THE EFFECT OF NUTRIENT ENRICHMENT ON PRIMARY PRODUCTION IN MICHIGAN FARM PONDS

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ABSTRACT

THE EFFECT OF NUTRIENT ENRICHMENT ON PRIMARY
PRODUCTION IN MICHIGAN FARM PONDS

By

Albert Massey

Three farm ponds were fertilized with phosphorus and nitrogen to determine the response of the primary producers. One was fertilized with phosphorus and nitrates to approximate enrichment with primary sewage effluent; another to approximate secondary sewage effluent; a third to approximate tertiary effluent; and a fourth was unfertilized to serve as a control.

Water samples were monitored for temperature, dissolved oxygen, pH, alkalinity, soluble phosphorus, total phosphorus and nitrate. Primary producers were also monitored. Changes in water chemistry were exhibited by all the treated ponds. In general, the severity of the changes produced by the treatments was relative to the nutrient load received. The highest nutrient loading rate caused the greatest fluctuations in mean dissolved oxygen concentrations, pH and alkalinity.

The phosphorus and nitrogen concentrations in the fertilized ponds never reached the theoretical concentration and decreased rapidly, shortly after fertilization. The rapid decline of phosphate

was probably due to a comination of factors, including bacterial activity, absorption, intake by plants and precipitation with Ca^{++}. The decline in nitrate was biogenic in origin.

Fertilizer-induced changes in productivity or standing crop of the macrophytes, phytoplankton and periphyton were observed under the conditions of the study. Results indicate that the highest nutrient loads favored the production of phytoplankton, and the lower nutrient loads favored the production of periphyton. Macrophyte production was highest in the control pond.

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Albert Massey

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INTRODUCTION

Eutrophication (enrichment) of natural waters is one of the most significant causes of water quality problems in North America (Lee, 1970). The term is usually defined as meaning an enrichment of water by plant nutrients through either man-created or natural means (Mackenthun, 1968). Man, with his current waste disposal and farming practices, has accelerated this natural process with the result being increased primary production (Lee, 1973).

Primary production is the process by which plants change inorganic matter and solar energy into energy containing material which acts as the base for the aquatic food chain. Primary producers can be separated into three general categories: (1) phytoplankton, (2) periphyton, (3) higher aquatic plants (Wetzel, 1964).

Primary producers play a vital role in the aquatic environment (Mulligan, 1969). Some of the functions primary producers perform are to oxygenate the water, serve as a base for the food web, convert inorganic material to organic material, and store plant nutrients. However, excessive growth of higher aquatic plants and algae can have a detrimental effect on the aquatic community. Excessive primary production can result in the destruction of fisheries (Scultorpe, 1967), block irrigation ditches (Westlake, 1960), interfere with navigation (Boughey, 1963), affect public health (Holm *et al.*, 1969), and affect the recreational uses of a body of water (Little, 1966).

Scientists agree that the way to slow cultural eutrophication is to find the key factor limiting primary production and limit its input to our waterways.

The question of "What limits the primary producers?" is complicated by the fact that the aquatic ecosystem is a multifactor system which is in dynamic equilibrium. The equilibrium is established by the particular geochemical character and biota of the lake and as a result the isolation of a single limiting factor from various lake types has proved a difficult and confusing task.

The objectives of this research project were:

1. To conduct an *in situ* bioassay to determine if phosphorus and nitrogen are the factors limiting primary production in typical Michigan farm ponds.

2. To quantify the biological response of primary producers to various concentrations of growth limiting nutrients.

LITERATURE REVIEW

Part I of this research project is based on Liebig's law of the minimum which states that "growth is limited by the substance that is present in minimal quantity in respect to the needs of the organism" (Liebig, 1840).

Investigators do not always agree on the element limiting primary production. These are reasonable findings because any one of the fifteen major elements needed for growth which is in short supply may limit growth. Due to particular geochemical situations some of the following have been found to be limiting: nitrogen, phosphorus, iron, sodium, silicon, copper, carbon and vitamin B_{12} (Vollenweider, 1968). If any one of these elements which is in short supply is added to an aquatic system, primary production will increase until some other element becomes limiting.

One of the confusing factors of the nutrient controversy is that algae and aquatic plant species vary in their nutritive requirements. Minor changes in nutrient levels may bring about succession of one species by another because of slight physiological advantages in competition for nutrients (Hutchinson, 1944). As trophic levels change, i.e., from oligotrophic to eutrophic, plankton populations become more and more dominated by less desirable species of algae such as the bluegreens (Rawson, 1956).

Wetzel (1965) points out that in the past the problem has been attacked by determining as many physical and chemical parameters as possible and then seeking correlation between these fluctuating parameters and oscillations in the growth of algae. He further states that the relationship is not simply related to a single dominant influencing factor. The instantaneous concentrations may also indicate actual influencing quantities of a limiting factor or be the resultant product of a regeneration of a declining population peak. Problems of rapid turnover, re-utilization, and even storage of some materials negate the value of results of the analysis to a large extent. Some critical factors such as dissolved organic compounds are ignored because of the present inability or difficulty in determining their concentrations. In natural situations, limiting factors are relative and highly dynamic.

This is in agreement with Moss (1967) who points out chemical analysis of lake waters will not necessarily indicate potentially limiting nutrients since the environment is a dynamic system with constant interaction between biotic and abiotic factors.

Phosphorus usually occurs in natural waters in the form of phosphates. Biologically, phosphorus is one of the most important elements. Phosphorus is necessary to create high energy phosphate bonds. Adenosine diphosphate (ADP) converted to adenosine triphosphate stores energy which is later released and is the fuel for all biochemical work within cells. The photosynthetic fixation of carbon would be fruitless if it were not followed by the phosphorylation of the sugar product. Thus, one atom of phosphorus per molecule of adenosine is absolutely necessary. No organism could live without phosphorus (Deevey, 1970).

The theory that phosphorus is the limiting element in the aquatic ecosystem has been in existence since the infancy of limnology. Atkins (1923) postulated that phosphorus might be the limiting factor in phytoplankton production in the sea. In 1931 Prescott found a correlation between the phosphorus content and production of plankton in Iowa lakes. He went on to theorize that the type of flora in a lake might be influenced by the difference in phosphate content. Yoshimura (1932) concluded that total phosphate was the limiting factor during certain periods of the year. Chu (1942, 1943), while trying to develop a culture medium for optimum algae growth, found that algae were limited when the concentration of nitrogen and phosphorus dropped below 0.1 mg/l and 0.009 mg/l, respectively.

Investigations of Wisconsin lakes by Sawyer et al. in 1945 popularized the idea that phosphorus was the limiting element in the growth of algae. Of the 17 lakes studied, a concentration of 0.015 mg/l inorganic phosphorus at the time of the spring turnover seemed to be the value below which few blooms of algae would develop. Although it was never Sawyer's intention to set a minimum concentration for which phosphorus would be limiting, many authorities have used this concentration for a standard. Numerous investigations (Ball, 1948; Rodhe, 1948; Hasler and Einsele, 1948; Hutchinson and Bowen, 1950; Thomas, 1953; Ketchum, 1954; Sawyer, 1954; Gerloff and Skoog, 1957; Putnam and Olson, 1960; Hooper and Ball, 1964; Wetzel, 1966; Edmondson, 1970) have, through independent research, found that phosphorus is usually the element limiting the growth of algae and aquatic plants. Over 40 years of research has led investigators to the understanding that phosphorus is the limiting factor in most aquatic systems.

However, there have been findings from limnologists which do not agree that phosphorus is the limiting element. Due to shifting equilibrium, phosphorus may be limiting at one time during the year and nitrogen at another time (Yoshimura, 1932). In other situations where large amounts of phosphorus are available, nitrogen may become limiting (Shapiro and Ribeiro, 1965).

Investigators have found that trophic increases in Lake Michigan have led to a change in the silicon cycle. The diatom population in the last 20 years has utilized silicon at a greater rate than the amount supplied from all sources relative to other essential nutrients. Continued depletion of silicon will tend to favor proliferation of phytoplankton other than diatoms and could result in blooms of blue-green or other undesirable algae (Schelske and Callender, 1970).

Blue-green algae, which cause most noxious blooms, have the ability to fix atmospheric nitrogen. *Nostoc paludosum* was found to fix atmospheric nitrogen as long as there was a sufficient supply of phosphorus. Low nitrate values are commonly associated with blooms of blue-green algae, presumably due to the ability of these algae to fix nitrogen and flourish when nitrogen is in short supply for other algae (Schelske *et al.*, 1970).

The nitrate nitrogen concentration in the Upper Great Lakes can be used to show that nitrogen is not the primary factor in the eutrophication of the Great Lakes. Lake Superior was found to have the highest nitrate concentration of the Great Lakes (Putnam and Olson, 1960); however, the nitrate was not used because the low phytoplankton population was limited by some other factor, probably phosphorus. In the other Great Lakes the nitrate concentrations were lower due to utilization by phytoplankton. Lake Michigan studies show that when phosphorus was added the algae were able to use the nitrogen (Schelske and Stoermer, 1971).

In Lake Erie blue-green algae populations most likely would not be reduced by the control of inorganic nitrogen, since blooms presently occur after this nutrient has all but disappeared from the lake in summer. It appears, however, that maximum populations, but not period of dominance, can be limited by phosphorus control (Hartley and Potos, 1971).

Three recent papers concerning the importance of carbon in biological systems (Kuentzel, 1969, 1970, 1971) have been widely publicized. The papers are basically literature reviews. They propose that carbon, not phosphorus, is the key limiting factor. Kerr et al. (1970) conducted a bioassay study on laboratory medium and infertile pond waters. Under the conditions of the study, where other nutrients were in adequate supply, carbon was limiting.

King (1970) comes to the conclusion that carbon can be limiting in situations where phosphorus and nitrogen are in adequate supply. In oligotrophic, mesotrophic and many eutrophic waters the carbon supply (from inorganic sources in the water, daily increment, the atmosphere, bacterial degradation of autochthonous and allochthonous material) would be more than adequate to force some other factor to be limiting (Goldman et al., 1971). In any event, the overwhelming bulk of evidence indicates that in aquatic systems normally encountered in nature, phosphorus limits primary production.

Part II of this project deals with the response of the primary producers to nutrient enrichment. Most of the research which has been done on the effect of enrichment has been done in ponds in relation to fish production. The fertilization of ponds to increase fish production dates back to the early days of China (Swingle et al., 1963). Nutrient enrichment increases primary production. The end result is an increase in food available to all higher trophic levels.

However, fertilization appears to affect the components of the primary producers in various ways. In the southeastern United States the fertilization of ponds with phosphorus, nitrogen and potassium is used to control macrophytes (Smith and Swingle, 1942). Fertilization proved to be effective in producing a rapid growth of phytoplankton which, by shading out sunlight, prevented the growth of submerged macrophytes. Fertilization of ponds in the southeast was for the purpose of increasing fish production and to control aquatic plants.

Not all enrichment studies have resulted solely in an increase in phytoplankton. Neess (1949) found that phosphate fertilizer often would stimulate the growth of macrophytes. Another difficulty frequently experienced has been the appearance of a heavy growth of filamentous algae which is detrimental to the designated use of the ponds.

Fertilizer applied in the early spring or late winter stimulated the growth of cold water filamentous algae (Swingle et al., 1963). Fertilization studies by Surber (1945) in ponds and Hasler and Jones (1949) in artificial ponds resulted in the growth of filamentous algae. Patriarche and Ball (1949) working on Michigan ponds were unable to produce a phytoplankton bloom with fertilizer until late in the summer. Their ponds contained large beds of rooted aquatic plants prior to fertilization. Instead of a plankton bloom a heavy growth of filamentous algae was produced which resulted in elimination of most of the submerged macrophytes.

Later, Ball and Tanner (1951) while working on a small warm water lake in northern Michigan found that three applications of fertilizer during the summer resulted in an increase in phytoplankton, yet a fourth application caused a filamentous algae bloom. During three years of work on a small marl lake in Michigan, Hooper and Ball (1964) were unable to stimulate a bloom of photoplankton. However, a rise in the production of the benthic community was noted.

More recently Mulligan and Baranowski (1969) conducting experiments at Cornell University found that high nutrient levels (5.0 mg/l phosphorus and 15-50 mg/l total nitrogen) cause an increased turbidity of the water, increased chlorophyll a and increased dry weight of phytoplankton. Lower levels of nutrient enrichment (0.17-1.5 mg/l nitrogen and 0.065-0.51 mg/l phosphorus) caused an increase in the production of macroscopic filamentous algae. The lowest fertilization levels tested stimulated the growth of higher aquatic plants. High yields of macrophytes never coincided with large phytoplankton populations.

Wetzel and Hough (1973) suggest that as nutrient limitations are decreased there is an increase in macrophyte production. At some point, as nutrient loading increases, there is a shift in production with attached and eulittoral algae playing a more important role. During this time span, when the attached and eulittoral algae were increasing, phytoplankton production also increases.

There appears to be a number of factors which control the component of the primary producers which will respond to nutrient enrichment. Some of these factors appear to be: season in which enrichment was started, water temperature, sunlight, standing crop of macrophytes prior to enrichment, rate of nutrient loading, concentration of fertilizer in the water and availability of carbon.

DESCRIPTION OF STUDY AREA

This study was conducted at the Michigan State University Experiment Station two miles south of Lake City, Missaukee County, Michigan. Michigan State University owns and operates an 850-acre farm as an agricultural experimental research station. Four man-made ponds, a laboratory and living facilities are maintained by the Fisheries and Wildlife Department for field research.

Construction of the ponds began in 1942. The original ponds were below a beaver dam on Mosquito Creek. However, the beaver dam proved unreliable and an earthen dam with concrete structures which allow control of water levels in the ponds was completed in 1945. The control structures allow the water level in each pond to be maintained independently of the other ponds (Figure 1).

Mosquito Creek is an outlet for Missaukee Lake. High water levels in the lake during recent years had resulted in increasing amounts of water passing through Mosquito Creek. The high water levels insured an adequate supply of water to the ponds during the study period.

The ponds were assigned the letters A, B, C and D (Figure 1). The physical dimensions of the ponds are listed in Table 1. The ponds were originally constructed with gravel bottoms. However, over the years sediments composed of silt, muck and decaying aquatic vegetation covered sections of the ponds. During the study period

Figure 1. Location and diagram of the Michigan State University Experimental Ponds at Lake City, Michigan, 1970.

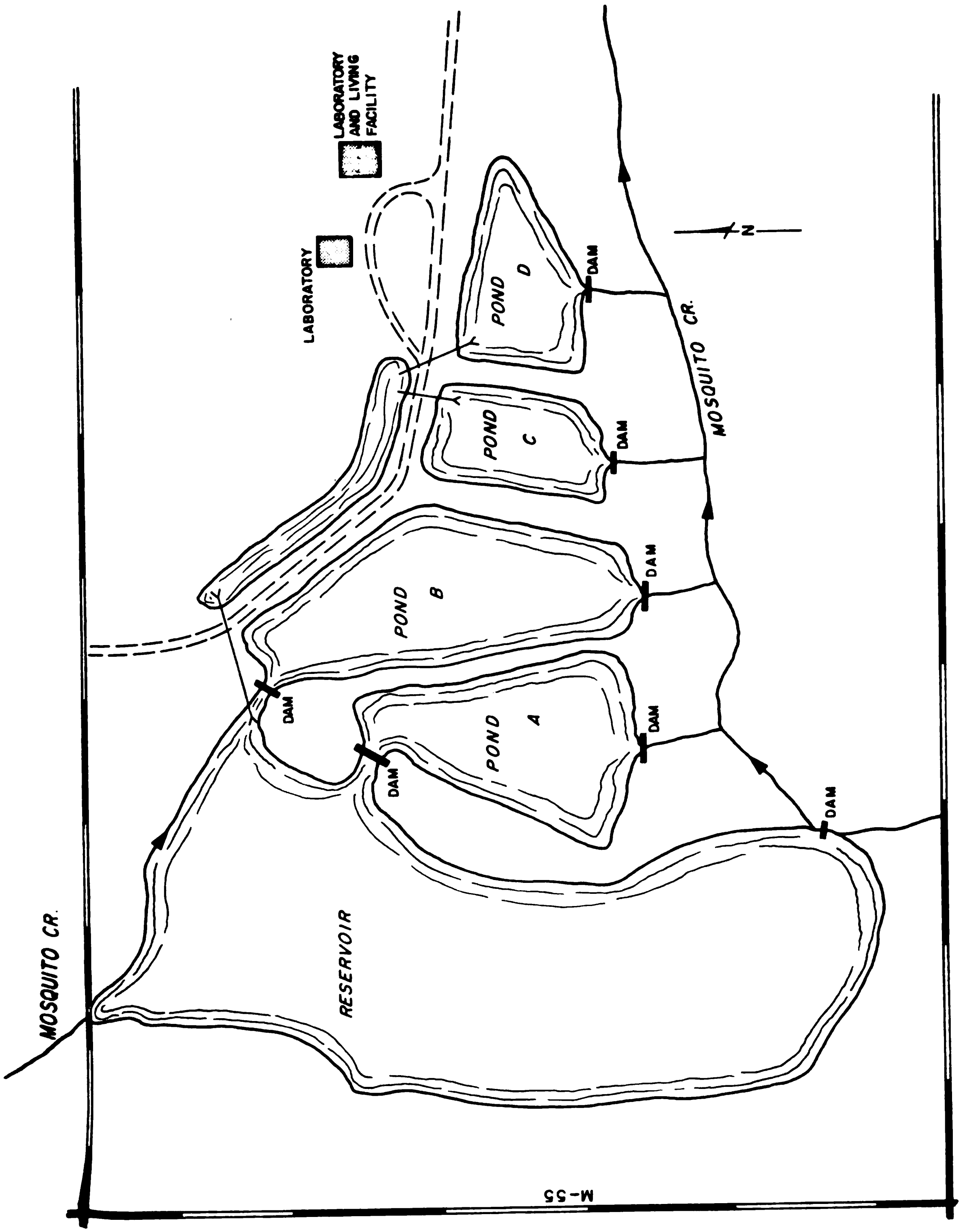

LABORATORY AND LIVING FACILITY
LABORATORY
POND D
POND C
POND B
POND A
DAM
MOSQUITO CR.
RESERVOIR
N
M-55

Table 1. Physical Dimensions of Lake City Experimental Ponds, Lake City, Michigan, 1970.

