statements concerning nutrient limitation (Dodds and Priscu 1990). In addition, bioassays provide information on phytoplankton yield relative to nutrient loads and species-specific responses to enrichment. While agreement among enrichment bioassays and indices for nutrient limitation is commonly observed (e.g., Smith and Kalff 1981; Lean et al. 1989; St. Amand et al. 1989), rapid changes in the nutrient-status of phytoplankton underscores the utility of applying both methods (Elser and Kimmel 1986). Thus, identification of reliable physiological indices of nutrient-limitation can ultimately be used to make rapid assessments of phytoplankton nutrient status (e.g., Lean et al. 1983), with only periodic validation with the more elaborate and time-consuming nutrient enrichment bioassays.

Choices concerning the application of specific physiological indicators are often influenced by their relative predictability, cost,

and ease of use. P-deficiency indicators (APA, HEP, and ^{32}P-turnover) appear to be predictable relative to enrichment experiments (Smith and Kalff 1981; Wetzel 1981; Lean et al. 1989) and consistent among themselves (Fitzgerald and Nelson 1966; Lean et al. 1987). However, agreement between N-indices and corresponding nutrient bioassays are less consistent (Lean et al. 1982). Dark ^{14}C-fixation by NH_4^+ enhancement may not provide unequivicol evidence for N-limitation of phytoplankton, given the confounding influence of fixation by NH_4^+-oxidizing bacteria (Dodds and Priscu 1991). While N-uptake kinetics appear to be sensitive to environmental conditions, such as light, measurements made under a variety of conditions can yield interpretable results (e.g., Dortch et al. 1991). Thus, relying strictly on various N-indices may produce misleading results, particularly in systems where N-fixation can become periodically important (Vincent 1981a).

At present, it is our belief that nutrient enrichment bioassays appear to be the most reliable indicator of nutrient limitation to phytoplankton in Lake Apopka. Early in this study it became evident that phytoplankton in Lake Apopka were generally growth limited by N. Because of this and the fact that very few good alternatives for measuring N-limitation exist, the focus of this study was to evaluate nutrient limitation using NEB. In addition to NEB, physiological indicators of N and P limitation can be used to provide an independent validation. For indices to be used as predictive tools of phytoplankton nutrient status, they need to be calibrated in parallel with NEB for phytoplankton assemblages of varying nutrient deficiency. We feel the most appropriate methods for this are HEP-

storage or P-uptake for evaluation of P-limitation and N-uptake to corroborate N-limitation. All three of these assays are non-hazardous (avoid the use of harsh chemicals), simple to conduct, and do not require any specialized equipment (e.g., radioisotopes) other than that used to measure basic water chemistry.

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Appendix A. Lake Apopka photosynthetically active radiation (PAR) for upwelling, downwelling and deck co-sine sensors, absorption coefficients (K_t), dissolved oxygen, water temperature at given depths.

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen ($mg\ L^{-1}$)	Temp. (^0C)
	0	1312.0	130	1540		11.7	25.0
891108	10	809.7	81	1191	2.25		
891108	20	630.1	63	1404	3.20		
891108	25					11.6	25.0
891108	30	403.4	40	1461	3.76		
891108	50	213.4	21	1584	3.69	11.7	25.0
891108	75	94.1	9	1640	3.60	11.8	25.0
891108	100					11.7	25.0
891206	0	938.9	94	1030			
891206	10	809.2	81	1309	3.86		
891206	20	496.4	50	1003	3.04		
891206	30	353.8	35	937	2.93		
891206	40	304.2	30	1007	2.76		
891206	50	199.9	20	990	3.01		
891206	75	119.6	12	1166	2.91		
900124	0	543.0	54	544		11.7	22.8
900124	10	276.9	28	526	5.45		
900124	25					12.2	21.1
900124	30	466.6	47	1219	2.88		
900124	40	450.9	45	1557	2.86		
900124	50	329.1	33	1495	2.84	12.4	20.8
900124	60	174.9	17	1060	2.85		
900124	70	124.9	12	1036	2.89		
900124	75					12.0	20.3
900124	80	74.8	8	948	3.05		
900124	90	13.9	1	315	3.36		
900124	100	9.3	1	319	3.44	12.0	20.2
900124	110	5.6	0	380	3.76		
900207	0	1427.0	143	1547		9.3	21.6
900207	10	1066.0	107	1550	2.94		
900207	20	732.0	73	1546	3.33		
900207	25					10.8	19.5
900207	30	498.6	50	1558	3.53		
900207	40	304.5	30	1566	3.89		
900207	50	203.4	20	1567	3.92	11.4	18.5
900207	60	130.2	13	1555	4.00		
900207	70	75.3	8	1568	4.22		
900207	75					9.9	18.2
900207	80	31.0	3	1583	4.82		
900207	90	14.4	1	1600	5.14		
900207	100	0.0	0	1602			

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen (mg L^{-1})	Temp. (^0C)
900221	0	797.0	64	904		8.4	22.5
900221	10	489.0	44	1062	6.42		
900221	20	190.1	28	1221	8.37		
900221	25					8.4	22.5
900221	30	30.5	3	761	10.22		
900221	40	13.5	1	755	9.71		
900221	50	6.5	1	927	9.56	8.4	22.5
900221	60	3.6	0	1420	9.75		
900221	70	1.3	0	1684	10.06		
900221	75					8.4	22.5
900221	80	0.0	0	980			
900221	100					8.3	22.5
900307	0	614.8	46	754		9.4	20.0
900307	10	274.5	21	850	9.24		
900307	20	210.5	18	1688	9.33		
900307	25					9.4	20.0
900307	30	95.5	9	1899	9.23		
900307	40	52.2	5	2460	9.09		
900307	50	17.5	2	2453	9.44	9.4	20.0
900307	60	5.1	0	2029	9.60		
900307	70	1.1	0	884	9.24		
900307	75					9.4	20.0
900307	80	0.4	0	785	9.31		
900307	90	0.2	0	788	9.05		
900307	100	0.0	0	819	10.08	9.4	20.0
900321	0	1702.0	84	1951		10.7	19.4
900321	10	779.2	51	1942	7.61		
900321	20	303.9	21	1947	8.52		
900321	25					10.6	19.4
900321	30	138.5	10	1967	8.32		
900321	40	59.3	4	1964	8.35		
900321	50	24.8	2	1980	8.44	10.6	19.3
900321	60	12.5	1	1980	8.16		
900321	70	5.6	0	1984	8.16		
900321	75					10.5	19.3
900321	80	2.4	0	1976	8.19		
900321	90	1.1	0	1971	8.15		
900321	100	0.3	0	1978	8.64	10.4	19.2
900404	0	1561.0	82	1804		8.1	20.5
900404	10	577.3	31	1799	9.91		
900404	20	139.5	11	1829	12.01		
900404	25					8.0	20.5
900404	30	52.1	4	1826	11.29		
900404	40	16.9	1	1827	11.27		
900404	50	5.2	0	1838	11.38	8.0	20.5
900404	60	2.0	0	1832	11.11		
900404	70	0.6	0	1836	11.20		

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen ($mg\ L^{-1}$)	Temp. (^0C)
900404	75					8.0	20.8
900404	80	0.1	0	1857	12.17		
900404	90	0.0	0	1848			
900404	100					8.0	20.8
900418	0	2013.0	174	1987		11.8	28.5
900418	10	1320.0	161	1993	3.93		
900418	20	1007.0	131	1987	3.27		
900418	25					12.3	28.0
900418	30	743.7	106	2002	3.18		
900418	40	563.1	85	2000	3.06		
900418	50	409.4	65	2010	3.08	12.7	27.5
900418	60	281.4	45	2001	3.18		
900418	70	211.8	35	2014	3.14		
900418	75					12.8	27.0
900418	80	141.7	23	2012	3.25		
900418	90	90.9	14	2026	3.39		
900418	100					12.0	26.5
900504	0	1894.0	139	1988		8.4	26.8
900504	10	1010.0	97	2000	6.14		
900504	20	591.0	55	2052	5.89		
900504	25					8.2	26.8
900504	30	264.0	30	2077	6.59		
900504	40	136.0	16	2100	6.62		
900504	50	70.5	8	2173	6.68	8.2	26.8
900504	60	38.3	5	2187	6.59		
900504	70	20.1	2	2072	6.49		
900504	75					8.2	26.8
900504	80	8.7	1	1834	6.58		
900504	90	2.5	0	1930	7.36		
900516	0	1885.0	143	1927		8.6	28.0
900516	10	1088.0	117	1953	5.34		
900516	20	571.3	80	1954	5.75		
900516	25					8.7	28.0
900516	30	352.2	49	1957	5.46		
900516	40	221.2	31	1971	5.27		
900516	50	129.0	18	1973	5.30	8.7	27.5
900516	60	72.7	10	1970	5.37		
900516	70	42.2	6	1978	5.39		
900516	75					8.5	27.5
900516	80	5.9	1	1991	7.14		
900516	90	0.0	0	1998			

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen (mg L^{-1})	Temp. (^0C)
900530	0	1846.0	131	1869		11.2	29.0
900530	10	906.3	107	1875	6.72		
900530	20	560.3	68	1874	5.74		
900530	25					11.3	28.2
900530	30	302.8	39	1877	5.87		
900530	40	161.5	20	1880	5.98		
900530	50	87.5	11	1884	6.01	10.8	27.0
900530	60	42.6	5	1903	6.23		
900530	70	22.8	3	1896	6.22		
900530	75					9.2	26.5
900530	80	0.0	0	1895			
900613	0	1969.0	99	2015		10.2	28.5
900613	10	900.4	70	2017	7.32		
900613	20	409.0	38	2007	7.51		
900613	25					10.2	28.5
900613	30	185.8	18	1967	7.55		
900613	40	38.9	4	936	7.70		
900613	50	28.8	3	1575	7.79	9.9	28.0
900613	60	8.9	1	1067	7.80		
900613	70	3.5	0	860	7.72		
900613	75					9.2	27.8
900613	80	4.1	0	2064	7.68		
900613	90						
900627	0	1015.0	86	1351		9.6	29.2
900627	10	772.0	74	1342	4.61		
900627	20	409.4	46	1301	5.25		
900627	25					10.0	28.8
900627	30	230.1	28	1233	5.21		
900627	40	106.8	14	1155	5.65		
900627	50	53.2	7	1111	5.83	8.3	28.0
900627	60	26.1	4	1089	6.01		
900627	70	15.3	2	1087	5.93		
900627	75					6.9	28.0
900627	80	9.1	1	1106	5.87		
900627	90	4.0	0	1102	6.17		
900718	0	1499.0	81	1604		10.4	26.0
900718	10	721.5	49	1744	8.16		
900718	20	307.1	25	1730	8.26		
900718	25					10.4	26.0
900718	30	129.7	11	1689	8.28		
900718	40	59.0	5	1678	8.15		
900718	50	24.9	2	1657	8.21	10.4	26.0
900718	60	11.5	1	1511	7.97		
900718	70	6.1	1	1568	7.79		
900718	75					10.3	26.0
900718	80	2.4	0	1465	7.89		

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen (mg L^{-1})	Temp. (^0C)
900718	90	1.2	0	1549	7.87		
900718	100					10.0	26.0
900801	0	1880.0	104	1884		10.9	31.0
900801	10	1270.0	106	1882	3.13		
900801	20	890.2	85	1889	3.31		
900801	25					11.0	31.2
900801	30	626.2	65	1896	3.36		
900801	40	430.6	48	1891	3.44		
900801	50	290.4	33	1896	3.54	10.5	31.2
900801	60	198.9	22	1915	3.60		
900801	70	133.3	15	1907	3.65		
900801	75					9.3	31.0
900801	80	85.3	9	1913	3.76		
900801	90	50.3	5	1930	3.95		
900801	100					8.2	31.0
900822	0	1690.0	133	1822		10.0	32.0
900822	10	1257.0	120	1819	2.78		
900822	20	935.8	98	1813	2.81		
900822	25					10.2	31.8
900822	30	659.4	76	1835	3.05		
900822	40	475.8	59	1851	3.11		
900822	50	316.3	41	1862	3.30	10.7	31.2
900822	60	228.8	30	1868	3.29		
900822	70	155.8	20	1850	3.36		
900822	75					10.4	30.5
900822	80	114.7	15	1850	3.32		
900822	90	78.2	10	1837	3.38		
900822	100	54.1	6	1843	3.43	9.8	30.5
900906	0	1915.0	126	2026		9.4	29.0
900906	10	1202.0	74	1405	0.97		
900906	20	695.3	72	2018	4.83		
900906	25					9.4	28.5
900906	30	443.0	48	2069	4.80		
900906	40	89.6	21	1393	6.33		
900906	50	38.3	4	696	5.58	9.3	28.0
900906	60	27.4	3	661	5.14		
900906	70	15.5	2	728	5.36		
900906	75					8.1	27.5
900906	80	7.4	1	705	5.57		
900906	90	14.9	1	1454	5.02		
900906	100	6.3	0	2102	5.73		

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen (mg L^{-1})	Temp. (^0C)
900920	0	449.0	38	526		8.7	26.0
900920	10	242.5	24	514	6.56		
900920	20	138.2	15	506	5.96		
900920	25					8.7	26.5
900920	30	87.4	10	507	5.49		
900920	40	57.1	7	508	5.17		
900920	50	31.3	4	509	5.33	8.6	26.5
900920	60	19.7	3	511	5.21		
900920	70	12.6	2	506	5.09		
900920	75					8.7	26.5
900920	80	7.9	1	504	5.05		
900920	90	4.8	1	506	5.04		
900920	100						
901002	0	1398.0	97	1493		10.4	25.5
901002	10	1014.0	91	1604	3.73		
901002	20	769.2	76	1624	3.26		
901002	25					10.1	25.5
901002	30	577.6	59	1661	3.20		
901002	40	436.7	44	1639	3.07		
901002	50	294.5	31	1672	3.27	10.2	25.5
901002	60	226.1	24	1653	3.15		
901002	70	166.0	17	1671	3.16		
901002	75					9.9	25.5
901002	80	129.2	13	1652	3.07		
901002	90			1661			
901017	0	994.3	81	1065		8.4	25.0
901017	10	543.0	47	1064	5.89		
901017	20	237.9	24	882	6.07		
901017	25					8.3	25.0
901017	30	134.1	15	821	5.69		
901017	40	79.9	9	757	5.36		
901017	50	45.1	5	757	5.42	8.3	25.0
901017	60	27.9	3	759	5.32		
901017	70	15.1	2	758	5.43		
901017	75					8.2	25.0
901017	80	9.2	1	720	5.31		
901017	90	5.8	1	712	5.22		
901017	100	1.8	0	714	5.84	8.2	25.0
901107	0	1411.0	120	1545		11.6	24.7
901107	10	981.8	108	1551	3.53		
901107	20	656.4	84	1555	3.71		
901107	25					11.8	23.7
901107	30	443.0	62	1557	3.76		
901107	40	320.0	46	1556	3.62		
901107	50	195.2	28	1558	3.88	12.3	22.2
901107	60	140.4	20	1555	3.78		

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen (mg L^{-1})	Temp. (^0C)
901107	70	93.9	14	1561	3.82		
901107	75					11.8	21.1
901107	80	65.1	10	1565	3.80		
901107	90	44.2	6	1567	3.83		
901107	100					10.6	22.2
901120	0	1298.0	96	1387		11.0	21.0
901120	10	837.0	81	1400	4.22		
901120	20	541.6	59	1404	4.24		
901120	25					11.5	20.5
901120	30	353.7	47	1417	4.21		
901120	40	237.5	33	1421	4.15		
901120	50	150.4	21	1422	4.24	11.7	20.0
901120	60	112.8	15	1429	4.02		
901120	70	75.7	10	1435	4.02		
901120	75					11.9	19.8
901120	80	53.5	7	1433	3.95		
901120	90	38.8	5	1425			
901120	100					11.6	19.5
901205	0	1141.0	112	1367		9.4	21.5
901205	10	578.1	69	1379	7.57		
901205	20	267.9	35	1379	7.58		
901205	25					9.6	21.5
901205	30	134.6	18	1377	7.32		
901205	40	72.9	10	1379	7.02		
901205	50	34.1	5	1386	7.15	9.5	22.0
901205	60	16.1	2	1391	7.21		
901205	70	9.1	1	1386	6.99		
901205	75					9.6	22.0
901205	80	5.1	1	1383	6.84		
901205	90	2.8	0	1380	6.78		
901205	100					9.7	22.0
901217	0	1106.0	101	1222		13.2	17.5
901217	10	751.8	91	1234	3.82		
901217	20	517.4	73	1247	3.74		
901217	25					13.1	17.0
901217	30	372.9	58	1249	3.55		
901217	40	268.5	44	1240	3.45		
901217	50	179.6	30	1225	3.53	13.3	16.8
901217	60	131.2	22	1323	3.59		
901217	70	95.7	16	1250	3.46		
901217	75					13.6	16.0
901217	80	65.6	10	1265	3.51		
901217	90	52.0	8	1268			
901217	100					13.5	15.8