Pond	Acres	Average Depth (feet)	Acre Feet
A	0.36	3.2	1.152
B	0.46	3.3	1.518
C	0.17	3.4	0.578
D	0.19	3.2	0.608

every attempt was made to avoid disturbing the sediments. Aquatic plants present at the onset of the study are listed in Table 2.

Table 2. Aquatic Plants in Lake City Ponds, June, 1970.

Pond A.

Chara

Elodea canadensis

Najas flexilis

Potamogeton natans

Pond B.

Chara

Elodea canadensis

Najas flexilis

Potamogeton natans

Pond C.

Chara

Potamogeton natans

Potamogeton paraelongus

Elodea canadensis

Nymphaea ordorata

Pond D.

Potamogeton amphifolius

Potamogeton natans

Najas flexilis

Elodea canadensis

METHODOLOGY

Fertilization Rates

The fertilization schedule for Pond A was modeled to simulate the rate of enrichment which would occur if three million gallons a day of primary wastewater treatment plant effluent, which contained 5 mg/l total phosphorus and 30 mg/l total nitrogen, were discharged to a 12.7 acre pond six feet deep.

The enrichment in Pond B simulated the nutrient load a 13.3 acre pond six feet deep would receive if three million gallons a day of wastewater treatment plant effluent containing 2.5 mg/l of total phosphorus and 15 mg/l total nitrogen were discharged into it.

Pond C received a nutrient load proportional to that which would occur if three million gallons a day of wastewater treatment plant effluent, which contained 1.25 mg/l of total phosphorus and 7.0 mg/l total nitrogen, were discharged into a 5.6 acre pond six feet deep.

Pond D was not fertilized and contained natural levels of total phosphorus and total nitrogen.

Phosphorus Application

Phosphorus as P_2O_5 is a well known fertilizer for terrestrial plants. Triple superphosphate which is 46 percent available P_2O_5 was used as a phosphorus source. The fertilizer was purchased locally in fifty-pound bags. Phosphorus was added at a rate which would bring the initial concentration of phosphorus in the pond water to the following levels. Subsequent applications would increase the phosphate concentration.

Pond	Amount of Phosphorus Added (pounds)	(kg)	(lb/acre foot)	Theoretical Concentration
A	1.85	0.84	1.60	0.59 mg/l
B	1.2	0.54	0.79	0.29 mg/l
C	0.52	0.24	0.90	0.33 mg/l
D	0.0	0.0	0.0	0.0

At 0900 on July 7, 1970, the first phosphorus application was made to Pond A. Prior to application the phosphorus was placed in 10 gallons of water in a plastic container overnight. One gallon of the phosphorus solution was then placed with four gallons of water in a manually operated water pump. The pump and operator were then rowed around the pond in a small boat. While the boat was moved around the pond the operator directed the nozzle of the pump at the water's surface. This process was repeated until all 10 gallons had been used. Every attempt possible was made to spread the phosphorus evenly. In addition, the boat was rowed around the pond for an additional 10 minutes to aid in mixing. Due to water quality problems discussed in the nitrogen section, phosphorus was not added on July 16, 17 and at only one-half the original rate on July 18 and 20.

The initial application was made to Ponds B and C on July 14 and July 17, respectively. Dates of fertilization and quantities added are shown in Table 3.

Table 3. Dates of Nutrient Enrichment and Quantities [a] Added in Experimental Ponds

Date	Pond A	Pond B	Pond C	Pond D
July 7, 1970	1.60 phosphorus	0.0	0.0	0.0
	10.57 nitrogen	0.0	0.0	0.0
July 8, 1970	1.60 phosphorus	0.0	0.0	0.0
	10.57 nitrogen	0.0	0.0	0.0
July 9, 1970	1.60 phosphorus	0.0	0.0	0.0
	10.57 nitrogen	0.0	0.0	0.0
July 10, 1970	1.60 phosphorus	0.0	0.0	0.0
	10.57 nitrogen	0.0	0.0	0.0
July 11, 1970	1.60 phosphorus	0.0	0.0	0.0
	10.57 nitrogen	0.0	0.0	0.0
July 14, 1970	1.60 phosphorus	0.79 phosphorus	0.0	0.0
	10.57 nitrogen	4.65 nitrogen	0.0	0.0
July 15, 1970	1.60 phosphorus	0.79 phosphorus	0.0	0.0
	10.57 nitrogen	4.65 nitrogen	0.0	0.0
July 16, 1970	1.60 phosphorus	0.79 phosphorus	0.0	0.0
	0.0 nitrogen	0.0 nitrogen	0.0	0.0
July 17, 1970	0.0 phosphorus	0.0 phosphorus	0.90 phosphorus	0.0
	0.0 nitrogen	0.0 nitrogen	2.99 nitrogen	0.0
July 18, 1970	0.80 phosphorus	0.79 phosphorus	0.90 phosphorus	0.0
	5.28 nitrogen	4.65 nitrogen	2.99 nitrogen	0.0
July 20, 1970	0.80 phosphorus	0.79 phosphorus	0.90 phospnorus	0.0
	5.28 nitrogen	4.65 nitrogen	2.99 nitrogen	0.0
July 23, 1970	1.60 phosphorus	0.79 phosphorus	0.90 phosphorus	0.0
	10.57 nitrogen	4.65 nitrogen	2.99 nitrogen	0.0
July 27, 1970	1.60 phosphorus	0.79 phosphorus	0.90 phosphorus	0.0
	10.57 nitrogen	4.65 nitrogen	2.99 nitrogen	0.0
July 29, 1970	1.60 phosphorus	0.79 phosphorus	0.90 phosphorus	0.0
	10.57 nitrogen	4.65 nitrogen	2.99 nitrogen	0.0
August 1, 1970	1.60 phosphorus	0.79 phosphorus	0.90 phosphorus	0.0
	10.57 nitrogen	4.65 nitrogen	2.99 nitrogen	0.0
August 3, 1970	1.60 phosphorus	0.79 phosphorus	0.90 phosphorus	0.0
	10.57 nitrogen	4.65 nitrogen	2.99 nitrogen	0.0
August 19, 1970	1.60 phosphorus	0.79 phosphorus	0.90 phosphorus	0.0
	10.57 nitrogen	4.65 nitrogen	2.99 nitrogen	0.0

[a] pounds per acre-foot

Nitrogen Application

Nitrogen in the form of urea and potassium nitrate are well known fertilizers for terrestrial plants. Initially in the study urea was used as a fertilizer. The urea was 45 percent NH_3, by weight.

A build-up of ammonia nitrogen levels was determined to be stressing fish in Pond A on July 16, 1970. On July 18, 1970, the nitrogen source was changed from urea to potassium nitrate. Potassium nitrate was purchased locally in 50-pound bags which contained 33 percent nitrate nitrogen by weight. Nitrogen was added at a rate which would bring the water to the following concentrations. Subsequent applications would increase the nitrate concentration.

Pond	Amount of Nitrate Added (pounds)	(kg)	(lb/acre foot)	Theoretical Concentration
A	11.05	5.02	10.57	3.52 mg/l
B	7.05	3.20	4.65	1.71 mg/l
C	3.2	1.45	2.99	1.1 mg/l
D	0.0	0.0	0.0	0.0 mg/l

At 0900 on July 7, 1970, the first application of urea was made to Pond A. Prior to application the urea was placed in 10 gallons of water to which the phosphorus had already been added and left to stand overnight. One gallon of the phosphorus-nitrogen solution was then placed with four gallons of water in a manually operated water pump. The pump and operator were then rowed around the pond in a small boat. While the boat was rowed around the pond the operator directed the spray from the nozzle of the pump at the water's surface. This process was repeated until all 10 gallons of solution had been applied. Every attempt was made to spread the solution evenly. In addition, the boat was rowed around the pond for an additional 10 minutes to aid mixing.

Due to the ammonia problem discussed earlier in this section, nitrogen was not added to Ponds A and B on July 16 and 17. On July 18 and 20, nitrogen loading was reduced by one-half in Pond A.

Initial application was made on Ponds B and C on July 14 and 17, respectively. Dates of fertilization and concentrations are shown in Table 3.

Water Exchange

Rate of water flow through the ponds was monitored by constructing straight notch weirs below the outlet structures of each pond. Boards on the inlet structures were adjusted to allow a sufficient water flow through the ponds to conform with a predetermined retention time. The retention time of the pond was calculated using the following formula:

$$t = \frac{v}{q}$$

where t is the theoretical retention time in days, v is the volume of the pond in millions of gallons and q is the rate of flow in millions of gallons. The theoretical retention time is the time in which there is complete replacement of all water in the pond. However, this is a volumetric replacement and not molecule for molecule replacement. Therefore, some water was not replaced. The theoretical retention times for the ponds were:

Pond A, 8.3 days

Pond B, 8.7 days

Pond C, 3.7 days

Pond D, 4.0 days

Physical and Chemical Parameters

Temperature

Water temperature in the ponds was monitored with a Taylor continuous recording thermometer. The sensor for this unit was placed in four feet of water near the outlet of Pond B. Weekly inspection of the recorder chart indicated the maximum and minimum temperatures which had occurred on any date. On several occasions water temperatures in all ponds were checked with a hand-held thermometer to determine if temperatures in Pond B were similar to temperatures in Ponds A, C and D. Ambient air temperature was recorded at and obtained from the weather station at the Lake City Experiment Station.

Dissolved Oxygen

Dissolved oxygen concentrations were determined between 10:00 and 11:00 A.M. using the azide modification of the Winkler method (Anon., 1965). Due to its chemical stability, phenylarsene oxide was substituted for sodium thiosulfate in the oxygen determinations. Water samples for determination of dissolved oxygen and the remainder of the chemical parameters were taken at three-day intervals from June 29 to August 8, 1970. From August 8 until August 22 a four-day sampling interval was used. Water samples were also analyzed on the initial day of fertilization in Ponds A, B and C and on August 24 and September 4, 1970.

pH

A Beckman pH meter was used to measure hydrogen ion concentration on the sample dates indicated in the dissolved oxygen section. The instrument was standardized at regular intervals.

Alkalinity

Phenolphthalein alkalinity is a measure of the concentration of carbonate $(CO_3)^=$ and hydroxide $(OH)^-$ ions in the sample. The amount of carbonate and hydroxide are determined by titration with an acid to the phenolphthalein end-point at pH $\approx$ 8.3, while the total alkalinity was titrated to the color end-point of pH between 4.5 and 5.4 using methyl orange as a color end-point indicator (yellow to pink).

Total and phenolphthalein alkalinity were measured following the procedures outlined in Standard Methods (Anon., 1965).

Phosphorus

The concentrations of orthophosphate and total phosphate were determined in all ponds at predetermined intervals. Orthophosphate was determined spectrophotometrically using the stannous chloride method (Anon., 1965). Under acid conditions orthophosphate forms a yellow complex with molybdate ions. The phosphate-molybdate complex was then reduced to a highly colored blue complex by the addition of stannous chloride. The color is proportional to the phosphate concentration. The blue color was then measured at 690 μ on a Beckman Model B spectrophotometer.

Total phosphate was determined spectrophotometrically using the stannous chloride method after an initial acid hydrolysis step (Anon., 1965). Boiling with acid for 90 minutes hydrolyzed polyphosphates, insoluble phosphates and some organic phosphates to orthophosphates. Orthophosphate was then determined as outlined above.

Samples for phosphorus determinations were taken in 250-ml glass-stoppered, acid-washed bottles. Samples were returned to the laboratory and processed within two hours. Water samples for phosphorus analysis were taken on the schedule outlined under dissolved oxygen. This sampling schedule was designed to monitor changes in phosphorus concentrations in pond waters.

Nitrate-Nitrogen

Nitrate was determined spectrophotometrically using the brucine method (Anon., 1965). Brucine is an organic compound which reacts with nitrates under acid conditions and at elevated temperatures develops a yellow color. The color of the resulting complex was measured at 410 μ on a Beckman Model B spectrophotometer. There are two factors which cause difficulty when this method is used: (1) The color development does not obey Beer's law. A curve rather than a straight line is obtained from graphing a set of standard solutions.

(2) The intensity of color development is a function of time and temperature. Temperature and time during analysis were carefully controlled. For each series of analysis a set of nitrate standard solutions was also analyzed. To insure accuracy, after analysis a new calibration curve was developed from the standard solutions.

Water samples for nitrate analysis were taken at the intervals indicated under the dissolved oxygen section. Samples for analysis were taken in 250-ml acid-washed bottles and returned to the laboratory. Analysis was undertaken within two hours.

Ammonia nitrogen was determined on July 17, 1970 in Pond A using the Nessler method (Anon., 1965).

Biological Parameters

Macrophytes

The standing crop of macrophytes was measured gravimetrically after the whole plant including the roots was cropped (Penfound, 1956; Sculthorpe, 1967). With the aid of stakes on shore to locate coordinates, all ponds were divided into one-meter square plots. Plots which contained emergent or floating aquatic plants or were within one meter of shore were not sampled. Each pond was sampled at seven-day intervals, with a plot to be sampled being selected using a table of random numbers. Ropes were stretched between coordinating stakes, thus locating the plot. Three 625 square centimeter subsamples were taken from each square meter plot.

Plants were harvested by placing a square (25 cm x 25 cm) frame within the one meter plot and removing all plants. An attempt was made to remove all plant roots. The plants were removed by hand with the worker hanging over the side of the boat wearing scuba gear or with the diver free in the water. Plant samples were washed in fresh water, blotted dry, then placed in a constant temperature oven (50°C) for 48 hours (Wetzel, 1960). Plants were then placed in a desiccator, cooled to room temperature and weighed on an analytical balance.

Sculthorpe (1967) lists four aquatic plant species found in the study ponds as containing from 14 to 23.9 percent of their dry weight as ash free dry weight. Tesar (1973) reported the ash free weight of macrophytes ranged from 29 to 43 percent of the net weight. A mean of 19.5 percent was calculated from the work of Sculthorpe and used to estimate ash free dry weight in the study. This calculated mean is similar to the 20 percent ash free weight from dry weight reported by Slādeček and Sládečková (1964).

Phytoplankton

In this study phytoplankton was defined as the free floating algae which could be removed from the water by filtration with a 0.45 μ Millipore R Filter. This group would include the tychoplankton which is normally found free floating with other vegetation, usually macrophytes, in the littoral zone (Hutchinson, 1957). No attempt was made to trace the change in species produced in the ponds since greater importance was attached to the total quantity of plankton produced.

The standing crop of phytoplankton was measured in each pond at four-day intervals. Chlorophyll a was used to determine standing crop and was measured spectrophotometrically using the methods of

Parsons and Strickland (1963) as modified by Moss (1967). Samples were obtained from all ponds between 10:00 A.M. and 12:00 noon on sampling days. This was done to reduce the possibility of diurnal changes in chlorophyll concentration (Yentsch and Ryther, 1957). To insure that the phytoplankton sample was representative of the plankton population, eight one-liter samples from various locations in the ponds were placed in a plastic bucket and mixed. A one-liter subsample was then removed and used for analysis. A water sample between 25 ml and 500 ml was filtered through an H.A. 0.45 ± 0.02 μ Millipore *R* Filter (Millipore Filter Corp., Bedford, Mass.) upon which three drops of $MgCO_3$ had been placed. Care was taken to keep the filter vacuum below 15 inch Hg. to avoid damage to the algal cells and loss of cell contents (Wetzel, 1964). The amount of sample filtered was dependent upon the plankton population present with high plankton densities reducing the volume of sample filtered.

After filtration the filter was placed in a mortar and ground vigorously with a pestle for two minutes in 2 ml of 90 percent redistilled acetone. The mixture was rinsed with 4 ml of acetone into a 15-ml glass-stoppered centrifuge tube. The centrifuge tube was then placed in a dark refrigerator for 24 hours prior to analysis for chlorophyll a and its degradation products. Chlorophyll a concentrations were determined with the aid of a Beckman Model B spectrophotometer.