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen (mg L^{-1})	Temp. (^0C)
910109	0	812.6	93	1083		9.9	20.2
910109	10	402.0	50	952	5.66		
910109	20	211.5	29	910	5.76		
910109	25					10.3	20.2
910109	30	99.9	14	733	5.60		
910109	40	53.6	8	688	5.59		
910109	50	46.4	7	954	5.41	10.3	20.3
910109	60	27.0	4	998	5.47		
910109	70	15.2	2	897	5.35		
910109	75					10.3	20.0
910109	80	14.2	2	1381	5.32		
910109	90	3.5	1	1460			
910109	100						20.0
910123	0	911.7	83	1191		13.6	14.0
910123	10	509.0	61	1119	4.95		
910123	20	316.9	48	1254	5.27		
910123	25					13.6	14.0
910123	30	267.9	41	1560	4.80		
910123	40	171.6	26	1563	4.72		
910123	50	88.6	14	1391	4.86	13.6	14.0
910123	60	53.0	8	1346	4.85		
910123	70	43.5	7	1820	4.87		
910123	75					13.4	13.2
910123	80	23.0	3	1531	4.85		
910123	90			1364			
910123	100						12.8
910206	0	716.3	53	918		12.2	18.0
910206	10	350.5	37	864	6.26		
910206	20	185.2	23	844	6.12		
910206	25					12.3	18.0
910206	30	191.8	24	1365	5.55		
910206	40	141.7	18	1510	5.17		
910206	50	67.6	9	1415	5.48	12.3	18.0
910206	60	36.9	5	1341	5.48		
910206	70	22.1	3	1306	5.39		
910206	75					12.2	17.5
910206	80	13.4	2	1494	5.51		
910206	90	8.6	1	1517	5.41		
910206	100	4.4	0	1652		11.9	17.5
910301	0	1448.0	121	1716		12.0	18.0
910301	10	289.8	30	787	8.11		
910301	20	246.8	28	1443	7.85		
910301	25					12.2	18.0
910301	30	62.3	7	801	7.87		
910301	40	86.4	9	2006	7.38		
910301	50	29.8	3	2121	8.13	12.2	18.0

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen ($mg\ L^{-1}$)	Temp. (^0C)
910301	60	7.4	1	1430	8.39		
910301	70	4.0	1	2020	8.53		
910301	75					12.2	17.5
910301	80	2.7	0	2079	8.03		
910301	90	1.2	0	2025	8.03	12.2	17.0
910301	100				8.03		
910302	0	438.8	38	549		11.5	18.5
910302	10	230.4	23	620	7.53		
910302	20	104.1	12	574	7.27		
910302	25					11.7	19.0
910302	30	47.0	7	545	7.25		
910302	40	30.4	4	616	6.84		
910302	50	19.5	3	910	7.13	11.7	18.8
910302	60	23.3	3	1761	6.75		
910302	70	14.3	2	2067	6.72		
910302	80	7.9	1	2033	6.60		
910302	90	0.8	0	1342	7.74	11.7	18.8
910303	0	144.9	11	214		7.6	18.2
910303	10	29.3	2	226	16.52		
910303	20	5.7	1	273	17.08		
910303	25					7.6	18.2
910303	30	0.7	0	235	17.87		
910303	40	0.3	0	241	15.92		
910303	50	0.0	0	260			
910303	75					7.5	18.2
910303	90					7.4	18.2
910320	0	1665.0	104	2017		14.5	18.0
910320	10	1060.0	85	1997	4.67		
910320	20	471.0	46	1994	6.30		
910320	25					14.8	18.0
910320	30	303.8	32	1962	5.59		
910320	40	192.3	23	2028	5.39		
910320	50	126.6	15	2041	5.16	14.7	18.0
910320	60	108.3	12	1727	4.29		
910320	70	65.7	8	1788	4.44		
910320	75					13.1	17.0
910320	80	39.6	5	1733	4.47		
910320	90	24.9	3	1921	4.61		
910320	100			1812	5.09	12.6	17.5
910403	0	1225.0	94	1561		11.2	20.5
910403	10	181.4	21	948	13.77		
910403	20	71.6	9	1146	12.44		
910403	25					11.6	20.5
910403	30	17.2	2	1087	12.84		

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen (mg L^{-1})	Temp. (^0C)
910403	40	3.7	0	827	12.79		
910403	50	0.9	0	812	13.06	11.6	20.5
910403	60	0.4	0	976	12.72		
910403	70	0.1	0	896	12.76		
910403	75					11.6	19.5
910403	80						
910403	90						
910403	100					11.4	19.5
910419	0	317.1	22	404		12.7	26.5
910419	10	188.7	19	440	5.74		
910419	20	121.4	15	466	5.29		
910419	25					12.7	26.5
910419	30	77.6	10	495	5.18		
910419	40	55.5	7	524	4.87		
910419	50	31.9	4	547	5.08	12.3	26.0
910419	60	22.9	3	568	4.84		
910419	70	13.8	2	578	4.89		
910419	75					8.6	24.5
910419	80	7.9	1	566	4.94		
910419	90	4.4	1	550	5.03		
910419	100	1.0	0	530		8.1	24.2
910516	0	1785.0	188	2118		10.8	33.2
910516	10	1374.0	186	2121	2.36	10.9	33.2
910516	20	995.3	172	2120	2.63	11.2	33.0
910516	30	660.0	137	2120	3.02	12.0	32.5
910516	40	469.3	105	2122	3.09	13.1	31.5
910516	50	299.7	70	2122	3.35	13.5	29.0
910516	60	216.3	51	2120	3.33	13.2	28.5
910516	70	147.3	35	2120	3.41	12.8	28.2
910516	80	106.8	25	2118	3.38	12.6	28.0
910516	90	68.2	16	2121	3.50	11.8	27.5
910516	100	45.8	11	2118		11.4	27.2
910516	110					10.7	27.2
910516	120					8.8	27.0
910516	125					5.0	27.0
910626	0	1688.0	153	2110		10.3	30.2
910626	10	1235.0	150	2113	2.86	10.3	30.2
910626	20	951.4	135	2097	2.61	10.4	30.2
910626	30	664.6	110	2097	2.87	10.4	30.0
910626	40	431.2	80	2098	3.19	10.5	30.0
910626	50	322.6	60	2093	3.12	11.0	29.0
910626	60	242.0	47	2093	3.07	10.9	28.0
910626	70	174.9	35	2095	3.09	10.8	28.0
910626	80	121.3	25	2095	3.16	10.0	27.5
910626	90	87.1	17	2087	3.18	9.6	27.0
910626	100	54.7	11	2091	3.33	8.3	26.8

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen (mg L^{-1})	Temp. (^0C)
910626	110	35.6	7	2083	3.41	7.0	27.0
910626	120	24.3	4	2070	3.46	6.9	27.0
910626	130	6.6	1	2073	4.23	6.6	26.8
910723	0	1680.0	153	1886		12.5	33.0
910723	10	1177.0	140	1884	3.30	11.8	33.0
910723	20	809.2	110	1851	3.36	12.1	33.0
910723	30	605.2	89	1809	3.10	12.1	33.0
910723	40	387.0	64	1824	3.42	11.8	32.5
910723	50	286.3	52	1868	3.36	11.7	32.5
910723	60	210.4	38	1872	3.32	11.8	31.5
910723	70	147.1	28	1866	3.34	12.0	31.5
910723	80	101.8	19	1867	3.39	12.6	31.0
910723	90	71.4	13	1865	3.41	12.0	31.0
910723	100	49.7	9	1857	3.43	12.2	30.5
910723	110	27.9	5	1853	3.65	10.5	28.5
910723	120	14.7	2	1853	3.91	10.0	28.0
910723	130	0.0	0	1829		8.1	28.0
910724	0	1750.0	133	1969		10.8	32.0
910724	10	1074.0	124	1965	4.66	10.6	32.0
910724	20	711.1	94	1960	4.30	10.5	32.0
910724	30	453.6	74	1550	3.50	10.7	32.0
910724	40	271.6	48	1940	4.44	10.9	31.8
910724	50	175.7	30	1936	4.42	11.1	31.2
910724	60	118.8	21	1940	4.34	11.0	31.0
910724	70	75.1	12	1949	4.39	11.1	31.0
910724	80	54.7	9	1939	4.24	10.8	30.5
910724	90	34.5	6	1935	4.27	10.4	30.5
910724	100	22.4	4	1924	4.26	10.1	30.0
910724	110	10.1	2	1920	4.60	9.0	30.0
910724	120	4.7	0	1935	4.98	8.2	30.0
910724	130						
910725	0	1513.0	119	1809		8.2	29.0
910725	10	779.0	76	1810	7.22	8.4	29.0
910725	20	448.2	51	1817	6.31	8.3	29.0
910725	30	221.4	29	1821	6.51	8.3	29.0
910725	40	126.9	16	1818	6.28	8.2	29.0
910725	50	86.3	12	1806	5.77	8.2	29.0
910725	60	52.5	7	1815	5.65	8.1	29.0
910725	70	30.9	4	1812	5.59	8.1	29.0
910725	80	18.3	2	1814	5.55	8.0	29.0
910725	90	10.8	2	1827	5.52	7.9	29.0
910725	100	6.8	1	1828	5.44	7.7	29.0
910725	110	3.9	0	1824	5.45	7.6	29.0
910725	120	2.5	0	1828	5.41	7.4	29.0
910725	130					7.1	29.0