Ash free dry weight was estimated using the values reported by Grzenda and Brehmer (1960). They reported that phyto-pigments constituted 1.3 percent of the ash free dry weight. These findings are in agreement with Weber (1973) who estimated the chlorophyll content of a cell to be 1 to 2 percent of the ash free dry weight.

Periphyton

Studies have shown the amount of biomass of periphyton is directly related to the amount of chlorophyll *a* present in a sample (Weber, 1973; Grzenda and Ball, 1968). Standing crop and colonization rates of this community can be measured and sample sites compared if the quantity of chlorophyll per unit area is known. Artificial substrates (plexiglass) were provided for periphyton growth, then the growth measured at predetermined time intervals.

Clear plexiglass plates with a total exposed surface area of 140 square centimeters (2" x 5" x 1/4") were used to collect periphyton growth. The plates were attached to a horizontal crossbar with Acco #1125 spring clips. The crossbar was attached to a wooden pole driven into the pond bottom. In each pond a total of 16 plates were held 12 inches below and with the flat surface parallel to the water's surface (King and Ball, 1966).

At four-day intervals duplicate samples were removed from each pond. By measuring the amount of biomass accumulation between sampling dates an estimate of the colonization rate was calculated. In this manner plates which had been in the ponds for time intervals of from 4 to 32 days were compared. At the end of the first 32-day period (July 28) all plates were replaced and the 32-day cycle restarted.

The plexiglass plates were removed from the clamps, placed in individual light-proof containers and taken directly to the laboratory for immediate processing. The plexiglass plates were scraped into a mortar to remove the periphyton community which had colonized it. The plexiglass slide was then rinsed with 2 ml of 90 percent redistilled acetone into the same mortar. An additional 5 ml of acetone was placed into the mortar. The periphyton-acetone mixture was vigorously ground with a pestle for one minute. The contents of the mortar were then rinsed with 3 mls of acetone into a 15-ml glass-stoppered centrifuge tube. The centrifuge tube was refrigerated at 6°C for 24 hours. After 24 hours the sample was examined spectrophotometrically for chlorophyll a as outlined by Parsons and Strickland (1963) and its degradation products (Moss, 1967).

Ash free dry weight was estimated using the values reported by Grzenda and Brehmer (1960). They reported that phytopigments constituted 1.3 percent of the ash free dry weight. These findings are in agreement with Weber (1973) who estimated the chlorophyll content of a cell to be 1 to 2 percent of the ash free dry weight.

RESULTS AND DISCUSSION

General Ecological Considerations

It is a well established ecological observation that individuals of a population in order to grow and multiply must be supplied with certain essential materials. Not only must specific essential elements be present, but each element to which the organism responds has a maximum and minimum limiting effect. These observations are usually stated as Liebig-Blackman law and Shelford law (Boughey, 1968). The former states that growth of an organism is dependent on the amount of foodstuff (e.g., inorganic, or organic) or level of factor (e.g., light, temperature) present in minimum quantity to the organism relative to the availability of essential elements and factors in the organism's environment and the proportionate cellular requirements of the organism. Shelford's law goes on to recognize the effect of an excess of one element or factor has on the growth of an organism. That is, Shelford recognized the growth or metabolism of an organism is injured when an element is supplied above or below a certain concentration (Bowen, 1966). Thus, an organism's success in populating a given ecosystem is controlled by the quantity and variability of materials for which there is a minimum requirement and physical factors which are critical and by the limits of tolerance of an organism to these factors and competing organisms (Odum, 1971).

The presence or success of a population of organisms in a given ecosystem is based on the completeness of a complex of conditions. What is essential is the identification of the operationally significant factors on the organism's growth from the array of possible controlling factors. Environmental factors that have been experimentally demonstrated to exert control on the growth and metabolism of plant and animal populations in aquatic ecosystems include light, temperature, organic compounds, dissolved oxygen, salinity, dissolved inorganic constituents, and suspended solids.

In discussing the abundance of the primary producers and causal factors responsible for the observed population shifts, it will be done in light of these ecological principles (Liebig and Shelford laws). They provide the framework to understand the chance of an organism to thrive and grow in a given environment and not in another (Odum, 1971).

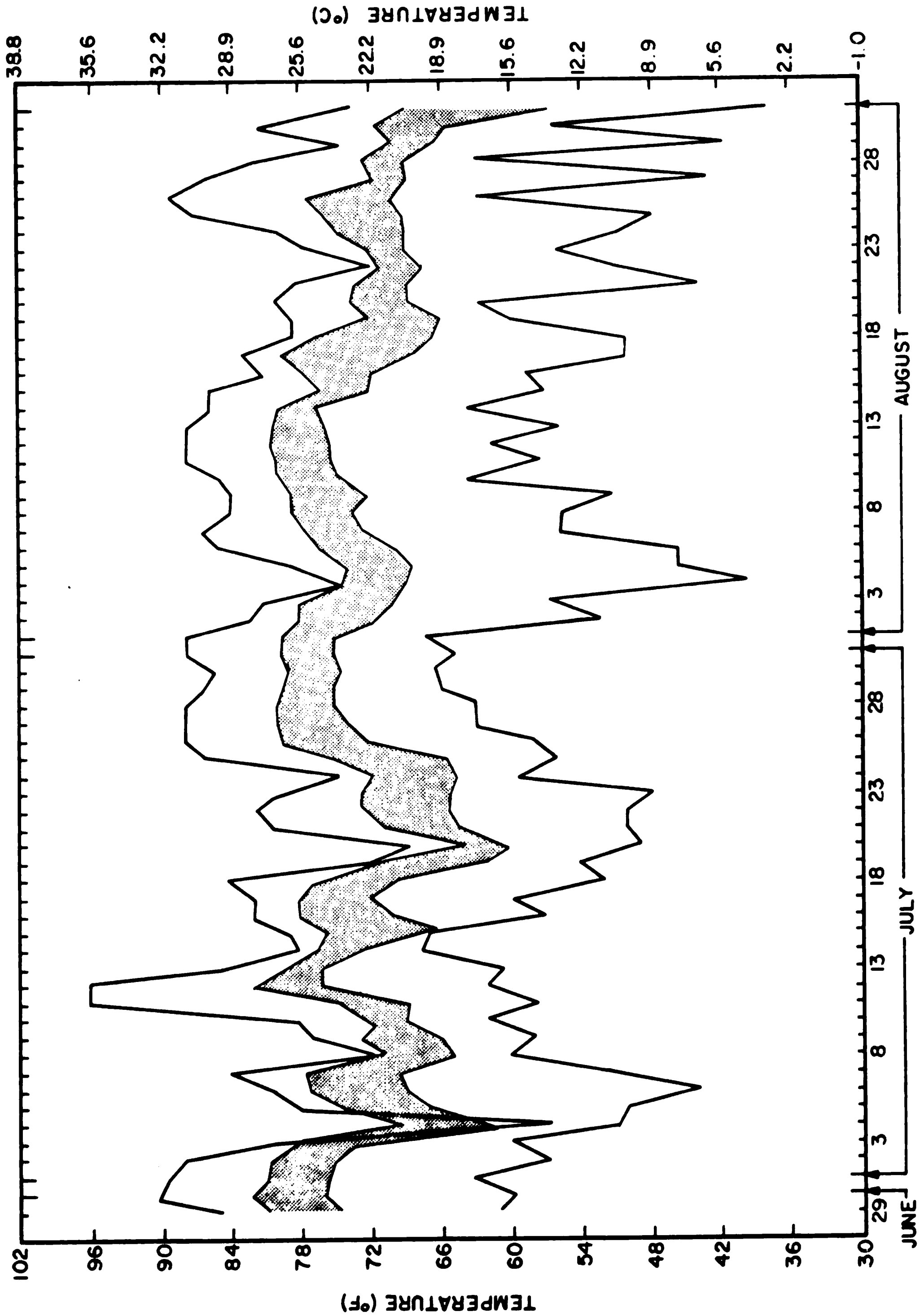
Physicochemical Characteristics of Pond A

Pond A was the first pond to be fertilized (July 7, 1970). It also received the highest rate of nitrogen and phosphorus loading, 5.02 (3.52 mg/l) and 0.84 kg (0.59 mg/l), respectively.

Temperature

The temperature in the four ponds followed similar patterns. Temperatures during the study ranged from a maximum of 28.1°C (82.5°F) on June 30 down to a minimum of 13.7°C (56.9°F) on August 31, 1970 (Figure 2). The maximum diurnal change in water temperature from a daytime high to a nighttime low was 5.0°C (9.9°F) and occurred on

Figure 2. The daily maximum and minimum air and water temperatures in Lake City ponds during the study period, 1970. (Maximum and minimum water temperatures are stippled; maximum and minimum air temperatures are clear.)


TEMPERATURE (°F)
30 36 42 48 54 60 66 72 78 84 90 96 102
TEMPERATURE (°C)
-1.0 2.2 5.6 8.9 12.2 15.6 18.9 22.2 25.6 28.9 32.2 35.6 38.8
JUNE 29
JULY 3 8 13 18 23 28
AUGUST 3 8 13 18 23 28

July 23. The maximum diurnal change in surface air temperature from a daytime high to a nighttime low was 23.9°C (43°F) and occurred on August 27, 1970. The stable temperature in the waters (1/5 the variation of the air) is due to the specific heat capacity of water (Hutchinson, 1967). The ecological importance of temperature lies in the seasonal and daily variation within the ponds. This flux is what directly affects the metabolic activities of the existing plant and animal communities.

Organisms such as algae may be greatly influenced by temperature (Hutchinson, 1967). Blue-green algae are very tolerant of high temperature. Diatoms such as <u>Fragilaria crotonensis</u>, however, grow best between 13-16°C. In most years <u>Fragilaria crotonensis</u> and other diatoms are largely replaced in the summer by blue-green algae (Wesenberg-Lund, 1904). Algae encountered in this study would be from the mesothermal temperature spectrum (15-30°C) (Lowe, 1974).

The solubility of oxygen in water varies inversely with temperature. In fresh water, the solubility of atmospheric oxygen is decreased by about 55 percent as the temperature increases from 0°C to 40°C (Mackenthun and Ingram, 1967). Therefore, high temperatures indirectly affect organisms by controlling the amount of oxygen available to them at a time when their metabolic rates are high.

<u>Oxygen</u>

Oxygen concentrations averaged 8.2 mg/l (94 percent saturation). An oxygen maximum appeared very early in the study (July 19) during the peak of the phytoplankton standing crop (Table 4). Levels

Table 4. Concentration of Selected Chemical Constitutents[a] in Pond A, Lake City, Michigan, 1970.

Date	Dissolved Oxygen	pH	Total Alkalinity	Phenolphthalein Alkalinity	Ortho Phosphorous	Total Phosphorous	Nitrate-Nitrogen
	Parameter						
6/29/70	8.1 (97)[b]	8.3	86.0	0.0	<0.01	0.01	<0.01
7/1/70	8.6 (100)	8.5	84.0	0.0	<0.01	0.02	0.02
7/4/70	7.6 (83)	7.9	85.0	0.0	<0.01	<0.01	0.02
7/7/70	8.3 (96)	7.9	86.0	0.0	0.42	0.45	0.03
7/10/70	6.8 (76)	7.8	91.0	0.0	1.15	1.28	0.07
7/13/70	16.5 (182)	8.9	105.0	16.0	1.27	1.34	0.30
7/16/70	17.2 (185)	9.0	104.0	22.0	1.98	2.12	0.75
7/19/70	19.0 (186)	9.3	107.0	38.0	1.40	1.61	2.51
7/23/70	13.0 (152)	9.8	93.0	42.0	1.49	1.69	3.01
7/25/70	11.5 (127)	9.6	87.0	26.0	1.06	1.20	2.25
7/28/70	10.3 (122)	9.5	92.0	21.0	1.15	1.26	3.08
7/31/70	12.7 (136)	9.8	87.0	29.0	1.05	1.17	3.84
8/3/70	8.0 (92)	8.9	85.0	11.0	1.41	1.59	6.46
8/6/70	8.1 (94)	9.1	82.0	7.0	1.01	1.11	4.05
8/10/70	4.8 (57)	8.0	90.0	0.0	0.66	0.75	3.07
8/14/70	5.0 (61)	8.0	91.0	0.0	0.30	0.44	1.75
8/18/70	5.2 (58)	7.9	88.0	0.0	0.19	0.26	0.82
8/22/70	4.5 (52)	7.6	93.0	0.0	0.38	0.49	2.13
8/27/70	7.6 (79)	7.8	86.0	0.0	0.16	0.23	0.7
9/4/70	7.9 (80)	8.1	84.0	0.0	0.01	0.06	0.10

[a]Expressed in mg/l except pH which is in standard units.

[b]Percent saturation.

of 19 mg/l (186 percent saturation) of oxygen were reached throughout the shallow pond. Following this peak in productivity, oxygen levels within the pond were reduced but still remained between 120 and 152 percent saturation at the existing temperatures. The very intensely developed oxygen maxima began in mid-July, persisted until late July and remained below 100 percent saturation throughout August. The primary productivity of the pond was particularly intense throughout the periods of the maxima in direct correlation with the development of high oxygen concentrations. These findings are similar to those reported by Wetzel (1966) in hypereutrophic Sylvan Lake. He reported a maximum oxygen concentration of 16.6 mg/l and 189.5 percent saturation during July.

Alkalinity and pH

Experimental Pond A at Lake City had an average total alkalinity of 90 mg/l. This is similar to the findings of Sohacki (1965) for the same ponds. It can be classified as a hardwater pond (Ruttner, 1940). Alkalinity is the buffering capacity of a body of water, i.e., its ability to withstand changes in pH. Alkalinity is imparted by the proton accepting species in the water, usually the bicarbonates, carbonates and hydroxide components of the water body. As plants remove CO_2 from the water for use in photosynthesis, a new chemical equilibrium must be established. Carbon dioxide is utilized by primary producers causing a loss of bicarbonate, and carbonate and hence a reduction in buffering capacity. A change in pH may result if the buffering capacity is exceeded.

Total alkalinity in Pond A ranged from 82 to 107 mg/l with a mean of 90.3 mg/l. Total alkalinity concentrations were slightly elevated on July 13, 16, and 19. The elevation was probably due to the potassium nitrate fertilizer used to enrich the ponds. Potassium would dissociate from the nitrate and be available to form potassium bicarbonate and carbonate (Hutchinson, 1957). Potassium bicarbonate and carbonate would increase the buffering capacity of the water. Barrett (1957) found that when fertilizer containing potassium was added to the lake it did go into solution but was only a small fraction of the amount already occurring in the lakes.

Phenolphthalein alkalinity was 0.0 except on several occasions when a supersaturation of dissolved oxygen and high pH values (> 8.9) occurred. The absence of phenolphthalein alkalinity indicates all of the alkalinity was due to bicarbonates. High primary production resulted in the loss of CO_2, high pH values and a supersaturation of dissolved oxygen. As a result of the loss of CO_2, the concentration of bicarbonate was lowered (indicated by increased phenolphthalein alkalinity) and could not effectively act as a buffer for hydroxyl ions produced by the photosynthetic uptake of CO_2. As a result, hydroxyl ions accumulate accounting for a dramatic rise in pH (Vallentyne, 1974).

Phosphorus Concentrations

The total phosphorus concentration in Pond A ranged from less than 0.01 mg/l on July 4 to 2.12 mg/l on July 16, 1970 (Table 4). The total phosphorus concentration increased immediately after fertilization to 0.45 mg/l which is somewhat lower than the theoretical concentration expected (0.59 mg/l). On July 11 after five days of receiving 1.85 pounds of phosphorus per day, an algal bloom began to develop. Fertilization was halted on July 12 and July 13. Fertilization was resumed on July 14, 15 and 16. On July 16, total phosphorus reached the highest concentration during the study period (2.12 mg/l). This value was, however, less than the expected theoretical value of 2.73 mg/l. Observed total phosphorus values ranged from 60 to 96 percent of the theoretical values (Table 5). The phosphorus maxima coincided with the maximum standing crop of phytoplankton.

Soluble phosphorus concentrations ranged from less than 0.01 mg/l on several occasions to 1.98 mg/l on July 16, 1970. Soluble phosphorus concentrations followed the same general pattern as total phosphorus concentrations, i.e., increased with fertilization, decreased when enrichment was halted.

Soluble phosphorus ranged from 93 percent of the total phosphorus, on the first day of the study, to 16 percent total phosphorus on the last day sampled (September 4, 1970).

Nitrogen

Although air contains 79 percent nitrogen, only a few organisms can use nitrogen in this form. Nitrogen is present in the aquatic ecosystem in the following forms: N_2, NH_3, NO_2^-, NO_3^-. Most primary producers are only able to utilize nitrogen that has been fixed (incorporated in a chemical compound). Therefore, most primary

Table 5. Observed and Theoretical[a] Total Phosphorus Concentrations[b] in Pond A, Lake City, Michigan, 1970.

Date	Theoretical Concentration	Observed Concentration	Percent $\left(\frac{\text{Observed}}{\text{Theoretical}}\right)$
July 7	0.59	0.45	76
July 10	1.96	1.29	65
July 13	1.76	1.34	76
July 16	2.73	2.12	78
July 19	2.08	1.61	77
July 23	2.01	1.69	84
July 25	1.54	1.20	78
July 28	1.54	1.26	82
July 31	1.49	1.17	79
August 3	2.04	1.59	78
August 6	1.37	1.11	81
August 10	0.80	0.75	94
August 14	0.47	0.44	94
August 18	0.27	0.26	96
August 22	0.56	0.49	88
August 27	0.27	0.23	79
September 4	0.10	0.06	60
			$\bar{x}$ 80.3 percent

[a]Takes into account flushing rate of pond.

[b]mg/l.

producers are unable to use atmospheric nitrogen. Nitrogen is one of the key elements in the building of amino acids.

Certain bacteria and some algae are capable of utilizing and incorporating it into protein. Nitrogen compounds are then released as waste products or at the death of the organism through decomposition. Bacteria convert protein to NH_3 which plants and algae can then use as a nutrient. If the NH_3 is not used, the Nitrosomonas group of bacteria under aerobic conditions will convert NH_3 to NO_2^-.

If the nitrite is still unused the Nitrobacter group of bacteria convert it into nitrate.

The nitrogen cycle is a gaseous cycle. Nitrogen may be lost or added, to or from the atmosphere at any point in the cycle as a gas. The nitrogen fertilizer originally used in Pond A was urea. Urea is converted to ammonia which can be toxic to aquatic organisms and becomes more toxic as the pH value increased above 8. On July 16, 1970, fish in Pond A were observed to be in distress. Fish had congregated at the inlet of the pond and were near the surface apparently under some physiological stress. Water samples were immediately taken and analyzed for ammonia-nitrogen. The results revealed that 0.5 mg/l of ammonia was present in the water. At a pH of 9.1 enough of the non-ionized ammonia would be present to be toxic to fish (Ball, 1967). Approximately 238 bass, bluegills, and bullheads were removed from the pond and released. An additional 35 fish died in the pond. Fish were not completely eradicated from the pond: several bluegills were seen later in the summer.

Subsequent applications were made using potassium nitrate. Nitrate is quite soluble in water, easily used by aquatic plants and algae and relatively chemically stable. Nitrate concentrations in the ponds were always lower than the theoretical concentrations calculated. This is probably due to the luxury consumption of aquatic plants, phytoplankton and periphyton (Gerloff, 1960). Maximum concentration of nitrate observed during the study was 6.46 mg/l which occurred on August 3, 1970 (Table 4).

The concentrations of available nitrate should not have limited primary production. However, nitrogen can limit primary production.

Yoshimura (1932) found phosphorus may be limiting at one time of the year and nitrogen at another. In other situations where large amounts of phosphorus are available, nitrogen may become limiting (Shapiro and Ribeiro, 1965). Thomas (1953) concluded that (1) phosphorus was the minimum factor in oligotrophic lakes, and (2) there are lakes in which, according to the weather, phosphorus may be limiting one year and nitrogen may be limiting the next.

There were other significant sources of nitrogen to the ponds. Precipitation in the form of rainfall and snowfall is a significant source of nitrogen in some parts of the world. In the midwest, average rainfall contains on the order of 1 mg/l of nitrate nitrogen. When this is compared to the normal 0.3 mg/l often cited as the critical concentration of nitrogen, rainfall rather than diluting the nutrients in a lake is actually contributing to them (Lee, 1970; Pecor _et al_., 1973).

Macrophytes

The standing crop of hydrophytes in Pond A remained relatively constant during the first three sampling periods (June 29, July 8 and July 15) of the summer (Figure 3a, Table 6). These same phenomena were observed by Penfound (1956); i.e., the productivity of macrophytes was highest in spring and lowest during the summer. Low productivity was due primarily to the relatively low photosynthesis compared with that of respiration during hot summer weather. Forsberg (1959) observed production rates of submerged macrophytes were higher in spring, lower in summer and negative in the fall. Aquatic macrophyte standing crop on the three above mentioned dates was 239.6 with a S.D. 7.6 g/m^2. There was no apparent stimulation of plant growth when enrichment began on July 7.

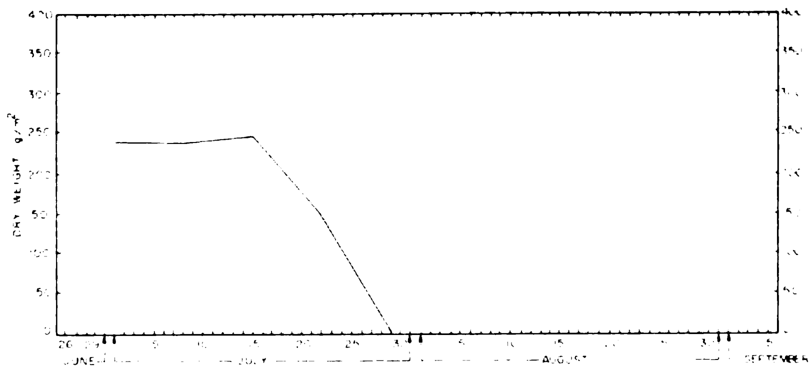
The July 15 sample from Pond A was obtained with a great deal of difficulty. The water was extremely turbid making collection of the macrophytes difficult. Plants which were sampled were found to be in poor physiological condition. Macrophytes sampled on July 22 were difficult to locate in the turbid water and those which were located were in a degenerated condition. Standing crop had been reduced to 132 g/m^2.

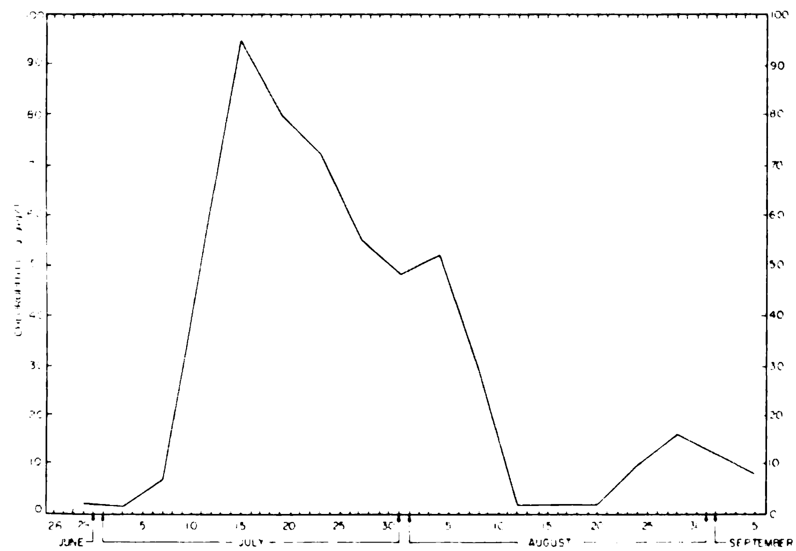
By the July 29 sampling date only a few plant fragments could be located anywhere in Pond A. The rate at which the aquatic plants degenerated was calculated to be 17 percent per day. These findings are higher but in agreement with those observed by Jewell (1970) who found degeneration of hydrophytes at 18°C to be about 9 percent per day. The decline in macrophyte standing crop may have

Figure 3a. Macrophyte standing crop in Pond A, Lake City, Michigan, 1970.

Figure 3b. Phytoplankton standing crop as determined by chlorophyll *a* concentrations in Pond A, Lake City, Michigan, 1970.

Figure 3c. Periphyton production rates and standing crop in Pond A, Lake City, Michigan, 1970.


DRY WEIGHT g/m²
JUNE
JULY
AUGUST
SEPTEMBER


JUNE
JULY
AUGUST
SEPTEMBER

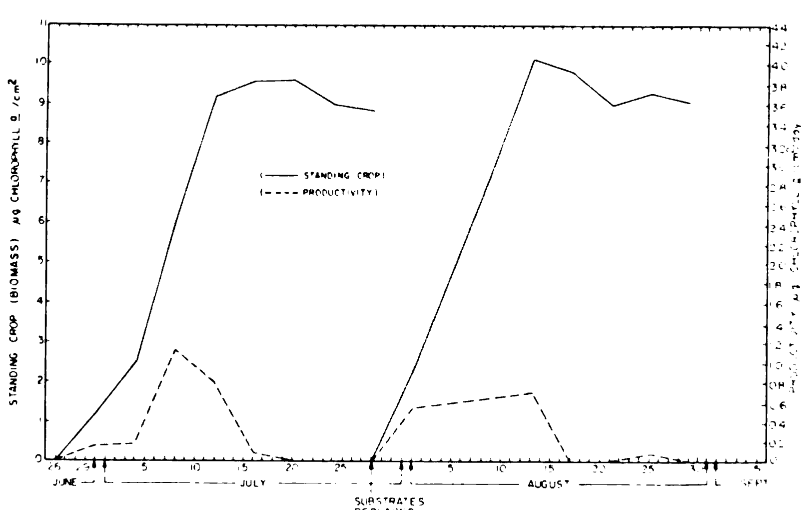

STANDING CROP (BIOMASS) µg CHLOROPHYLL a /cm²
STANDING CROP
PRODUCTIVITY
JUNE
JULY
AUGUST
SUBSTRATES REPLACED

Table 6. Standing Crop (biomass) of Primary Producers in Pond A. Lake City, Michigan, 1970.

Date	Fertilizer Added	Phytoplankton Mean Standing Crop µg/l chlorophyll a	Periphyton Mean Standing Crop µg/cm² chlorophyll a	Macrophytes Mean Standing Crop g/m²	Chemistry
6/29/70		<5			X
6/30/70			1.2		
7/ 1/70				238	X
7/ 2/70					
7/ 3/70		<5			X
7/ 4/70			2.5		
7/ 5/70					
7/ 6/70					X
7/ 7/70	X	7			
7/ 8/70	X		5.0	233	
7/ 9/70	X				X
7/10/70	X				
7/11/70	X	53			
7/12/70			9.2		X
7/13/70					
7/14/70	X				
7/15/70	X	95		248	X
7/16/70	X		9.6		
7/17/70					
7/18/70	X				X
7/19/70		80			
7/20/70	X		9.6		
7/21/70					X
7/22/70				134	
7/23/70	X	72			
7/24/70			9.0		X
7/25/70					
7/26/70					
7/27/70	X	55			X
7/28/70			8.8		
7/29/70	X			Decomposed	
7/30/70					X
7/31/70		48			
8/ 1/70	X		2.3		
8/ 2/70					X
8/ 3/70	X				
8/ 4/70		PPT			
8/ 5/70		52	Lost	0.0	X
8/ 6/70					
8/ 7/70					
8/ 8/70		29			
8/ 9/70			7.3		
8/10/70					X
8/11/70					
8/12/70		25			
8/13/70			10.1		
8/14/70					X
8/15/70					
8/16/70		<5			
8/17/70			9.8		
8/18/70					X
8/19/70	X				
8/20/70		<5			
8/21/70			9.0		
8/22/70					X
8/23/70					
8/24/70		10			
8/25/70			9.3		
8/26/70					
8/27/70					X
8/28/70		16			
8/29/70			9.1		
8/30/70					
8/31/70					
9/ 1/70		12			
9/ 2/70					
9/ 3/70					
9/ 4/70					
9/ 5/70					
9/ 6/70		8			X
9/ 7/70					

been due to plant growth being inhibited by high phosphate concentrations. Wetzel (1968) reported increasing phosphate levels above those found in natural waters results in a slight but consistent inhibition of certain species of aquatic plants. He reported 0.305 mg/l as being inhibiting the growth of some aquatic plant species.

The response of Chara to phosphate levels above 30 μg/l was found to be distinctly inhibitory (Forsberg, 1964). The decline in the macrophyte population corresponded to an increase in the phytoplankton population.

Phytoplankton

The phytoplankton population of Pond A showed an immediate response to enrichment. Phytoplankton standing crop increased from 7 μg/l chlorophyll a on July 7 to 53 μg/l on July 11 (Figure 3b). A number of researchers (Hutchinson and Bowen, 1950; Hayes and Phillips, 1958) have found that phytoplankton can absorb nutrients very quickly. Phytoplankton response to nutrients can be measured in a matter of minutes (Coffin et al., 1949) and blooms of phytoplankton in natural waters have been reported to appear very quickly (Comita and Anderson, 1959). The phytoplankton biomass in Pond A continued to increase until the maximum standing crop (95 μg/l chlorophyll a) was observed on July 15. Chlorophyll values were indicative of highly eutrophic (productive) waters which have a high concentration of phosphorus (Bachmann and Jones, 1974; EPA, 1974). Using the relationship between total phosphorus and average chlorophyll concentrations suggested by Dillon and Rigler (1974), the concentration of total phosphorus observed on July 16 (2.12 mg/l) could have resulted in over 1000 μg/l of chlorophyll a. The vertical extinction of light is effectively accomplished by dense phytoplankton populations and is directly density dependent. The rapid diminishment of light intensity, and consequent increase in photo-inhibition by high biogenic turbidity is probably a very important factor in the high levels of photosynthesis achieved in the first few centimeters of water in a hypereutrophic situation (Wetzel, 1966).

The self-shading effect described by Wetzel may be the factor limiting the phytoplankton population to 95 μg/l chlorophyll a making the 1000 μg/l suggested by Dillon and Rigler unobtainable. The highest chlorophyll a concentration observed in 150 Michigan lakes surveyed during 1974 was 125 μg/l (Michigan Department of Natural Resources Unpublished Data, 1975) indicating that indeed the phytoplankton population may be self-limiting.

Chlorophyll concentrations remained high until August 8 when a decline was noted. Turbidity decreased and chlorophyll concentrations remained low until August 20. The next application of fertilizer stimulated a small pulse of phytoplankton (maximum of 16 μg/l chlorophyll a).

The reduction in phytoplankton biomass with an apparent excess of available phosphorus (Table 4) may possibly be accounted for by examining the food web in the pond. Phytoplankton is normally consumed by zooplankton which are consumed by fish. Due to some yet undetermined set of physical, chemical and biological circumstances the phytoplankton were able to respond to the fertilization by very quickly increasing their biomass. A time lag could be expected for the zooplankton population to increase in size for the available food supply (Comita and Anderson, 1959). The increase in the zooplankton population could be expected to increase rapidly due to the fish kill which occurred on July 18. The removal of almost all fish from the pond would remove predator pressure from the zooplankton, allowing for a very rapid population expansion.

A reciprocal relationship between zooplankton and phytoplankton was indicated by Wright (1965). He found that during periods when zooplankton were increasing there was a simultaneous decrease in phytoplankton biomass (as measured by chlorophyll a) becoming practically zero at the height of the zooplankton pulse. Following the decline of the zooplankton, the population of phytoplankton increased. A very large but undetermined number of zooplankters were observed in Pond A from August 3 through August 10.

During the period of high phytoplankton populations, macrophyte biomass continued to decline. The decline in macrophyte biomass may be attributed to the increased phytoplankton reducing the light intensity or possibly changing the spectral qualities of light reaching the plants. Shading with phytoplankton is a common method used to control macrophytes in the southeastern United States (Swingle et al., 1963).

Knight et al. (1962) suggest that aquatic macrophytes have several adaptive mechanisms which allow them to grow in luxuriant beds at high light intensities which may have caused phytoplankton to be light inhibited. He felt that starting ponds (flooding) in early summer when there is little cloudiness may favor the growth of aquatic macrophytes as compared to phytoplankton, whereas the lower light intensities of a cloudy spring may favor growth of phytoplankton.

Weather records obtained from the Lake City Weather Station indicate cloudy, mostly cloudy or overcast weather from July 7 through July 11, clear skies on July 12 and overcast on July 13 and 14. The author speculates that overcast skies may have reduced light intensities sufficiently to allow the phytoplankton population, with nutrients available, to rapidly increase in biomass until they become self-limiting.

Periphyton

The periphyton community in Pond A responded to enrichment by an increase in production. The artificial substrate method does not truly measure production; it measures the rate of colonization (Wetzel, 1964). The estimate of colonization or production is the additional $\mu g/cm^2/day$ of chlorophyll a which has occurred by attachment and growth of organisms since the last sample was taken. The two sampling dates prior to enrichment had production rates of 0.30 and 0.33 μg chlorophyll a cm^2/day. One day after fertilization production was determined to be 1.12 μg chlorophyll a cm^2/day. Standing crop of periphyton for the next 16 days averaged 9.2 μg chlorophyll a cm^2. These findings are similar to those of Szczepánski and Szczepánska (1966) who measured chlorophyll content of algae on slides exposed for various times. They found a rapid rise in the amount of chlorophyll for the first two weeks or so, then a period of about five weeks of more or less constant chlorophyll levels. The carrying capacity or maximum biomass of periphyton which could utilize the artificial substrate was determined to be approximately 9 μg chlorophyll a cm^2. Slādeček and Sládečkvoá (1964) defined turnover time as the time needed for renewal of the periphyton community. The turnover time for the periphyton community in Pond A with the first set of substrates was 16 days.