DATE (YYMMDD)	DEPTH (cm)	down-welling ($\mu Em^{-2}s^{-1}$)	up-welling ($\mu Em^{-2}s^{-1}$)	deck ($\mu Em^{-2}s^{-1}$)	Absorption Coefficient (K_t)	Dissolved Oxygen (mg L^{-1})	Temp. (^0C)
910726	0	2087.0	175	2366		12.3	31.2
910726	10	1345.0	156	2399	4.40	12.3	31.2
910726	20	936.0	125	2415	3.97		
910726	25					12.3	31.0
910726	30	580.7	85	2420	4.21		
910726	40	385.7	59	2444	4.19		
910726	50	270.3	42	2460	4.07	12.8	30.5
910726	60	171.5	27	2466	4.15		
910726	70	113.5	19	2476	4.15		
910726	75					12.9	29.5
910726	80	77.9	12	2506	4.12		
910726	90	53.5	8	2571	4.11		
910726	100	34.6	5	2524	4.12	10.3	29.0
910726	110	21.0	3	2372	4.15		
910726	120	8.9	1	1947	4.39	10.1	29.0
910726	130						

APPENDIX B - Lake Apopka light primary productivity, and chlorophyll for light and dark bottle oxygen method.

Key: Treatments are initial, dark, final and 0, 1, 2, 4, and 6 neutral density screens. Depth equivalent is the calculated depth of Iz for each treatment (except inital and final which were taken at 0.30 m). D.O. means and S. D. represent the average and standard deviation of triplicate dissolved oxygen measurements. Gross and net production (GP and NP) were calculated from dissolved oxygen data. Community respiration (CR) is equal to the value for NP in the dark treatment. See text (Chapter 2) for methods.