Artificial substrates were replaced on July 28. On August 1 when the first samples were taken, the rate of production was 0.56 μg chlorophyll a cm^2/day. This low rate even with excessive nutrients available was probably the result of initial colonization. During the first three days of exposure, substrates placed in artificial streams by Kevern (1962) showed little or no measurable periphyton growth. He subtracted the three days from total exposure time for calculating production rates. This situation did not appear to hold completely true for the nutrient enriched Pond A.

Due to a laboratory accident, the samples for August 5 were destroyed. Production rates on August 9 and 13 were 0.63 and 0.70 μg chlorophyll a cm^2/day, respectively. These rates were slightly lower than those observed during the first exposure period. Carrying capacity averaged 9.4 μg chlorophyll a cm^2. Turnover time was 16 days. In 1968 Gehring (1969) found the turnover time for these same ponds to be 16-22 days.

Differences in periphyton standing crop and production are apparent from Figure 3C. Standing crop is the quantity or biomass of material which was present when the sample was taken, and productivity is the increase in biomass between sampling dates. Productivity was high (July 5-10 and August 1-13) when biomass was being accumulated on the plexiglass plates. On July 20 and August 15, productivity appeared to decline to near zero while the standing crop remained fairly constant. However, production was taking place to make up for losses from the standing crop. Materials can be lost from the standing crop via grazing organisms, cell sluffing and cell decay. Production of the periphyton community was approximately equal to the loss of material from the standing crop.

Physicochemical Characteristics of Pond B

Fertilization of Pond B was begun on July 14, 1970, seven days after the fertilization of Pond A started. It received the second highest rate of nitrogen and phosphorus loading, 3.20 and 0.54 kg, respectively.

Temperature

A complete description of the temperature variation observed during the study and its impact on the ecosystem can be found in the temperature section of the Results and Discussion, Pond A (page 30).

Oxygen

Prior to enrichment, oxygen concentrations averaged 9.1 mg/l (104 percent saturation). The maximum observed concentration of oxygen occurred late in July (between July 19 and 28). Levels of 13 mg/l (142-155 percent saturation) of oxygen were reached on two occasions in the pond. Following this peak, oxygen levels declined below 100 percent saturation and remained there for the balance of the study period. Primary production, as estimated by standing crop, was particularly intense during the periods of the maxima in direct correlation with the development of high oxygen concentrations.

The low values for dissolved oxygen occurred on August 18 when levels of dissolved oxygen and percent saturation were 5.9 mg/l and 65 percent, respectively. These reduced levels of dissolved oxygen occurred during a period when there was a high standing crop of filamentous algae covering much of the surface of Pond B. It can be theorized that the floating algae which was beginning to decay exerted a high B.O.D. on the pond water, reducing the dissolved oxygen. In addition, the floating algae may have effectively kept light from reaching other primary producers (shading effect).

Alkalinity and pH

Experimental Pond B contained hardwater with a total alkalinity which ranged from 61 to 108 mg/l and a mean of 77 mg/l. Total alkalinity was elevated on July 19, probably due to the use of potassium nitrate fertilizer used to enrich the ponds. Phenolphthalein alkalinity was low (< 10) except for a 9-day period near the end of July when it averaged 24 mg/l.

The elevated phenolphthalein alkalinity occurred during a period of high pH values (> 9.1) and high phytoplankton standing crop. The depletion in the bicarbonate alkalinity occurred in the pond during the period of maximal productivity. Such removal of bicarbonate from the water is largely biogenic in origin.

Phosphorus and Nitrate Concentrations

Total phosphorus concentrations in Pond B ranged from 0.01 mg/l on July 13 to 0.75 mg/l on August 8. Total phosphorus measurements were always somewhat lower than the expected theoretical concentration (Table 8). Observed concentrations of total phosphorus ranged from 48 to 85 percent of the theoretical values. A questionable value of 31 percent was observed but not used. Soluble phosphorus ranged from less than 0.01 mg/l to 0.65 mg/l (Table 7).

Macrophytes

Standing crop of macrophytes in Pond B remained relatively constant after fertilization began, $x = 201$ g/m^2 (Figure 4, Table 9). Biomass appeared to have increased on the August 12 sampling date; however, the addition was not due entirely to macrophyte growth. A bloom of filamentous algae occurred the week before and it was impossible to remove all of this material from the plants prior to their being dried and weighed. The periphyton material which remained on the aquatic plants accounted for some of the weight addition. Subsequent sampling of the macrophytes found the plants to be heavily overgrown with filamentous algae and in poor physiological condition. From August 12 until the termination of the study, the macrophyte population continued to decline. The biomass on the last sampling date (September 1) had been reduced to 120 g/m^2.

Table 7. Concentration of Selected Chemical Constituents[a] in Pond B, Lake City, Michigan, 1970.

	Parameter						
Date	Dissolved Oxygen	pH	Total Alkalinity	Phenolphthalein Alkalinity	Ortho-Phosphorus	Total Phorphorus	Nitrate-Nitrogen
6/29/70	8.9 (108)[b]	8.8	66.0	4.0	<0.01	0.01	0.03
7/1/70	9.2 (107)	8.9	63.0	5.0	<0.01	0.01	0.03
7/4/70	8.0 (87)	8.7	72.0	6.0	<0.01	0.02	0.02
7/7/70	8.1 (95)	8.7	70.0	4.0	<0.01	0.02	0.02
7/10/70	9.3 (104)	8.8	76.0	4.0	<0.01	0.01	0.03
7/13/70	10.0 (120)	9.0	79.0	8.0	<0.01	0.01	0.02
7/14/70	10.4 (124)	8.5	68.0	7.0	0.22	0.24	0.11
7/16/70	11.1 (128)	8.7	76.0	6.0	0.40	0.45	0.26
7/19/70	13.0 (142)	9.2	108.0	36.0	0.58	0.63	1.07
7/23/70	12.3 (136)	9.5	67.0	21.0	0.65	0.72	2.44
7/25/70	11.0 (122)	9.6	61.0	21.0	0.47	0.56	1.95
7/28/70	13.0 (155)	9.7	65.0	19.0	0.43	0.58	2.32
7/31/70	7.9 (95)	9.1	59.0	9.0	0.42	0.53	2.53
8/3/70	7.6 (86)	9.0	61.0	5.0	0.60	0.75	3.76
8/6/70	8.1 (93)	8.7	66.0	5.0	0.43	0.54	2.48
8/10/70	6.5 (78)	8.3	79.0	0.0	0.15	0.27	1.41
8/14/70	6.1 (73)	7.9	87.0	0.0	0.05	0.11	0.81
8/18/70	5.9 (65)	8.0	84.0	0.0	0.01	0.04	0.46
8/22/70	5.8 (64)	7.8	92.0	0.0	0.17	0.21	0.96
8/27/70	6.6 (87)	8.1	79.0	0.0	0.02	0.11	0.31
9/4/70	7.8 (89)	8.4	68.0	3.0	<0.01	0.04	0.05

[a]Expressed in mg/l except pH which is in standard units.

[b]Percent saturation.

Table 8. Observed and Theoretical[a] Total Phosphorus Concentrations[b] in Pond b, Lake City, Michigan, 1970.

Date	Theoretical Concentration	Observed Concentration	Percent($\frac{\text{Observed}}{\text{Theoretical}}$)
July 14	0.29	0.24	83
July 16	0.77	0.45	58
July 19	0.77	0.63	82
July 23	0.93	0.72	77
July 25	0.71	0.56	79
July 28	0.73	0.58	79
July 31	0.71	0.53	75
August 3	0.99	0.75	76
August 6	0.67	0.54	81
August 10	0.39	0.27	69
August 14	0.23	0.11	48
August 18	0.13	0.04	31
August 22	0.26	0.21	81
August 27	0.13	0.11	85
September 4	0.05	0.04	80
			$\bar{x}$ 72.3 percent

[a]Takes into account flushing rate of pond.

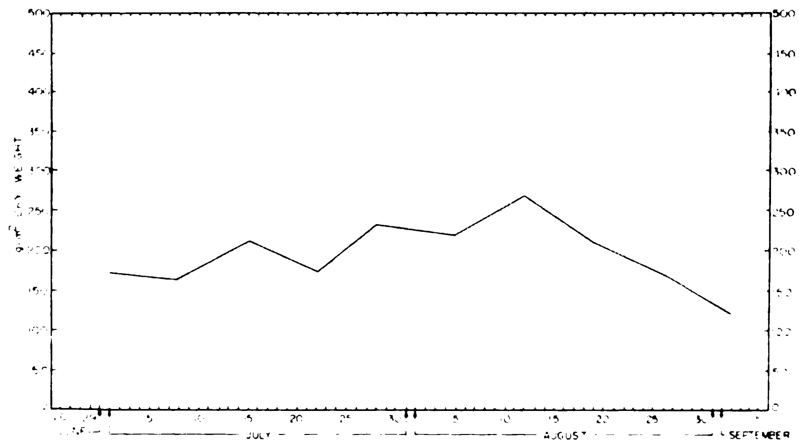
[b]mg/l.

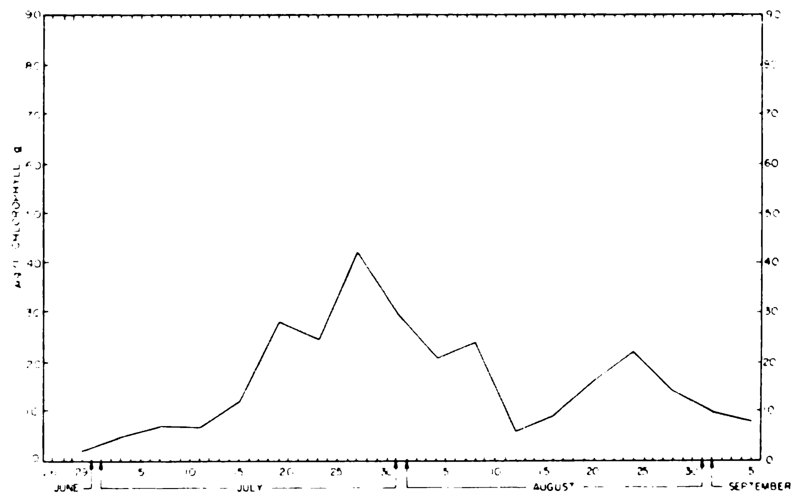
Figure 4a. Macrophyte biomass (dry weight) in Pond B, Lake City, Michigan, 1970.

Figure 4b. Phytoplankton biomass as determined by chlorophyll a concentrations in Pond B, Lake City, Michigan, 1970.

Figure 4c. Periphyton production rates and standing crop in Lake City, Michigan, 1970.

Pond B as determined by chlorophyll a concentration (————————standing crop; --------------production).


JULY
AUGUST
SEPTEMBER


90
40
30
20
10
0
JUNE
JULY
AUGUST
SEPTEMBER

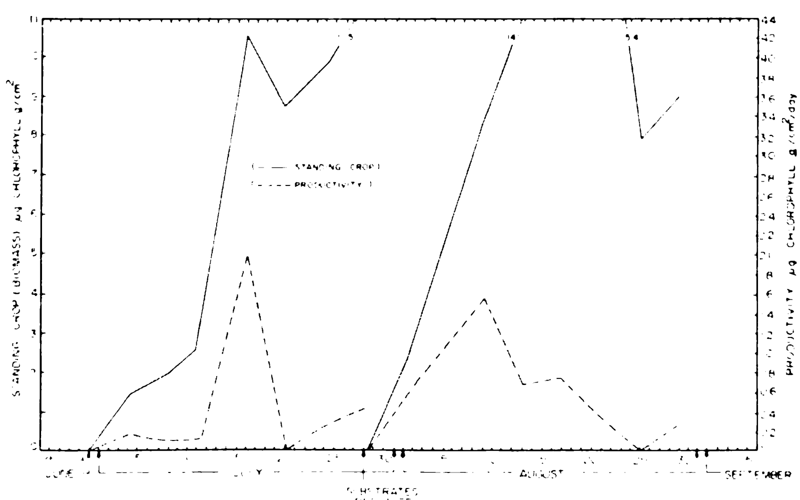

STANDING CROP
AUGUST
SEPTEMBER

Table 9. Standing Crop (biomass) of Primary Producers in Pond B. Lake City, Michigan, 1970. Dates of Fertilization and Water Chemistry Analysis Indicated.

Date	Fertilizer Added	Phytoplankton Mean Standing Crop μg/l chlorophyll a	Periphyton Mean Standing Crop μg/cm² chlorophyll a	Macrophytes Mean Standing Crop g/m²	Chemistry
6/29/70		<5			X
6/30/70			<1		
7/ 1/70				183	X
7/ 2/70					
7/ 3/70		5			
7/ 4/70			1.4		X
7/ 5/70					
7/ 6/70					
7/ 7/70		7			X
7/ 8/70			1.9	164	
7/ 9/70					
7/10/70					X
7/11/70		7			
7/12/70			2.6		
7/13/70					X
7/14/70	X				
7/15/70	X	12		212	
7/16/70	X		10.6		X
7/17/70	X				
7/18/70	X				
7/19/70		28			X
7/20/70	X		8.7		
7/21/70					
7/22/70				187	X
7/23/70	X	25			
7/24/70			9.7		
7/25/70					X
7/26/70					
7/27/70	X	42			
7/28/70			11.5		X
7/29/70	X			240	
7/30/70					
7/31/70		30	2.24		X
8/ 1/70	X				
8/ 2/70					
8/ 3/70	X				X
8/ 4/70		21			
8/ 5/70			–	225	
8/ 6/70					X
8/ 7/70					
8/ 8/70		24	8.4		
8/ 9/70					
8/10/70					X
8/11/70					
8/12/70		6		270	
8/13/70			11.1		
8/14/70					X
8/15/70					
8/16/70		9			
8/17/70			14.0		
8/18/70					X
8/19/70	X			212	
8/20/70		16			
8/21/70			15.4		
8/22/70					X
8/23/70					
8/24/70		22			
8/25/70			7.8		
8/26/70				171	
8/27/70					X
8/28/70		14			
8/29/70			8.9		
8/30/70					
8/31/70					
9/ 1/70		10			
9/ 2/70				120	
9/ 3/70					
9/ 4/70					
9/ 5/70		8			X
9/ 6/70					
9/ 7/70					

Ball and Tanner (1951) reported a similar occurrence while fertilizing a small lake. They observed that after fertilization a bloom of filamentous algae appeared and covered the submerged vegetation. The combination of reduced light and the burden of the filamentous algae all but destroyed the Chara and potamogentons.

Phytoplankton

Phytoplankton standing crop in Pond B responded immediately to enrichment. Chlorophyll a increased from 7 μg/l on July 11 to a maximum of 42 μg/l on July 27. During this same time period, total phosphorus concentration increased from < 0.01 mg/l to 0.56 mg/l. Phytoplankton biomass gradually declined reaching 6 μg/l chlorophyll a on August 12. This decrease in phytoplankton biomass corresponds with the appearance of the filamentous algae bloom described in the preceding section. Estimates were that about 25 percent of the surface area of the ponds was covered with mats of floating algae. From August 7 until August 12 approximately 33 cubic feet of algae (wet) were removed from the pond's surface. The 25 percent of the pond surface which was covered by floating mats of algae resulted in the shading out of light to other primary producers. Hence, the decrease in phytoplankton may have been the result of decreased availability of light. No dramatic increase in the zooplankton population was visible, unlike the pulse of zooplankton noted in Pond A.

An increase in the phytoplankton standing crop was noted as the filamentous algae bloom subsided and fertilization was resumed.

Periphyton

Periphyton in Pond B had an accrual rate of less than 0.20 $\mu g/cm^2$/day chlorophyll a prior to enrichment. Accrual rate increased to 2.0 after enrichment then decreased as the substrate reached its carrying capacity estimated to be 10.6 $\mu g/cm^2$ chlorophyll a. Turnover time was determined to be 20 days.

The second set of substrates in Pond B had a higher initial accrual rate than the previous set (0.56 compared to < 0.20). Turnover time was less than 16 days and the substrate carrying capacity was greater (11.4 $\mu g/cm^2$ compared to 10.6 $\mu g/cm^2$ chlorophyll a).

During the second colonization study, a filamentous algae bloom appeared. The filamentous algae bloom occurred during a period when water temperatures were high and relatively stable. Ball and Tanner (1951) while fertilizing a small lake observed that filamentous algae occurrence was more conspicuous during a period in early August, when there was a prolonged period of hot quiet weather. At Lake City, filaments were visible on the artificial substrates during the bloom. However, the exact quantification of this material was not possible. The filamentous form of algae did not appear to be anchored securely to the substrates. Pieces of the mats were observed on several occasions to break from the substrate and float to the surface. When maximum care was taken in removing the substrates for analysis, some material was observed to be lost. Evidence of this can be seen in large variation in the biomass on the last six sampling dates.

Although no exact measurement of the quantity of filamentous algae produced during the bloom was made, approximately 33 cubic feet of filamentous algae was removed. Most of the filamentous algae produced stayed in the pond. A great deal of it could be observed in various stages of decomposition throughout the pond after the bloom had subsided. This material contributed to total pond primary production, but was not accurately measured.

Pieczýnska (1971) reported that filamentous algae growing on and around plants in the shallow zone of a lake may produce 750 mg/dm^2 of material (dry weight) and contributes significantly to the total primary production of some lakes.

Initial response to enrichment in Pond B was an increase in phytoplankton biomass followed by a bloom of filamentous algae. The filamentous algae subsequently caused a reduction in both the phytoplankton and macrophyte standing crops.

Physicochemical Characteristics of Pond C

Pond C was the last pond to be fertilized (July 17, 1970), ten and three days, respectively, after Ponds A and B were first fertilized. It received the lowest rate of nitrogen and phosphorus loading, 1.45 and 0.24 kg, respectively.

Temperature

A complete description of the temperature variation observed during the study and its impact on the ecosystem can be found in the temperature section of the Results and Discussion, Pond A (page 30).

Oxygen

Enrichment had no apparent effect on the dissolved oxygen concentration in Pond C (Table 10). Prior to fertilization, oxygen concentrations averaged 9.1 mg/l (105 percent saturation). The maximum observed dissolved oxygen concentration occurred in mid-July (on July 13 and 17). Levels of 11.2 mg/l (133 percent saturation) of oxygen were reached in the pond. On July 8, 1970, a heavy rainstorm occurred at Lake City; 0.42 inches of rain fell on that date. Between July 13 and 17, 0.49 inches of rain fell on the Lake City Experiment Station. Rainfall can carry substantial quantities of nutrients (Weibel, 1969). It is possible that the natural nutrient contributions in rainfall stimulated productivity in the pond increasing the dissolved oxygen concentration. Following this peak and the commencement of enrichment, percent saturation of dissolved oxygen declined, averaging 97 percent for the remainder of the study.

Alkalinity and pH

Experimental Pond C had an average total alkalinity of 82 mg/l, and is classified as hard water. Phenolphthalein alkalinity was low (< 10) except for two occasions when it rose to 10 and 11 mg/l. During this same time interval, dissolved oxygen levels were 10.2 and 11.0 mg/l. High pH levels (> 8.5) occurred simultaneously. The depletion of the bicarbonate alkalinity and rise in pH occurred in the pond during periods of high productivity.

Table 10. Concentration of Selected Chemical Constituents[a] in Pond C, Lake City, Michigan, 1970.

	Parameter						
Date	Dissolved Oxygen	pH	Total Alkalinity	Phenophthalein Alkalinity	Ortho-Phosphorus	Total Phosphorus	Nitrate-Nitrogen
6/29/70	8.0 (96)[b]	8.3	87.0	0.0	<0.01	0.01	0.02
7/2/70	8.2 (97)	8.8	76.0	2.0	<0.01	0.01	0.03
7/4/70	7.5 (84)	8.2	85.0	0.0	<0.01	0.02	0.02
7/7/70	8.7 (100)	8.4	78.0	3.0	<0.01	0.02	0.02
7/10/70	9.6 (106)	8.5	74.0	5.0	<0.01	0.01	0.03
7/13/70	11.2 (133)	8.6	68.0	11.0	<0.01	0.01	0.02
7/16/70	10.8 (122)	8.4	83.0	6.0	<0.01	0.01	0.03
7/17/70	11.2 (130)	8.1	79.0	0.0	0.24	0.28	0.83
7/19/70	8.0 (88)	8.3	77.0	3.0	0.47	0.50	1.07
7/23/70	9.0 (99)	8.6	77.0	5.0	0.51	0.56	0.9
7/25/70	9.0 (100)	8.5	71.0	6.0	0.23	0.31	0.06
7/28/70	10.0 (118)	8.7	71.0	10.0	0.25	0.35	0.21
7/31/70	8.2 (99)	9.0	81.0	6.0	0.25	0.32	0.07
8/3/70	8.1 (92)	8.7	69.0	3.0	0.42	0.57	0.23
8/6/70	8.3 (96)	8.8	80.0	.5	0.14	0.24	0.02
8/10/70	7.8 (95)	8.7	74.0	0.0	0.02	0.07	<0.02
8/14/70	9.0 (110)	9.2	78.0	0.0	<0.01	0.03	<0.02
8/18/70	8.6 (97)	8.0	79.0	0.0	<0.01	0.03	<0.02
8/22/70	7.0 (78)	8.2	94.0	0.0	0.08	0.12	0.03
8/27/70	7.4 (83)	8.0	87.0	0.0	<0.01	0.03	<0.02
9/4/70	7.1 (81)	7.9	83.0	0.0	<0.01	0.03	<0.02

[a]Expressed in mg/l except pH which is in standard units.

[b]Percent saturation.

Phosphorus and Nitrate Concentrations

Total phosphorus concentrations in Pond C ranged from 0.01 mg/l prior to enrichment to a maximum of 0.51 mg/l on August 3. Total phosphorus concentrations were always lower than the expected theoretical concentration. Observed concentrations of total phosphorus ranged from 85 to 93 percent of the theoretical concentrations (Table 11). Soluble phosphorus ranged from less than 0.01 mg/l to 0.51 mg/l (Table 10).

Maximum nitrate nitrogen values (1.07 mg/l) occurred on July 19. Successive applications of nitrogen fertilizer did not result in the "build-up" in the nitrate concentration. A "build-up" of nitrate over the sample period did occur in Ponds A and B.

Macrophytes

The standing crop of macrophytes in Pond C was increasing prior to enrichment and continued to increase for the first week after enrichment (Figure 5, Table 12). The July 30 sample contained slightly less biomass than the sample from the previous sampling period. This may be due to the lack of homogeneity in natural conditions which reduces the quantitative validity that can be attained with this sampling technique. Biomass appeared to have increased on the August 12 sampling date. The addition was not due entirely to macrophyte growth. The filamentous algae bloom described in Pond B also occurred in Pond C. An attempt was made to remove as much of the filamentous algae from the harvested plants as was possible. However, it was impossible to remove all the algae from the

Table 11. Observed and Theoretical[a] Total Phosphorus Concentrations[b] in Pond C, Lake City, Michigan, 1970

Date	Theoretical Concentration	Observed Concentration	Percent(Observed / Theoretical)
July 17	0.33	0.28	85
July 19	0.43	0.40	93
July 23	0.61	0.56	92
July 25	0.34	0.31	91
July 28	0.39	0.35	90
July 31	0.35	0.32	91
August 3	0.66	0.57	86
August 6	0.28	0.24	86
August 10	0.08	0.07	88
August 14	0.03	0.03	-
August 18	0.00	0.03	-
August 22	0.14	0.12	86
August 27	0.03	0.03	-
September 4	0.01	0.03	-
			$\bar{x}$ 89 percent

[a]Takes into account flushing rate of pond.

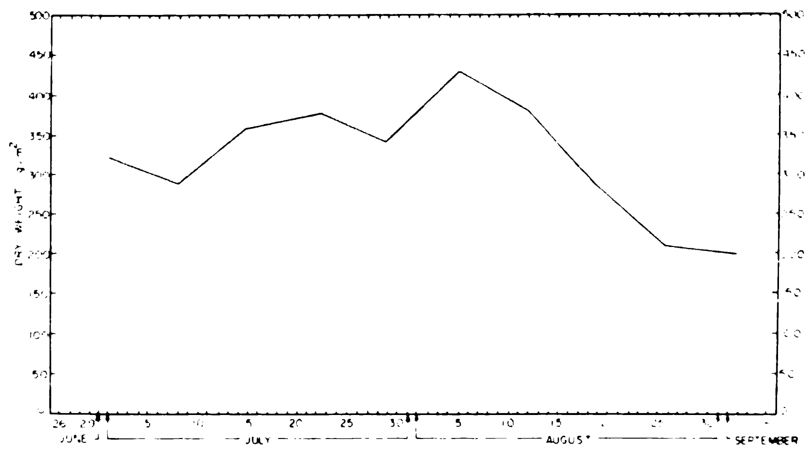
[b]mg/1.

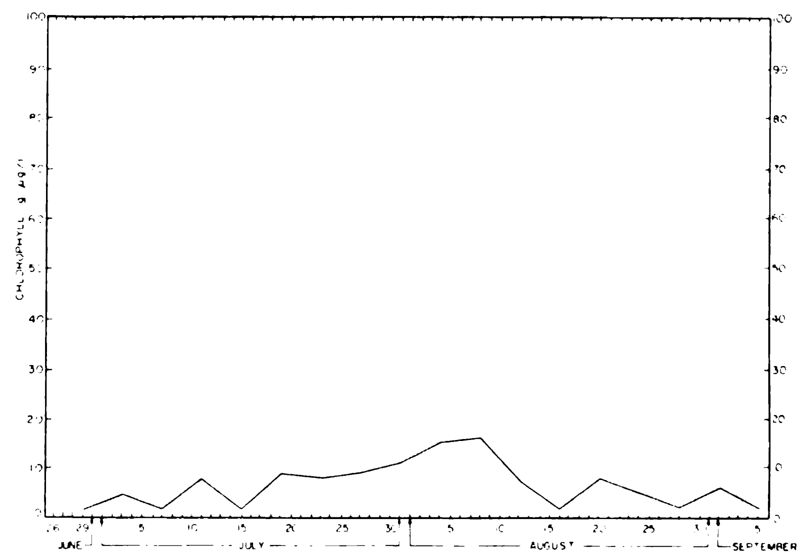
Figure 5a. Macrophyte biomass (dry weight) in Pond C, Lake City, Michigan, 1970.

Figure 5b. Phytoplankton biomass as determined by chlorophyll *a* concentrations in Pond C, Lake City, Michigan, 1970.

Figure 5c. Periphyton production rates and standing crop in Pond C, Lake City, Michigan, 1970.

(————————standing crop; ----------production rate)


JUNE
JULY
AUGUST
SEPTEMBER


JUNE
JULY
AUGUST
SEPTEMBER

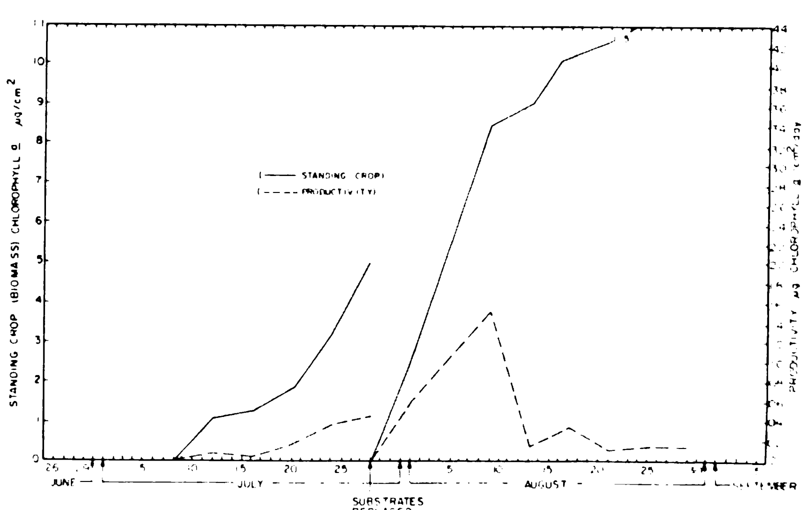

STANDING CROP (BIOMASS) CHLOROPHYLL a µg/cm²
STANDING CROP
PRODUCTIVITY
JUNE
JULY
AUGUST
SUBSTRATES
REPLACED

Table 12. Standing crop (biomass) of Primary Producers in Pond C. Lake City, Michigan, 1970. Dates of Fertilization and Water Chemistry Analysis Indicated.

Dates	Fertilizer Added	Phytoplankton Mean Standing Crop μg/l chlorophyll a	Periphyton Mean Standing Crop μg/cm² chlorophyll a	Macrophytes Mean Standing Crop g/m²	Chemistry
6/29/70		<5			X
6/30/70			<1		
7/ 1/70				320	X
7/ 2/70					
7/ 3/70		5			
7/ 4/70			<1		X
7/ 5/70					
7/ 6/70					
7/ 7/70		<5			X
7/ 8/70			<1	290	
7/ 9/70					
7/10/70					X
7/11/70		8			
7/12/70			1.1		
7/13/70					X
7/14/70					
7/15/70		<5		360	
7/16/70			1.2		X
7/17/70	X				
7/18/70	X				
7/19/70		9			X
7/20/70	X		1.8		
7/21/70					
7/22/70				375	X
7/23/70	X	8			
7/24/70			3.2		
7/25/70					X
7/26/70					
7/27/70	X	9			
7/28/70			4.9		X
7/29/70	X			340	
7/30/70					
7/31/70		11			X
8/ 1/70	X		2.4		
8/ 2/70					
8/ 3/70	X				X
8/ 4/70		15			
8/ 5/70			-	430	
8/ 6/70					X
8/ 7/70					
8/ 8/70		16			
8/ 9/70			8.4		
8/10/70					X
8/11/70					
8/12/70		7		382	
8/13/70			8.8		
8/14/70					X
8/15/70					
8/16/70		<5			
8/17/70			9.3		
8/18/70					X
8/19/70	X			285	
8/20/70		8			
8/21/70			10.5		
8/22/70					X
8/23/70					
8/24/70		5			
8/25/70			11.1		
8/26/70				210	
8/27/70					X
8/28/70		<5			
8/29/70			11.7		
8/30/70					
8/31/70					
9/ 1/70		6			
9/ 2/70					
9/ 3/70				200	
9/ 4/70					
9/ 5/70					
9/ 6/70		<5			X
9/ 7/70					

macrophytes prior to their being dried and weighed. Subsequent sampling of the macrophytes found the plants to be heavily overgrown with filamentous algae and in poor physicological condition. From August 12 until termination of the study the macrophyte population continued to decline. Biomass on the last sampling date was 200 gm/m^2. Macrophyte biomass was reduced 37.5 percent during the study.

Phytoplankton

Phytoplankton standing crop showed little initial response to the addition of nitrogen and phosphorus. Biomass as measured by chlorophyll a concentration did increase to a maximum of 16 μg/l on August 8. By August 8, a bloom of filamentous algae was spreading throughout the pond and subsequent sampling of phytoplankton showed a decrease in the biomass. The phytoplankton population increased to prefertilization levels after the bloom and remained at the low levels for the remainder of the season.

Periphyton

Periphyton in Pond C had a maximum accrual rate of 0.43 $\mu g/cm^2/day$ chlorophyll a. The 28-day time interval the first series of substrates was submerged was not sufficient to establish the turnover time or substrate carrying capacity with such a slow accrual rate.

During the second colonization study a filamentous algae bloom began to materialize. Initial rate of periphyton colonization was 0.6 $\mu g/cm^2/day$. The sample taken at the onset of the bloom showed a rate of 1.5 $\mu g/cm^2/day$. This rate was in all probability higher. Some filaments of periphyton were always observed to be lost when the substrates were removed for analysis. Substrate carrying capacity was determined to be 10.85 $\mu g/cm^2$. Turnover time was 24 days. During the period when the maximum bloom was in progress, approximately 21 cubic feet of algae (wet weight) was removed from the surface of Pond C.

The response in Pond C to enrichment was a doubling of the phytoplankton biomass followed by a bloom of filamentous algae which resulted in a decline in the macrophyte and phytoplankton standing crop.

Physicochemical Characteristics of Pond D

Pond D served as a control during the study and was not fertilized.

Temperature

A complete description of the temperature variation observed during the study and its impact on the ecosystem can be found in the temperature section of the Results and Discussion, Pond A (page 30).

Oxygen

Average oxygen concentration during the study period was 8.0 mg/l with 96 percent saturation (Table 13). The maximum observed concentration of oxygen occurred in mid-July. A maxima of 10.9 mg/l (130 percent saturation) was reached on July 13. The maximum oxygen concentration observed in Pond D coincided with that observed in Pond C. These maxima were probably the results of climatic conditions

Table 13. Concentration of Selected Chemical Constituents[a] in Pond D, Lake City, Michigan, 1970.