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
900207	INITIAL	0.30	0.000	11.00	0.09				
900207	DARK	1.83	0.000	9.78	0.22		-0.151		-1.22
900207	6.00	1.35	0.014	10.80	0.21	0.106	-0.020	1.02	-0.20
900207	4.00	0.71	0.184	11.58	0.18	0.187	0.060	1.80	0.58
900207	2.00	0.48	0.453	12.18	0.07	0.250	0.123	2.40	1.19
900207	FINAL	0.30	0.942	11.88	0.08	0.219	0.092	2.10	0.89
900207	1.00	0.25	1.133	12.07	0.17	0.238	0.112	2.29	1.07
900207	0.00	0.05	2.535	11.70	0.04	0.199	0.073	1.92	0.70
NOTE:	quantum flux for incubation period= 9.27 MOL/M2, DURATION= 3 HOURS, CHLA = 53 MG/M3								
900221	INITIAL	0.00	0.000	10.00	0.09				
900221	DARK	0.70	0.000	7.47	0.60		-0.317		-2.54
900221	6.00	0.57	0.016	8.62	0.51	0.121	-0.143	1.16	-1.38
900221	4.00	0.31	0.185	10.81	0.30	0.349	0.085	3.35	0.81
900221	FINAL	0.30	0.211	8.34	0.07	0.091	-0.172	0.88	-1.66
900221	2.00	0.22	0.443	13.50	0.25	0.629	0.365	6.04	3.51
900221	1.00	0.13	1.073	14.68	0.01	0.752	0.488	7.23	4.69
900221	0.00	0.05	2.336	13.80	0.12	0.660	0.396	6.34	3.81
Note:	quantum flux for incubation period= 11.33 MOL/M2, DURATION= 3 HOURS, CHLA = 151 MG/M3								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
900307	INITIAL	0.00	0.000	10.69	0.27				
900307	DARK	0.69	0.000	8.42	0.08		-0.180		-0.90
900307	6	0.52	0.021	9.46	0.17	0.068	-0.081	0.41	-0.49
900307	FINAL	0.30	0.156	9.76	0.02	0.088	-0.061	0.53	-0.37
900307	4	0.29	0.177	12.15	0.17	0.246	0.096	1.48	0.58
900307	2	0.21	0.377	15.33	0.39	0.455	0.305	2.74	1.84
900307	1	0.12	0.813	12.71	0.26	0.282	0.133	1.70	0.80
900307	0	0.05	1.598	13.51	0.04	0.335	0.185	2.02	1.12
Note:	quantum flux for incubation period= 12.09 MOL/M2, DURATION= 4.75 HOURS, CHLA= 166 MG/M3								
900321	INITIAL	0.00	0.000	10.21	0.24				
900321	DARK	0.90	0.000	9.81	0.05		-0.048		-0.30
900321	6	0.56	0.053	11.56	0.09	0.172	0.133	1.04	0.80
900321	4	0.31	0.428	13.87	0.43	0.401	0.361	2.41	2.17
900321	FINAL	0.30	0.461	11.92	0.17	0.209	0.169	1.26	1.02
900321	2	0.22	0.897	15.55	0.08	0.566	0.527	3.41	3.17
900321	1	0.13	1.903	15.17	0.05	0.528	0.488	3.18	2.94
900321	0	0.05	3.683	13.78	0.15	0.391	0.351	2.36	2.12
Note:	quantum flux for incubation period= 17.69 MOL/M2, DURATION= 3.17 HOURS, CHLA =161.3 MG/M3								
900404	INITIAL	0.00	0.000	7.37	0.08				
900404	DARK	0.65	0.000	6.91	0.03		-0.051		-0.29
900404	6	0.41	0.051	8.67	0.13	0.195	0.144	1.11	0.82
900404	FINAL	0.30	0.181	10.29	0.07	0.373	0.323	3.59	3.10
900404	4	0.23	0.385	11.00	0.21	0.452	0.402	2.57	2.28
900404	2	0.17	0.791	14.15	0.66	0.800	0.749	4.54	4.25
900404	1	0.11	1.643	14.07	0.21	0.791	0.740	4.49	4.20
900404	0	0.05	3.121	13.64	0.02	0.744	0.693	4.23	3.94
Note:	quantum flux for incubation period= 15.6 MOL/M2, DURATION = 2.83 HOURS, CHLA= 176.3 MG/M3								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
900418	INITIAL	0.00	0.000	13.03	0.18				
900418	DARK	2.41	0.000	11.45	0.06		-0.200		-4.03
900418	6	1.38	0.075	12.07	0.01	0.064	-0.100	1.28	-2.02
900418	4	0.73	0.590	12.38	0.15	0.097	-0.067	1.95	-1.35
900418	2	0.49	1.232	12.61	0.00	0.121	-0.044	2.43	-0.89
900418	FINAL	0.30	2.271	13.34	0.03	0.200	0.033	4.02	0.66
900418	1	0.26	2.599	12.06	0.06	0.063	-0.101	1.27	-2.03
900418	0	0.05	5.008	11.97	0.15	0.054	-0.110	1.09	-2.22
Note:	quantum flux for incubation period = 17.6 mol/m2, duration= 2.95 hours, chla = 49.7 mg/m3								
900504	INITIAL	0.00	0.000	8.43	0.10				
900504	DARK	1.50	0.000	8.11	0.46		-0.041		-0.33
900504	6	0.79	0.025	8.76	0.34	0.068	0.034	0.55	0.28
900504	4	0.42	0.259	10.56	0.07	0.256	0.222	2.09	1.81
900504	FINAL	0.30	0.583	10.47	0.37	0.246	0.212	2.01	1.74
900504	2	0.30	0.598	12.04	0.05	0.410	0.376	3.35	3.07
900504	1	0.17	1.400	12.40	0.38	0.447	0.413	3.66	3.38
900504	0	0.05	2.953	11.59	0.15	0.363	0.329	2.97	2.69
Note:	quantum flux for incubation period = 12.257 mol/m2, duration = 3.0 h, chl-a = 122.3 mg/m3								
900516	INITIAL	0.00	0.000	9.03	0.11				
900516	DARK	1.39	0.000	6.57	0.23		-0.307		-2.98
900516	6	0.81	0.061	7.87	0.48	0.135	-0.121	1.31	-1.17
900516	4	0.43	0.501	9.33	0.29	0.287	0.032	2.79	0.31
900516	2	0.30	1.058	10.56	0.56	0.415	0.159	4.03	1.55
900516	FINAL	0.30	1.069	10.31	0.18	0.389	0.133	3.78	1.29
900516	1	0.17	2.262	10.28	0.24	0.386	0.130	3.75	1.27
900516	0	0.05	4.410	10.01	0.16	0.357	0.102	3.47	0.99
Note:	quantum flux for incubation period= 17.57 mol/m2, duration= 3 hours, chla =103.0 mg/m3								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
900530	INITIAL	0.30	0.00	9.86	0.05				
900530	DARK	1.26	0.00	8.41	0.33		-0.211		-1.93
900530	6	0.78	0.06	9.63	0.05	0.150	-0.030	1.37	-0.27
900530	4	0.42	0.49	11.78	0.12	0.410	0.230	3.75	2.11
900530	2	0.29	1.05	12.78	0.16	0.530	0.350	4.85	3.21
900530	1	0.16	2.27	12.64	0.10	0.510	0.340	4.67	3.11
900530	0	0.05	4.48	12.44	0.20	0.490	0.310	4.49	2.84
900530	final	0.30	1.01	12.05	0.08	0.44	0.27	4.04	2.43
Note:	quantum flux for incubation period = 15.57 , duration = 2.58 h, chl-a = 109.2 ug/l.								
900613	INITIAL	0.00	0.000	8.77					
900613	DARK	0.98	0.000	7.86	0.03		-0.128		-0.84
900613	6	0.62	0.061	8.88	0.02	0.120	0.013	0.79	0.09
900613	4	0.34	0.534	11.20	0.67	0.394	0.287	2.59	1.89
900613	FINAL	0.30	0.722	10.06	0.36	0.259	0.152	1.70	1.00
900613	2	0.24	1.153	12.44	0.23	0.539	0.433	3.55	2.85
900613	1	0.14	2.517	12.31	0.47	0.525	0.418	3.45	2.75
900613	0	0.05	5.000	12.14	0.34	0.504	0.397	3.31	2.61
Note:	quantum flux for incubation period = 19.51 mol/m2(estimated), duration = 2.65 h, chl-a =152 ug/l								
900627	INITIAL	0.30	0.000	9.69	0.05				
900627	DARK	1.22	0.000	8.26	0.00		-0.290		-5.06
900627	6	1.08	0.016	8.98	0.13	0.119	-0.117	2.08	-2.05
900627	4	0.57	0.214	10.32	0.08	0.341	0.104	5.94	1.81
900627	2	0.39	0.542	11.47	0.01	0.531	0.294	9.27	5.14
900627	FINAL	0.30	0.876	11.00	0.03	0.462	0.221	8.07	3.85
900627	1	0.21	1.395	11.91	0.09	0.604	0.367	10.54	6.41
900627	0	0.05	3.200	11.72	0.02	0.572	0.335	9.98	5.85
Note:	quantum flux for incubation period= 7.67 mol/m2, duration= 1.85 hours, chla = 57.3 mg/m3								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
900718	INITIAL	0.30	0.000	10.51	0.07				
900718	DARK	0.91	0.000	9.43	0.07		-0.199		-1.51
900718	6	0.58	0.045	10.33	0.94	0.139	-0.027	1.05	-0.21
900718	4	0.32	0.370	14.33	0.53	0.755	0.589	5.74	4.48
900718	FINAL	0.30	0.434	n/a		-1.451	-1.617	-11.03	-12.29
900718	2	0.23	0.781	16.68	0.25	1.116	0.950	8.48	7.22
900718	1	0.13	1.666	16.29	0.14	1.056	0.890	8.02	6.76
900718	0	0.05	3.242	15.50	0.36	0.935	0.769	7.11	5.85
Note:	quantum flux for incubation period= 9.839 mol/m2, duration =2.03 hours, chla =131.6 mg/m3								
900801	INITIAL	0.30	0.000	9.28	0.02				
900801	DARK	2.07	0.000	8.21	0.23		-0.182		-1.74
900801	6	1.27	0.067	10.67	0.01	0.346	0.195	3.30	1.85
900801	4	0.67	0.595	10.85	0.12	0.372	0.220	3.54	2.09
900801	2	0.46	1.294	10.20	0.09	0.281	0.129	2.67	1.23
900801	FINAL	0.30	2.288	10.69	0.09	0.350	0.198	3.33	1.88
900801	1	0.24	2.848	9.08	0.21	0.124	-0.028	1.18	-0.27
900801	0	0.05	5.697	10.15	0.13	0.273	0.121	2.60	1.16
Note:	quantum flux for incubation period = 15.18 mol/m2(estimated), duration = 2.22 h, Chl-a = 72.4 mg/m3..								
900822	INITIAL	0.30	0.000	9.83	0.07				
900822	DARK	2.34	0.000	8.02	0.68		-0.317		-4.40
900822	6	1.47	0.061	9.38	0.14	0.199	-0.065	2.76	-0.91
900822	4	0.77	0.566	10.23	0.15	0.321	0.058	4.46	0.80
900822	2	0.52	1.248	10.76	0.08	0.399	0.136	5.55	1.88
900822	FINAL	0.30	2.553	10.20	0.85	0.317	0.053	4.41	0.74
900822	1	0.27	2.790	10.60	0.10	0.376	0.112	5.22	1.55
900822	0	0.05	5.654	10.09	0.05	0.302	0.038	4.19	0.53
Note:	quantum flux for incubation period = 14.25 mol/m2(estimated), duration = 2.15 h, Chl-a = 74.4 mg/m3.								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (*mgC*/ mgChla/h)	NP/CHLA (*mgC*/ mgChla/h)
900906	INITIAL	0.30	0.000	8.95	0.41				
900906	DARK	1.42	0.000	7.90	0.10		-0.190		-2.25
900906	6	0.95	0.031	9.82	0.82	0.288	0.130	3.42	1.55
900906	4	0.51	0.334	10.37	0.08	0.371	0.213	4.40	2.52
900906	2	0.35	0.772	10.85	0.16	0.442	0.284	5.25	3.38
900906	FINAL	0.30	1.010	10.51	0.17	0.392	0.234	4.65	2.78
900906	1	0.19	1.810	11.33	0.46	0.515	0.357	6.12	4.25
900906	0	0.05	3.827	10.18	0.13	0.342	0.184	4.06	2.19
Note:	quantum flux for incubation period = 10.39 mol/m2(estimated), duration = 2.08 h, Chl-a = 62.1* mg/m3.								
900920	INITIAL	0.30	0.000	8.29	0.06				
900920	DARK	1.15	0.000	6.28	0.09		-0.362		-7.86
900920	6	0.83	0.036	6.94	0.64	0.099	-0.203	2.14	-4.41
900920	4	0.45	0.257	6.60	0.24	0.047	-0.254	1.03	-5.52
900920	2	0.31	0.517	8.34	0.36	0.309	0.008	6.73	0.18
900920	FINAL	0.30	0.543	8.50	0.57	0.333	0.032	7.24	0.69
900920	1	0.17	1.052	8.97	0.38	0.403	0.102	8.77	2.22
900920	0	0.05	1.963	8.44	0.48	0.325	0.024	7.06	0.51
NOTE:	quantum flux for incubation period= 5.28 MOL/M2, DURATION= 2.08 HOURS, CHLA= 46.0 MG/M3								
901017	INITIAL	0.30	0.000	7.52	0.07				
901017	DARK	1.26	0.000	6.91	0.07		-0.102		-1.10
901017	6	0.76	0.044	7.47	0.48	0.078	-0.007	0.85	-0.07
901017	4	0.41	0.304	8.94	0.12	0.282	0.197	3.05	2.13
901017	FINAL	0.30	0.563	9.05	0.07	0.227	0.162	2.45	1.75
901017	2	0.29	0.606	10.18	0.13	0.454	0.369	4.91	3.99
901017	1	0.16	1.218	9.88	0.07	0.412	0.327	4.45	3.53
901017	0	0.05	2.251	9.86	0.02	0.410	0.325	4.44	3.52
NOTE:	quantum flux for incubation period= 6.68 MOL/M2, DURATION= 2.25 HOURS, CHLA= 92.5 MG/M3								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
901107	INITIAL	0.30	0.000	10.98	0.09				
901107	DARK	1.94	0.000	10.19	0.03		-0.107		-0.69
901107	6	1.23	0.055	10.95	0.18	0.085	-0.004	0.55	-0.02
901107	4	0.65	0.489	10.27	1.21	0.009	-0.080	0.06	-0.52
901107	2	0.45	1.061	10.61	0.61	0.047	-0.042	0.30	-0.27