	Parameter						
Date	Dissolved Oxygen	pH	Total Alkalinity	Phenolphthalein Alkalinity	Ortho-Phosphorus	Total Phosphorus	Nitrate-Nitrogen
6/29/70	8.0 (96)[b]	8.4	76.0	3.0	<0.01	0.01	0.03
7/1/70	8.4 (98)	8.8	74.0	4.0	<0.01	0.01	0.04
7/4/70	7.7 (84)	8.5	79.0	2.0	<0.01	0.02	0.02
7/7/70	8.6 (99)	8.6	80.0	4.0	<0.01	0.01	0.02
7/10/70	8.7 (96)	8.8	78.0	3.0	<0.01	0.02	0.04
7/13/70	10.9 (130)	8.8	74.0	10.0	<0.01	0.02	0.02
7/16/70	9.5 (110)	8.7	75.0	7.0	<0.01	0.02	0.02
7/19/70	9.0 (98)	8.5	80.0	5.0	<0.01	0.02	0.03
7/23/70	8.3 (90)	8.8	78.0	11.0	<0.01	0.01	0.03
7/25/70	8.5 (95)	8.7	76.0	10.0	<0.01	0.03	0.04
7/28/70	8.4 (99)	8.7	78.0	7.0	<0.01	0.02	0.02
7/31/70	8.0 (95)	8.5	70.0	4.0	<0.01	0.02	0.02
8/3/70	7.7 (86)	8.8	66.0	4.0	<0.01	0.01	0.02
8/6/70	7.9 (92)	8.7	68.0	5.0	<0.01	0.03	0.02
8/10/70	8.2 (98)	8.4	65.0	6.0	<0.01	0.02	0.02
7/14/70	8.6 (104)	8.7	66.0	6.0	<0.01	0.03	0.03
8/18/70	8.1 (92)	8.5	74.0	4.0	<0.01	0.02	0.02
8/22/70	7.9 (87)	8.6	72.0	3.0	<0.01	0.02	0.02
8/27/70	8.3 (91)	8.8	74.0	7.0	<0.01	0.03	0.02
9/4/70	7.7 (86)	8.5	68.0	4.0	<0.01	0.04	0.03

[a]Expressed in mg/l except pH which is in standard units.

[b]Percent saturation.

favoring high production during this period. The standing crop of phytoplankton as estimated by chlorophyll a concentration was near the maximal observed during this study on July 13. The high oxygen concentrations were caused by high primary production during this period. Following this peak, oxygen levels in the pond were slightly reduced but still remained between 86 and 110 percent saturation for the remainder of the study.

Alkalinity and pH

Experimental Pond D contained hard water with a total alkalinity which ranged from 65.0 to 80.0 mg/l and a mean of 73 mg/l. Phenolphthalein alkalinity was elevated on those occasions when pH was elevated. The increases in pH and phenolphthalein alkalinity were biogenic.

Phosphorus and Nitrate Concentrations

Total phosphorus concentrations in Pond D ranged from 0.01 to 0.04 with a mean value of 0.02 mg/l. This value is in agreement for phosphorus concentrations found in natural waters. Juday and Birge (1931) determined the mean phosphorus concentration of 479 Wisconsin lakes to be a mean value of 0.023 mg/l. Ten years later, Hutchinson (1941) found phosphorus concentration of Linsley Pond to be 0.021 mg/l.

Soluble phosphorus was below the level of detectability (< 0.01 mg/l) during the study period. Armstrong and Schindler (1971) reported an average dissolved phosphorus concentration of 0.007 mg/l from 40 small lakes within a small area of the Canadian shield in northwestern Ontario. Total and soluble phosphorus levels do not appear to be elevated beyond the levels which naturally occur in the surface water of this region.

Nitrate nitrogen concentrations ranged from 0.02 mg/l to 0.04 mg/l with a mean value of 0.025 mg/l. On July 1, 10 and 25, nitrate levels were 0.04 mg/l. On these dates, or within 48 hours preceding these dates, rainfall of 0.20, 0.42 and 0.55 inches, respectively, was recorded. This may well account for the slight elevations in nitrate concentrations.

Mean nitrate concentrations of 0.02 mg/l are in agreement with the data reported by other researchers in natural lentic waters of this region. Armstrong and Schindler (1971) reported mean nitrate nitrogen values of 0.01 mg/l (range < 0.001 to 0.04 mg/l) for surface samples taken in 1968 and 1969 from ten Canadian Experimental Lakes.

Macrophytes

Standing crop of macrophytes in Pond D increased at the onset of the study from 277 g/m^2 to 330 g/m^2 (Table 14). A slight decline in biomass was noted for the next two sampling dates (Figure 6). Macrophyte biomass increased from July 21 until the termination of the study. A maximum standing crop of 380 g/m^2 was observed on September 3, 1970. The net primary production of the higher aquatic plant community as determined by the harvest method was 1.69 g/m^2/day. These findings are in strong agreement with those reported by Knight, Ball and Hooper (1962), who reported a primary production rate of 1.45 g/m^2/day in a Lake City pond used as a control during another study.

Table 14. Standing Crop (biomass) of Primary Producers in Pond D. Lake City, Michigan, 1970. Dates of Fertilization and Water Chemistry Analysis Indicated.

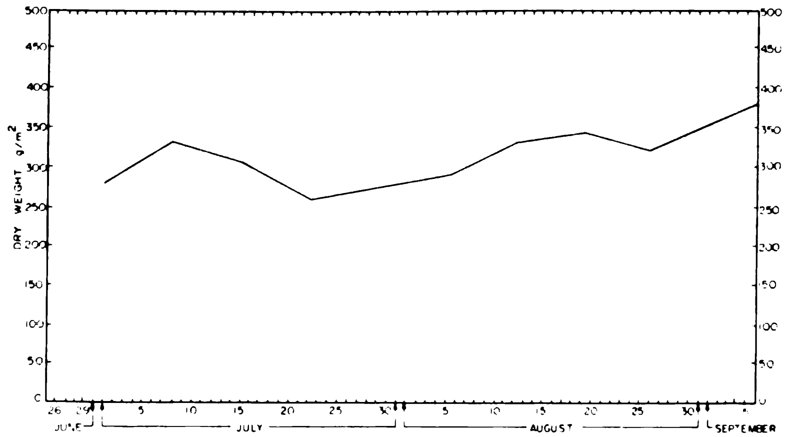
Date	Fertilizer Added	Phytoplankton Mean Standing Crop μg/l chlorophyll a	Periphyton Mean Standing Crop μgl/cm² chlorophyll a	Macrophytes Mean Standing Crop g/m²	Chemistry
6/29/70		<5			X
6/30/70			<1		
7/ 1/70				277	X
7/ 2/70					
7/ 3/70		<5			
7/ 4/70			<1		X
7/ 5/70					
7/ 6/70					
7/ 7/70		5			X
7/ 8/70			1.0	330	
7/ 9/70					
7/10/70					X
7/11/70		7			
7/12/70			1.1		
7/13/70					X
7/14/70					
7/15/70		5		305	
7/16/70			1.2		X
7/17/70					
7/18/70					
7/19/70		<5			X
7/20/70			1.3		
7/21/70					
7/22/70				260	X
7/23/70		6			
7/24/70			1.5		
7/25/70					X
7/26/70					
7/27/70		7			
7/28/70			1.8		X
7/29/70				271	
7/30/70					
7/31/70		8			X
8/ 1/70			<1		
8/ 2/70					
8/ 3/70					X
8/ 4/70		5			
8/ 5/70			---	293	
8/ 6/70					X
8/ 7/70					
8/ 8/70		<5			
8/ 9/70			<1		
8/10/70					X
8/11/70					
8/12/70		<5		350	
8/13/70			1.0		
8/14/70					X
8/15/70					
8/16/70		<5			
8/17/70			1.0		
8/18/70					X
8/19/70				348	
8/20/70		6			
8/21/70			1.1		
8/22/70					X
8/23/70					
8/24/70		7			
8/25/70			1.0		
8/26/70				362	
8/27/70					X
8/28/70		5			
8/29/70			1.1		
8/30/70					
8/31/70					
9/ 1/70		8			
9/ 2/70					
9/ 3/70					
9/ 4/70					
9/ 5/70		7			X
9/ 6/70					
9/ 7/70					

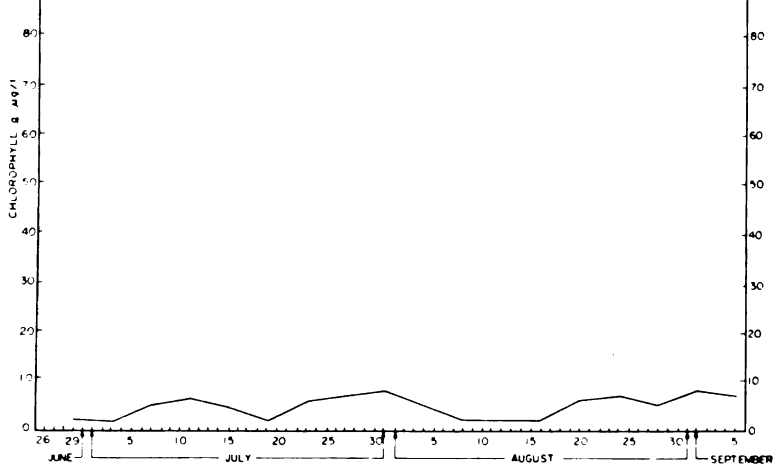
Figure 6a. Macrophyte biomass (dry weight) in Pond D, Lake City, Michigan, 1970.

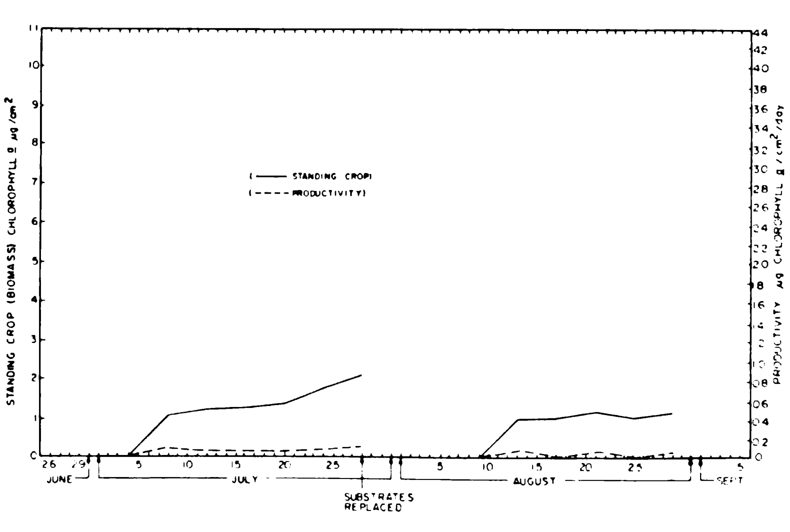
Figure 6b. Phytoplankton biomass as determined by chlorophyll a concentration in Pond D, Lake City, Michigan, 1970.

Figure 6c. Periphyton production rates and standing crop in Pond D, Lake City, Michigan, 1970.

(——————standing crop; --------------production)


DRY WEIGHT g/m2
JUNE
JULY
AUGUST
SEPTEMBER


CHLOROPHYLL a µg/l
JUNE
JULY
AUGUST
SEPTEMBER


STANDING CROP (BIOMASS) CHLOROPHYLL a µg/cm2
(STANDING CROP)
(PRODUCTIVITY)
JUNE
JULY
AUGUST
SEPT
SUBSTRATES REPLACED

Phytoplankton

The phytoplankton population, as determined by chlorophyll a concentrations, ranged from below the level of detectability (< 5 μg/l) to 8 μg/l with a mean of 4.6 μg/l (Figure 6B). This mean is probably an underestimate of the actual chlorophyll concentration. When the mean was calculated, values below the level of detectability were assigned a numerical value of 1.0 μg/l.

Bachmann and Jones (1974) suggest a direct relationship between chlorophyll a concentration and total phosphorus concentration. Using their model, a mean chlorophyll a concentration of 4.6 μg/l should have a total phosphorus value of 0.015 mg/l. Pond D contained a mean total phosphorus concentration of 0.02 mg/l and is in agreement with their model.

Periphyton

The periphyton community of Pond D was initially colonized very slowly (Figure 6C). There was a 10-day time lag between the placement of the substrates and the first detectable level of chlorophyll a (> 1 $\mu g/cm^2$). Kevern (1962) found a 3-day period after substrate placement when little growth occurred. He subtracted the first 3 days from total exposure time for calculating production rates. Lag time was not subtracted in this study, so these results would be comparable with the other ponds.

The maximum standing crop of periphyton observed was 1.8 $\mu g/cm^2$ and occurred during the first exposure period. Maximum colonization rate was 0.08 $\mu g/cm^2$/day. It appears as if the turnover time was greater than the 28-day exposure period.

GENERAL RESULTS

Dissolved Oxygen

A number of factors affect the concentration of dissolved oxygen in the water, two of which are temperature and the biota. Temperature affects the solubility of dissolved oxygen and the metabolic rate of organisms. Temperature can affect the seasonal and daily concentrations of dissolved oxygen, but it has a more pronounced effect on the seasonal variation of oxygen than on daily fluctuations.

Daily fluctuations in dissolved oxygen occur and they are in part related to the photosynthesis and respiration of the aquatic community (Odum, 1956). Oxygen is released into the water by primary producers as a by-product of photosynthesis during daylight. During darkness, the primary producers as well as the remainder of the aquatic community utilize dissolved oxygen for respiration. In addition, there is some exchange of oxygen with the atmosphere. The direction of exchange depends on the saturation gradient.

Diurnal fluctuations in dissolved oxygen are an indication of the amount of photosynthetic activity (primary production) being carried out by the aquatic community (Verduin, 1956; Ryther, 1956). Diurnal fluctuations were not monitored during this study. However, percent saturation of dissolved oxygen was calculated.

Percent saturation is the amount of oxygen which is dissolved in a volume of water compared to the maximum which could be dissolved in the same volume of water at the same temperature and pressure.

Supersaturation is a situation where greater than the maximum amount of a substance has been dissolved at standard temperature and pressure. With dissolved oxygen, this is an unstable condition and the oxygen tends to escape from solution until a new equilibrium is established near 100 percent saturation. In an aquatic system where plants and algae produce oxygen through photosynthesis, the amount of dissolved oxygen added to the water can often exceed the expected capacity of the water and it becomes supersaturated with oxygen (Pecor et al., 1973).

Oxygen supersaturation is often attributable to photosynthesis by primary producers. Consequently, marked oxygen supersaturation is commonly associated with highly productive waters (Hutchinson, 1957).

At the beginning of this project it was assumed that primary production in the four ponds would be similar. With the exception of nutrient loading, all ponds were exposed to the same chemical, physical and environmental variables. Therefore, water samples taken at the same time from all ponds and analyzed for dissolved oxygen and percent saturation should, in gross terms, reflect the relative production of oxygen by aquatic macrophytes and algae.

The results of this comparison are presented in Table 15. Pond A, which received the greatest amount of enrichment and hence was assumed to have the highest productivity, had a mean dissolved oxygen saturation value of 107 percent.

Table 15. Percent Dissolved Oxygen Saturation in Michigan State University, Lake City Experiment Ponds, 1970.

Date	Pond A	Pond B	Pond C	Pond D
6-29	97	108	96	96
7-1	100	107	97	98
7-4	83	87	84	84
7-7	96	95	100	99
7-10	76	104	106	96
7-13	182	120	133	130
7-16	185	128	122	110
7-19	186	142	88	98
7-23	k52	136	99	90
7-25	127	122	100	95
7-28	122	155	118	99
7-31	136	95	99	95
8-3	92	86	92	86
8-6	94	93	96	92
8-10	57	78	95	98
8-14	61	73	110	104
8-18	58	65	97	92
8-22	52	64	78	87
8-27	79	87	83	91
	$\bar{x}$ = 107	$\bar{x}$ = 102	$\bar{x}$ = 99	$\bar{x}$ = 96
	Range 57-186	Range 64-155	Range 83-133	Range 86-130
	S.D. = 42.0	S.D. = 25.0	S.D. = 13.0	S.D. = 9.0

A decrease in mean percent saturation occurred with a decrease in nutrient loading. Pond B was greater than Pond C which was greater than Pond D. The mean percent saturation of Ponds B, C and D was 102, 99 and 96, respectively.

The range and standard deviation of the four ponds indicate that the stability of the pond decreased with increased nutrient loading. Pond A had a larger range and standard deviation than Pond B which was larger than C which was larger than D. The large standard deviation was caused by the response of the aquatic community to reach a new equilibrium with changing levels of nutrient availability.

Phosphorus

The mean and range of total and soluble phosphorus concentrations in Ponds A, B, C and D are given in Table 16. Total phosphorus concentrations are graphically presented in Figure 7. Ponds B and C followed patterns similar to those described in Pond A, i.e., total phosphorus being always lower than the theoretical value and soluble phosphorus being some fraction of total phosphorus. The mean observed total phosphorus values were 80.3 (Pond A), 72.3 (Pond B) and 89 (Pond C) percent of the theoretical values (Tables 5, 8 and 11).