901107	FINAL	0.30	1.825	12.17	0.04	0.222	0.133	1.44	0.86
901107	1	0.23	2.329	10.30	1.14	0.012	-0.077	0.08	-0.50
901107	0	0.05	4.648	11.21	0.08	0.114	0.026	0.74	0.17
Note:	quantum flux for incubation period = 15.58 mol/m2 (estimated), duration = 2.78 h, Chl-a 112.9* mg/m3.								
901120	INITIAL	0.30	0.000	11.78	0.05				
901120	DARK	1.74	0.000	11.01	0.07		-0.092		-1.06
901120	6	1.07	0.062	11.72	0.13	0.071	-0.006	0.81	-0.07
901120	4	0.57	0.490	12.46	0.15	0.146	0.069	1.67	0.79
901120	2	0.39	1.022	12.63	0.11	0.163	0.086	1.87	0.99
901120	FINAL	0.30	1.477	12.56	0.05	0.156	0.079	1.79	0.91
901120	1	0.21	2.154	12.24	0.04	0.124	0.047	1.42	0.54
901120	0	0.05	4.148	11.77	0.02	0.076	-0.001	0.87	-0.01
Note:	quantum flux for incubation period = 15.81 mol/m2 (estimated), duration = 3.10 h, Chl-a = 82.7 mg/m3.								
901205	INITIAL	0.30	0.000	10.46	0.07				
901205	DARK	0.98	0.000	9.95	0.08		-0.089		-2.13
901205	6	0.66	0.039	10.67	0.16	0.106	0.032	2.54	0.76
901205	4	0.36	0.313	11.76	0.10	0.267	0.193	6.37	4.60
901205	FINAL	0.30	0.471	10.89	0.20	0.139	0.065	3.32	1.54
901205	2	0.25	0.654	11.75	0.10	0.264	0.190	6.31	4.54
901205	1	0.14	1.384	11.27	0.30	0.194	0.120	4.63	2.86
901205	0	0.05	2.673	10.77	0.07	0.121	0.046	2.88	1.11
Note:	quantum flux for incubation period= 8.02 mol/m2, duration= 2.12 h, chl-a= 41.9 mg/m3								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (*mgC*/ mgChla/h)	NP/CHLA (*mgC*/ mgChla/h)
901217	INITIAL	0.30	0.000	13.72	0.14				
901217	DARK	1.97	0.000	13.00	0.08		-0.131		-2.54
901217	6	1.23	0.044	13.41	0.03	0.062	-0.047	1.20	-0.92
901217	4	0.65	0.343	13.69	0.10	0.104	-0.005	2.02	-0.09
901217	2	0.44	0.710	13.59	0.12	0.090	-0.020	1.74	-0.38
901217	FINAL	0.30	1.177	13.94	0.07	0.142	0.033	2.75	0.63
901217	1	0.23	1.484	13.38	0.29	0.057	-0.052	1.10	-1.01
901217	0	0.05	2.838	13.44	0.04	0.066	-0.043	1.29	-0.83
Note:	quantum flux for incubation period = 7.04 mol/m2, duration = 2.08 h, chl-a= 51.6 mg/m3								
910109	INITIAL	0.30	0.000	10.22	0.71				
910109	DARK	1.21	0.000	9.06	0.02		-0.215		-2.95
910109	6	0.80	0.032	9.76	0.01	0.109	-0.071	1.49	-0.97
910109	4	0.43	0.252	10.89	0.13	0.285	0.105	3.90	1.44
910109	2	0.30	0.523	11.69	0.03	0.408	0.228	5.58	3.12
910109	FINAL	0.30	0.526	11.39	0.06	0.362	0.182	4.95	2.49
910109	1	0.17	1.096	11.40	0.19	0.364	0.185	4.98	2.52
910109	0	0.05	2.099	11.13	0.11	0.322	0.142	4.40	1.95
Note:	quantum flux for incubation period = 5.59 mol/m2, duration = 2.01 h, chl-a = 73.1 mg/m3								
910123	INITIAL	0.30	0.000	12.42	0.07				
910123	DARK	1.39	0.000	12.09	0.48		-0.057		-0.88
910123	6	0.93	0.026	12.25	0.04	0.023	-0.025	0.35	-0.38
910123	4	0.50	0.215	13.25	0.18	0.169	0.122	2.60	1.87
910123	2	0.34	0.455	13.29	0.10	0.176	0.128	2.70	1.97
910123	FINAL	0.30	0.561	13.33	0.03	0.181	0.134	2.79	2.05
910123	1	0.19	0.976	12.91	0.08	0.119	0.072	1.83	1.10
910123	0	0.05	1.907	12.66	0.09	0.083	0.035	1.27	0.54
NOTE:	quantum FLUX for incubation period = 5.187 MOL/M2, DURATION = 2.13 H, CHL-A = 65.1 MG/M3								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
910206	INITIAL	0.30	0.000	11.73	0.04				
910206	DARK	1.60	0.000	11.07	0.39		-0.121		-1.89
910206	6	0.76	0.070	11.79	0.16	0.110	0.009	1.71	0.14
910206	4	0.41	0.480	12.94	0.05	0.284	0.183	4.41	2.84
910206	FINAL	0.30	0.886	12.09	0.04	0.155	0.054	2.42	0.84
910206	2	0.29	0.953	13.60	0.10	0.385	0.284	5.98	4.41
910206	1	0.16	1.914	13.13	0.22	0.312	0.211	4.86	3.29
910206	0	0.05	3.531	12.51	0.07	0.220	0.118	3.41	1.84
Note:	quantum flux for incubation period = 9.59 mol/m2(estimated), duration = 2.06 h, Chl-a = 64.3 mg/m3.								
910301A	INITIAL	0.30	0.000	10.78	0.15				
910301A	DARK	0.91	0.000	10.17	0.11		-0.073		-0.49
910301A	6	0.60	0.025	10.87	0.10	0.070	0.009	0.47	0.06
910301A	4	0.33	0.221	12.49	0.25	0.235	0.174	1.58	1.17
910301A	FINAL	0.30	0.285	12.73	0.08	0.260	0.199	1.75	1.34
910301A	2	0.23	0.481	13.66	0.06	0.354	0.293	2.38	1.97
910301A	1	0.14	1.059	13.62	0.17	0.350	0.289	2.35	1.94
910301A	0	0.05	2.118	13.11	0.07	0.298	0.237	2.00	1.59
Note:	quantum flux for incubation period= 9.757 mol/m2, duration= 3.08 hours, chla= 148.7 mg/m3								
910301B	INITIAL	0.30	0.000	12.73	0.08				
910301B	DARK	0.91	0.000	11.79	0.07		-0.106		-0.71
910301B	6	0.60	0.036	12.71	0.21	0.086	-0.002	0.58	-0.01
910301B	4	0.33	0.324	14.51	0.08	0.254	0.166	1.71	1.12
910301B	FINAL	0.30	0.416	na					
910301B	2	0.23	0.704	15.30	0.21	0.328	0.240	2.20	1.61
910301B	1	0.14	1.549	13.46	0.37	0.156	0.068	1.05	0.46
910301B	0	0.05	3.098	13.58	0.17	0.167	0.079	1.13	0.53
Note:	quantum flux for incubation period=15.506 mol/m2, duration=3.35 hours, chla= 148.7 mg/m3								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
910320	INITIAL	0.00	0.000	14.40	0.06				
910320	DARK	1.46	0.000	12.52	0.06		-0.287		-2.76
910320	6	0.90	0.040	13.33	0.12	0.103	-0.136	0.99	-1.31
910320	4	0.48	0.337	14.75	0.09	0.285	0.046	2.74	0.44
910320	2	0.33	0.719	15.28	0.06	0.352	0.113	3.38	1.08
910320	FINAL	0.30	0.857	15.01	0.03	0.318	0.079	3.06	0.76
910320	1	0.18	1.554	14.85	0.06	0.297	0.057	2.85	0.55
910320	0	0.05	3.059	14.20	0.20	0.214	-0.026	2.05	-0.25
Note:	quantum flux for incubation period = 9.688 mol/m2, duration= 2.45 h, chl-a= 104 mg/m3								
910403	INITIAL	0.30	0.000	11.09	0.04				
910403	DARK	0.56	0.000	9.83	0.01		-0.176		-0.81
910403	6	0.39	0.025	11.38	0.00	0.181	0.034	0.83	0.16
910403	FINAL	0.30	0.080	10.79	0.20	0.111	-0.035	0.51	-0.16
910403	4	0.22	0.214	13.83	0.31	0.466	0.319	2.15	1.47
910403	2	0.16	0.457	16.15	0.08	0.737	0.590	3.39	2.72
910403	1	0.10	0.988	16.46	0.20	0.772	0.626	3.56	2.88
910403	0	0.05	1.945	15.77	0.38	0.693	0.546	3.19	2.52
Note:	quantum flux for incubation period = 9.873 mol/m2, duration = 2.68 h, chl-a= 217.1 mg/m3								
910419	INITIAL	0.30	0.000	11.22	0.08				
910419	DARK	1.15	0.000	9.75	0.06		-0.200		-1.82
910419	6	0.92	0.032	10.30	0.04	0.063	-0.104	0.57	-0.95
910419	4	0.49	0.268	11.69	0.18	0.221	0.054	2.01	0.49
910419	2	0.34	0.573	12.50	0.05	0.313	0.146	2.85	1.33
910419	FINAL	0.30	0.694	11.60		0.210	0.043	1.91	0.39
910419	1	0.18	1.238	13.10	0.17	0.381	0.213	3.46	1.94
910419	0	0.05	2.436	12.68	0.41	0.334	0.166	3.03	1.51
Note:	quantum flux for incubation period = 8.61 mol/m2, duration = 2.75 h, chl-a = 110 mg/m3.								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
910516	INITIAL	0.30	0.000	11.93	0.14				
910516	DARK	2.38	0.000	10.04	0.04		-0.269		-3.80
910516	6	1.43	0.084	10.73	0.16	0.082	-0.142	1.16	-2.01
910516	4	0.75	0.712	11.72	0.08	0.200	-0.024	2.83	-0.34
910516	2	0.51	1.521	11.68	0.27	0.195	-0.029	2.76	-0.41
910516	1	0.27	3.287	11.69	0.08	0.196	-0.028	2.78	-0.40
910516	0	0.05	6.470	11.51	0.11	0.175	-0.049	2.48	-0.70
Note:	quantum flux for incubation period = 15.88 mol/m2, duration = 2.63 h, Chl-a = 70.7 mg/m3								
910626a	INITIAL	0.30	0.00	9.58	0.02				
910626a	DARK	2.42	0.00	7.85	0.10		-0.235		-3.55
910626a	6	1.46	0.084	8.68	0.15	0.094	-0.102	1.42	-1.54
910626a	4	0.77	0.711	10.17	0.01	0.263	0.067	3.98	1.02
910626a	2	0.52	1.518	10.52	0.05	0.303	0.107	4.58	1.62
910626a	FINAL	0.30	2.998	11.36	0.06	0.398	0.202	6.01	3.06
910626a	1	0.27	3.281	10.11	0.02	0.256	0.060	3.87	0.91
910626a	0	0.05	6.459	9.56	0.00	0.194	-0.002	2.93	-0.03
Note:	total quantum flux for incubation period = 20.71 mol/m2 , duration = 2.75 h, chl-a = 66.2 mg/m3. The "a" assay run in polystyrene culture flask.								
910626b	INITIAL	0.30	0.00	9.70	0.02				
910626b	DARK	2.42	0.00	7.85	0.10		-0.251		-3.80
910626b	6	1.46	0.084	na					
910626b	4	0.77	0.711	10.77	0.01	0.331	0.122	5.00	1.84
910626b	2	0.52	1.518	11.04	0.18	0.362	0.152	5.46	2.30
910626b	final	0.30	2.998	11.36	0.06	0.398	0.189	6.01	2.85
910626b	1	0.27	3.281	10.98	0.01	0.355	0.146	5.37	2.20
910626b	0	0.05	6.459	10.28	0.09	0.275	0.066	4.16	0.99
Note:	"b" assay same as "a" assay except incubation was done in glass standard glass bottles.								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mg O_2/L)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
910724	INITIAL	0.30	0.00	11.08	0.02				
910724	DARK	1.76	0.00	9.75	0.09		-0.196		-2.09
910724	6	1.07	0.048	10.25	0.09	0.062	-0.102	0.66	-1.08
910724	4	0.57	0.405	11.16	0.06	0.173	0.010	1.85	0.11
910724	2	0.39	0.864	11.91	0.11	0.265	0.102	2.83	1.08
910724	1	0.21	1.868	11.60	0.07	0.227	0.064	2.42	0.68
910724	0	0.05	3.678	10.88	0.13	0.138	-0.025	1.48	-0.27
910724	FINAL	0.30	1.274	na				0.00	0.00
Note:	total quantum flux for incubation period = 11.594 MOL/M2 , duration = 2.55 h, chl-a = 93.7 mg/m3.								
910725a	INITIAL	0.30	0.00	6.12	0.02				
910725a	DARK	1.19	0.00	5.79	0.02		-0.041		-0.34
910725a	6	0.79	0.036	6.33	0.10	0.055	0.021	0.46	0.17
910725a	4	0.43	0.309	7.13	0.07	0.137	0.103	1.14	0.86
910725a	2	0.30	0.659	8.59	0.14	0.286	0.252	2.39	2.11
910725a	1	0.17	1.425	9.48	0.10	0.376	0.342	3.14	2.86
910725a	0	0.05	2.806	9.52	0.07	0.380	0.346	3.18	2.89
910725a	FINAL	0.30	0.650	7.19	0.20			0.00	0.00
NOTE:	Total quantum flux for incubation period = 11.54 MOL/M2 , duration = 3.07 h, chl-a = 119.5 mg/m3. Incubation time 0712h to 1017h.								
910725b	INITIAL	0.30	0.00	7.19	0.20				
910725b	DARK	1.27	0.00	6.59	0.00		-0.074		-0.62
910725b	6	0.79	0.060	8.25	0.04	0.172	0.110	1.44	0.92
910725b	4	0.43	0.507	10.57	0.08	0.411	0.349	3.44	2.92
910725b	2	0.30	1.084	11.30	0.08	0.486	0.424	4.07	3.55
910725b	1	0.17	2.343	10.77	0.04	0.432	0.370	3.61	3.09
910725b	0	0.05	4.611	9.59	0.16	0.310	0.248	2.60	2.08
910725b	FINAL	0.30	1.068	11.42	0.04			0.00	0.00
Note:	total quantum flux = 18.72 MOL/M2 , duration = 3.03 h , chl-a = 119.5 mg/m3. Incubation time 1112h to 1415h.								

DATE (YYMMDD)	TREAT-MENT	depth equiv(m)	Iz (mol/m2/h)	D.O. MEANS (mgO/l)	D.O. S.D.	GROSS PRODUCT (GP) (gC/m3/h)	NET PRODUCT (NP) (gC/m3/h)	GP/CHLA (mgC/ mgChla/h)	NP/CHLA (mgC/ mgChla/h)
910725c	INITIAL	0.30	0.00	11.42	0.04				
910725c	DARK	0.95	0.00	10.08	0.00		-0.120		-1.00
910725c	6	0.79	0.009	10.12	0.04	0.003	-0.097	0.03	-0.81
910725c	4	0.43	0.074	10.93	0.04	0.064	-0.036	0.53	-0.30
910725c	2	0.30	0.159	11.93	0.07	0.138	0.038	1.15	0.32
910725c	1	0.17	0.344	12.15	0.12	0.154	0.054	1.29	0.46
910725c	0	0.05	0.677	12.15	0.03	0.154	0.054	1.29	0.45
910725c	final	0.30	0.157	na				0.00	0.00
Note:	total quantum flux for incubation period = 3.81 MOL/M2 , duration = 4.2 h, chl-a = 119.5 mg/m3. Incubation time was 1447h to 1900h.								
910726	INITIAL	0.30	0.00	10.57	0.03				
910726	DARK	1.71	0.00	9.18	0.01		-0.134		-1.27
910726	6	1.10	0.044	9.92	0.08	0.059	-0.052	0.57	-0.50
910726	4	0.59	0.368	11.13	0.01	0.156	0.045	1.49	0.43
910726	2	0.40	0.787	12.39	0.03	0.257	0.146	2.45	1.39
910726	1	0.21	1.701	12.28	0.08	0.248	0.137	2.37	1.30
910726	0	0.05	3.349	11.81	0.14	0.211	0.100	2.01	0.95
910726	final	0.30	1.196	na				0.00	0.00
Note:	total quantum flux for incubation period = 16.05 MOL/M2 , duration = 3.9 h, chl-a = 104.9 mg/m3. Incubation time was 1447h to 1900h.								

* average of SJRWMD & regression prediction

Appendix C. Summary of data collected in Lake Apopka, February 1990 to July 1991. See following pages for key to abbreviations.

Date (ddmmyy)	Chla (mg/m3)	NPmax (mgC/ m3/hr)	GPmax (mgC/ m3/hr)	CR (mgC/ m3/hr)	Comp Pt N (m)	ANP (mgC/ m2/hr)	NPmax /CR	GPmax /CR	PBmax (mgC/ Chl/hr)	Comp Pt G (cm)	AGP (mgC/ m2/hr)	I Flux (mol/ m2)	Time (hr)	I/hr (mol/ m2/hr)	Temp (oC)	Secchi Disc (m)	Kt (m-1)	Lake Level (ft)	Dev mean (cm)
2/7/90	53.0	123	274	151	1.20	86	0.81	1.81	5.17	1.70	305	9.27	3.00	3.09	21.0		3.96	66.73	10.48
2/21/90	151.0	488	805	317	0.42	116	1.54	2.54	5.33	0.72	297	11.33	3.00	3.78	22.5		9.61	66.78	12.00
3/7/90	166.0	305	485	180	0.42	63	1.69	2.69	2.92	0.68	162	12.09	4.75	2.55	20.0		9.31	66.96	17.49
3/21/90	161.3	527	575	48	0.82	209	10.98	11.98	3.56	0.90	250	17.69	3.17	5.58	19.4		8.31	66.81	12.92
4/4/90	176.3	749	800	51	0.58	213	14.69	15.69	4.54	0.63	244	15.60	2.83	5.51	20.5	20.0	11.38	66.83	13.53
4/18/90	49.7	-44	156	200		*	-0.22	0.78	3.14			17.60	2.95	5.97	29.0	34.0	3.16	66.51	3.77
5/4/90	122.3	413	454	41	0.93	192	10.07	11.07	3.71	1.06	233	12.26	3.00	4.09	26.8	20.0	6.49	66.24	-4.47
5/16/90	103.0	159	466	307	0.50	50	0.52	1.52	4.52	1.22	314	17.57	3.00	5.86	28.0	16.5	5.67	66.05	-10.26
5/30/90	109.2	350	561	211	0.77	178	1.66	2.66	5.14	1.08	373	15.57	2.58	6.03	29.0	19.0	5.97	65.85	-16.36
6/13/90	152.0	433	561	128	0.62	184	3.38	4.38	3.69	0.72	270	19.51	2.65	7.36	28.5	18.0	7.74	66.11	-8.43
6/27/90	57.3	367	657	290	0.81	182	1.27	2.27	11.47	1.25	481	7.67	1.85	4.15	29.0	26.0	5.18	66.19	-5.99
7/18/90	131.6	950	1149	199	0.47	331	4.77	5.77	8.73	0.63	440	9.84	2.03	4.85	26.0	18.0	8.04	66.43	1.33
8/1/90	72.4	220	402	182	1.20	235	1.21	2.21	5.55	1.40	472	15.80	2.22	7.12	31.0	24.0	3.65	66.49	3.16
8/22/90	74.4	136	453	317	1.10	77	0.43	1.43	6.09	2.00	568	14.25	2.15	6.63	32.5	28.0	3.18	66.65	8.04
9/6/90	43.6	357	515	190	1.45	249	1.88	2.71	11.81	1.90	567	10.39	2.08	5.00	31.0	29.0	5.39	66.55	4.99
9/20/90	46.0	102	464	362	0.31	15	0.28	1.28	10.09	0.52	165	5.28	2.08	2.54	27.0	27.5	5.14	66.31	-2.33
10/17/90	92.5	369	471	102	0.77	116	3.62	4.62	5.09	0.92	202	6.68	2.25	2.97	28.0	31.0	5.54	66.43	1.33
11/7/90		26	133	107		3	0.24	1.24			3	15.58	2.78	5.60	24.0	36.0	3.74	66.26	-3.86
11/20/90	58.9	86	178	92	1.05	44	0.93	1.93	3.02	1.40	157	15.81	3.10	5.10	23.0	31.0	4.13	66.15	-7.21
12/5/90	171.9	193	282	89	0.77	81	2.17	3.17	1.64	0.87	154	8.02	2.12	3.78	18.0	23.5	6.94	66.18	-6.30
12/17/90	51.6	-5	126	131		*	-0.04	0.96	2.44			7.04	2.08	3.38	19.0	30.0	3.52	66.03	-10.87
1/9/91	73.1	228	443	215	0.65	87	1.06	2.06	6.06	0.95	259	5.59	2.02	2.77	21.5	28.0	5.53	65.93	-13.92
1/23/91	65.1	128	185	57	0.85	64	2.25	3.25	2.84	0.98	116	5.19	2.13	2.44	15.0	24.0	4.89	65.94	-13.62
2/6/91	64.3	284	405	121	0.80	118	2.35	3.35	6.30	0.93	223	9.59	2.06	4.66	19.5	28.5	5.53	65.96	-13.01
3/1/91	148.7	293	366	73	0.63	108	4.01	5.01	2.46	0.68	156	9.76	3.08	3.17	17.0		8.03	65.74	-19.72
3/1/91	148.7	240	346	106	0.60	65	2.26	3.26	2.33	0.72	135	15.51	3.35	4.63	17.0		8.03	65.74	-19.72
3/20/91	104.0	113	352	287	0.60	29	0.39	1.23	3.38	1.13	277	9.67	2.45	3.95	20.5	28.0	5.09	66.28	-3.25
4/3/91	217.1	626	802	176	0.42	151	3.56	4.56	3.69	0.45	228	9.87	2.68	3.68	20.5	17.0	12.77	66.26	-3.86
4/19/91	110.0	213	413	200	0.62	75	1.07	2.07	3.75	0.95	232	8.61	2.75	3.13	28.0	23.0	5.02	66.44	1.63
5/16/91	70.7	-24	245	269		*	-0.09	0.91	3.47			15.88	2.63	6.04	32.2	25.0	3.14	66.58	5.90
6/26/91	66.2	107	342	235	1.05	59	0.46	1.46	5.17	1.85	400	20.71	2.75	7.53	30.0	27.0	3.07	66.92	16.27
7/24/91	93.7	102	298	196	0.62	27	0.52	1.52	3.18	1.15	201	11.59	2.55	4.55	32.0	31.0	4.24	66.94	16.88
7/25/91	119.5	424	498	74	0.95	252	5.73	6.73	4.17	1.10	328	18.72	3.03	6.18	29.0	23.0	5.85	66.94	16.88
7/26/91	104.9	146	280	134	0.82	74	1.09	2.09	2.67	1.15	206	16.05	3.90	4.12	31.0	28.0	4.12	66.93	16.58
Avg.	103.9	270.1	439.5	171.7	0.76	120.4	2.55	3.54	4.76	1.05	271.5	12.4	2.68	4.64	24.9	25.5	5.92	66.39	0.00
Std. Dev.	45.4	217.0	215.8	87.2	0.26	80.6	3.29	3.29	2.47	0.40	129.9	4.38	0.59	1.44	5.15	5.13	2.36	0.367	11.19
CV (%)	43.6	80.3	49.1	50.8	34.8	66.9	129.2	93.1	52.0	37.7	47.8	35.3	22.0	31.0	20.7	20.1	39.9	0.6	

Key to Abbreviations for Variables

Date of sampling is given as month (mm)/day (dd)/year (yy).

Chl a refers to concentration of chlorophyll *a* in mg m^{-3}.

NPmax is maximum net primary production (mg C m^{-3} h^{-1}).

GPmax is maximum gross primary production (mg C m^{-3} h^{-1}).

CR is community respiration (mg C m^{-3} h^{-1}).

Comp Pt (N) is the compensation depth for net photosynthesis (m).

ANP is areal net production (mg C m^{-2} h^{-1}), an integration of the rate of net primary production to the compensation depth.

NPmax/CR is the ratio (dimensionless) of maximum net primary production to community respiration.

GPmax/CR is the ratio (dimensionless) of maximum gross primary production to community respiration.

PBmax (PB_{max}) is the maximum rate of gross primary production normalized to the chlorophyll concentration (GPmax/chl *a*). It is a biomass specific rate of production or assimilation number (mg C mg chl a^{-1} h^{-1}).

Comp Pt (G) is the compensation depth for gross photosynthesis (m).

AGP is areal gross production (mg C m^{-2} h^{-1}), an integration of the rate of gross primary production to the compensation depth.

I flux is the quanta of incident PAR received during the incubation period for primary production measurements (mol m^{-2}). Quanta of light are measured in units of moles or Einsteins (E).

Time is the incubation period for primary production measurements (h).

I/h is the average hourly PAR during the incubation period for primary production measurements (mol m^{-2} h^{-1}).

Temp refers to water temerature in °C (Celsius).

Secchi Disc is water transparency measured with a Secchi disc (m).

K_t is the extinction coefficient (ln units) of photosynthetically active radiation (PAR) in water.

Lake level is the elevation of the lake surface above mean sea level (ft).

Dev mean is the deviation of the lake surface (cm) from the mean lake level obtained from data collected from 1989-1991. 1.0 inch equals 2.54 centimeters (cm).

Avg. is the arithmetic mean of each variable.

Std. Dev. is the standard deviation of the mean for each variable.

CV(%) is the coefficient of variation of the standard deviation divided by the average and multiplied by 100 to obtain a percentage.