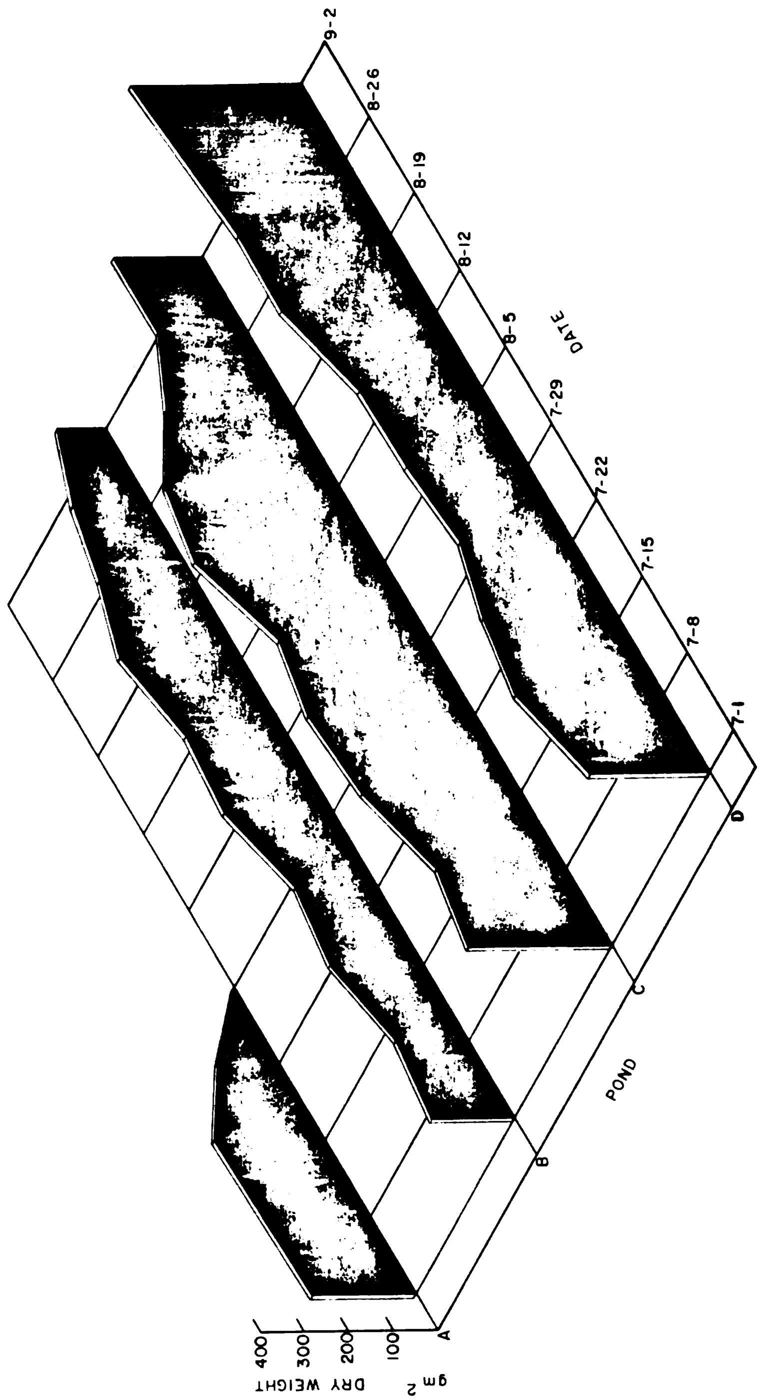
Total phosphorus levels in Pond D were within the range of values commonly found in Michigan inland lakes (Michigan Department of Natural Resources, 1975 unpublished data). Therefore, this concentration encountered could be considered a natural level. Soluble phosphorus concentrations were always below the level of detectability (< 0.01 mg/l).

Table 16. Mean and Range of Total and Soluble Phosphorus Concentrations[a] in Michigan State University Experimental Ponds, 1970.

		Pond A	Pond B	Pond C	Pond D
Total Phosphorus	Range	< 0.01-2.15	0.01-0.72	0.01-0.57	0.01-0.04
	$\overline{X}$	0.85	0.29	0.18	0.02
Soluble Phosphorus	Range	< 0.01-1.49	< 0.01-0.65	< 0.01-0.51	< 0.01
	$\overline{X}$	0.75	0.21	0.13	< 0.01

[a]Expressed in mg/1.

Figure 7. Total phosphorus concentrations observed in experimental ponds in Lake City, Michigan, 1970.


DRY WEIGHT
gm 2
400
300
200
100
A
B
C
D
POND
DATE
7-1
7-8
7-15
7-22
7-29
8-5
8-12
8-19
8-26
9-2

Some explanation of the phosphorus cycle may explain why concentrations of phosphorus in the enriched ponds were lower than theoretical values.

The decline of phosphorus concentrations after fertilization has been noted by many scientists (Zeller, 1952; Hepher, 1958).

Under normal conditions phosphate is found in aquatic systems due to natural and man-made activities. Distribution may depend to a great degree on basin form, chemical composition of the surrounding terrain, land use, waste disposal practices, watershed size, behavior of other substrates in the particular body of water (Reid, 1961).

Phosphate, pyrophosphate, tripolyphosphate and polyphosphate anions are known to form complexes, chelates and insoluble salts with a number of metal ions. How much complexing and chelating goes on will depend on the concentrations of phosphates and metal ions, the pH and the presence of other factors in the water (Stumm and Morgan, 1970).

Three forms of phosphates are commonly found in natural waters: soluble inorganic, soluble organic, and organic. Many researchers studying aquatic systems employ the molybdenum blue method of measuring orthophosphate. Rigler (1968) questions whether this method really measures orthophosphate. He postulates that the concentration of inorganic phosphate in lakewater is much lower than indicated by this method. Hydrolysis of organic compounds may be the source of error. The measurement of total and soluble phosphorus in the Lake City study would eliminate this error.

Watt and Hayes (1968) concluded analytical techniques can measure only net changes in phosphorus and cannot provide information about an equilibrated cycle. A steady state may exist such that during the period between two sets of observations, no change can be found in the phases of phosphorus. This does not mean that exchanges have not taken place, but rather that all changes are compensated--the system is in a state of dynamic equilibrium.

The following chemical explanations are given for the lower than theoretical concentrations of phosphate in the water column.

Large quantities of phosphorus are consistently found deposited in lake muds, but often the quantities of soluble inorganic phosphorus found in water are very small (Zicker et al., 1956).

Insoluble ferric phosphate is precipitated when ferric iron and inorganic phosphate occur together under alkaline aerobic conditions. When oxygen is added to the system, any excess iron is precipitated out as ferric hydroxide. When oxygen is lacking in the bottom muds, iron is reduced from ferric to the ferrous state and phosphorus is free to go into solution (Hutchinson, 1957).

Mortimer (1941) is in basic agreement with these findings. He concluded that most lake muds just below the surface are anaerobic and contain ferrous iron which is soluble. Ferrous iron cannot exist in oxygenated water, but is precipitated in ferric form on the mud surface carrying phosphorus with it. In a later paper, Mortimer (1942) elaborates on the mud-water relations. The thickness of the surface-oxidized layer decreases, adjusting to the new balance of supply and demand. The decrease in thickness takes place from below and results in the liberation of some previously adsorbed or precipitated ions (Fe, Mn, $PO_4^{\equiv}$).

Hutchinson (1957) agrees, stating that in productive lakes with clinograde oxygen curves there is an increase in soluble phosphate in the oxygen deficient part of the hypolimnion. This is in part due to decomposition of sinking plankton, but in most cases is primarily caused by liberation of phosphate from sediments under reducing conditions.

Excess ferric hydroxide forms a gel which while precipitating out of solution, also adsorbs phosphates on its surfaces (Zicker et al., 1956). They also concluded that calcium carbonate had a depressing effect on the concentration of soluble phosphate. By adding calcium carbonate to a lake containing 8 ppb soluble phosphorus, the water concentration was reduced to 0.5 ppb. This reduction may have been accomplished in two ways: (1) formation of a new compound such as hydroxylapatite, and (2) phosphate ions being adsorbed on the surface of the $CaCO_3$ crystals. The crystals that settle to the mud surface act in two ways: (1) form a seal that restricts the diffusion of phosphate ions released by the mud, and (2) some of the calcium particles entering the mud can convert some of the iron and aluminum held phosphates to calcium phosphates. If the pH is lowered, as may happen during anoxic conditions, some of the phosphorus is released.

The effect of high calcium concentrations on phosphorus concentrations was also investigated by Otsuki and Wetzel (1972). They considered chemical precipitation, without the agency of living organisms, responsible for several marl lakes in Indiana. By evaporation of natural waters, $CaCO_3$ is deposited and takes soluble salts with it. Even a change in temperature or free CO_2 concentration can cause precipitation.

Hepher (1958) felt that in hardwater ponds phosphorus added to the water in soluble forms disappears rapidly from the water mainly by being precipitated as calcium phosphate. He felt the process is influenced by pH and alkalinity of the water. The high pH values and moderately hard water in the ponds would account for some of the phosphorus loss.

Hooper and Ball (1964), while fertilizing marl lakes to increase primary production, concluded that a decrease in suspended carbonate and phosphorus coincided with an increase in transparency; it seemed as if phosphorus settled along with considerable marl.

The addition of living organisms to the cycle serves to clarify what may be happening. Fertilization of eutrophic lakes will very often cause increased primary production. At times, the increase is in phytoplankton (Swingle _et al_., 1963) or periphyton population (Hooper and Ball, 1964). At other times, it is displayed in the macrophytes (Mulligan and Baranowski, 1969). This may be due to the fact that macrophytes seem unable to utilize organic phosphate compounds (Hayes and Phillips, 1958). When Hayes _et al_. (1952) added large amounts of inorganic phosphate to a lake, it was found that the rooted aquatic _Ericaulon_ sp. had taken up phosphorus in hours. Examination revealed that the leaves showed uptake before the roots. It appears that material does not have to go through the roots in order to reach the leaves. The concentration in the plant tissue can be correlated with the concentration in the water (Lee, 1969). Hutchinson and Bowen (1950) reported that within one week, some species of Potamogenton in fertilized water can concentrate phosphorus in their tissue 1000 times that found in the tissue of similar plants in unfertilized water.

Hutchinson and Bowen (1950) relate that when inorganic phosphate is added to a lake, almost immediately all of the phosphorus is taken up by the phytoplankton which can then easily be sedimented out of the epilimnion. Hayes and Phillips (1958) found that Chlamydomonas dysosmos established a new phosphorus equilibrium within 2.5 minutes after the addition of phosphorus. Under natural conditions, such microorganisms can very rapidly make over any inorganic phosphorus present into an organic form. The microorganisms then return a fraction of the phosphorus via cell leakage and metabolic by-products to the water where it can be used by other organisms. In all cases, there was a rapid loss of phosphorus from the open water of the lake, the loss being due not only to storage by phytoplankton, but also the zooplankton and littoral vegetation.

Mackereth (1952) thought the uptake of phosphorus may be a purely physical phenomenon by adsorption on some part of the algae cell.

Chu (1946) tested to see if phytoplankton could use various forms of phosphorus. The following forms were tested: orthophosphate, phytin (a stable organic phosphorus compound of plant origin) and pyrophosphate. The results were: (1) orthophospate--plankton grew well; (2) phytin--plankton grew well; (3) pyrophosphate--there was some conversion to orthophosphate, but not enough to sustain growth. Therefore, the change of dissolved organic phosphorus may follow two courses: (1) turning into orthophosphate and (2) being absorbed and utilized directly by living organisms.

This is in keeping with the findings of Karl-Kroupa et al. (1957). The findings indicate that polyphosphates, which are stable compounds, are easily converted to orthophosphates by living organisms (plants and bacteria).

Hayes and Phillips (1958) do not believe that any specific natural physiochemical or bacteriological layering of the muds in lakes is important. The dominant role in exchange was previously attributed to the state of oxidation or reduction with bacteria only playing a minor role but their results indicate that bacteria can to a high degree suppress the classical inorganic mechanism.

Macpherson et al. (1958) found that when inorganic phosphate is added to water, a large part is likely to be quickly converted to an organic form by bacteria. If bacteria are absent, plants rapidly absorb it. Therefore, under conditions in nature, only a minor fraction is likely to be left for the mud adsorption reaction.

Hayes et al. (1958) concluded that bacteria appear to be the principal dynamic agents that affect the redox conditions of the mud. The lower strata of muds are at all times reduced but the layer at the interface may be oxidized by the water above, but this oxidized layer is never more than 1 mm thick. Pomeroy et al. (1965) present a conflicting view of the exchange mechanism. They feel exchange consists of a two-step ion exchange between clay minerals and water, plus a minor exchange between interstitial microorganisms and water.

Hayes and Phillips (1958) found that more total phosphorus remains in water at equilibrium when bacteria were present than when they were absent. The ability of bacteria to keep phosphorus in water has been explained in two ways: (1) by an acceleration of the rate of phosphorus return from the sediments to the water by bacteria in the mud decomposing dead or decaying organic matter; (2) by rapid uptake of the phosphorus, it is held from chemical or colloidal adsorption mechanisms of mud and water.

Kuznetsov (1968) provides some interesting material on bacteria. The mineralization of iron humates and the oxidation of ferrous forms of iron with subsequent sedimentation are, to a considerable extent, the result of certain species of iron bacteria. There are also bacteria capable of reducing ferric to ferrous compounds; these can filter through the silt surface and form an iron nodule. This indicates that these bacteria can affect the iron concentration directly and the phosphorus concentration indirectly.

Hargrave and Geen (1968) found that the regeneration of inorganic phosphorus by zooplankton is also of great importance. The daily phosphate requirements of phytoplankton were compared with phosphorus released by zooplankton. The rates of excretion and the abundance of zooplankton were used to calculate an average daily addition of inorganic phosphorus to the photic zone. In one lake the zooplankters released twice the average daily phytoplankton phosphorus requirement, but in a pond only one-fifth was supplied by animal excretion.

As Wetzel (1960) points out, primary producers affect marl deposition in lakes. Autotrophic and heterotrophic bacteria are capable of withdrawing CO_2 and causing a chemical precipitate. Chara, some filamentous algae and all macrophytes can also perform this task. This precipitation can then remove inorganic phosphate which is necessary for their growth. This is partially counter-balanced by another plant mechanism. Dissolved organic material excreted by the primary producers can act as a chelator for metal ions (Wetzel, 1968). Chelation, an equilibrium reaction between a metal ion and an organic chelating agent, results in the formation of a stable ring structure incorporating the metal ion. It assumes importance in marl lakes that exhibit many chemical characteristics unfavorable to high sustained productivity. Many essential ions such as iron and phosphorus are usually precipitated out of the aerobic zone and become unavailable for plant use. Chelation by dissolved organic matter can maintain metallic ions complexed in a soluble state and are available for metabolism. The organic chelate can successfully compete for cations such as Ca^{++} from $Ca_3(PO_4)_2$ and release the phosphate for use.

The biologic cycle as outlined by Rigler (1956) is the uptake of inorganic phosphate by plankton and bacteria which settle to the bottom. They are decomposed and their nutrients released. There then appears to be a horizontal movement of the nutrients to the littoral area via bacteria and plankton. Once in the littoral areas the macrophytes utilize them. The macrophytes in turn release phosphorus to the open water to complete the cycle. Hayes and Phillips (1958) conclude there is five times as much participating inorganic phosphorus in the mud phase as in the water (obtained from the sedimentation of phytoplankton and bacteria). The ratio of soluble organic to inorganic phosphorus in water is also 5 to 1. Hayes and Phillips (1958) combine the ratios and it is seen that the phosphorus exchange in the mud is about equal to the total phosphorus in the water. If the mud phosphorus were released, it would double the supply (this would then trigger more equilibrium reactions with underlying mud).

Watt and Hayes (1968) state that when an aquatic biological system approaches a relatively stable condition, the distribution of dissolved inorganic phosphorus, particulate phosphorus, and dissolved organic phosphorus may be dealt with as though it were the result of the equilibrium of a chemical exchange system. This statement is in line with Rigler's (1964) observations that in eight of the nine lakes studied, the proportions of inorganic, soluble organic and organic phosphorus were almost identical.

There is a strong interaction between the physical environment and its biological components.

In this study it appeared only a small portion of the added nutrients would contribute to the continued development and sustenance of the primary producers. A major quantity of the phosphorus and nitrogen added could not be accounted for as having left the pond via the outflow. Most phosphorus remained in the pond presumably sedimented in the bottom and unavailable to plants and algae. However, certain rooted aquatic plants can obtain phosphorus from the sediments via their root systems. Certain conditions prevailed throughout the study and contributed to the return of the ponds to prefertilization nutrient levels. The shallow study ponds remained in an oxidized state, so that the added phosphorus, which did not leave via the outfall, could be held out of the biologic cycle as poorly soluble ferric phosphate.

Primary Production

An examination and comparison of the total primary production in each pond would indicate whether fertilizer had increased the amount of plant material in each pond or whether it had just favored one component of primary producers over another. Estimated ash free dry weight of each component of primary producer was compared (Table 17). Pond C, which received the smallest of the three artificial nutrient loads, had the highest mean standing crop (ash free dry weight). Using this criterion, the second most productive pond was D which was unfertilized and acted as a control. In all cases, the bulk of the standing crop came from the macrophytes. The macrophytes appear to be the most important component in the production of organic material in this pond situation.

However, care must be taken when using mean standing crop to estimate production. Macrophytes, while difficult to measure, are relatively stable throughout the growing season. Phytoplankton and periphyton are less stable and pulses in populations may appear and

Table 17. Estimate of Mean Standing Crop[a] of Primary Producers in Lake City, Michigan Ponds, 1970.

COMPONENT	Pond A	Pond B	Pond C	Pond D
<u>Macrophytes</u>				
g/m^2 day wt.	85.3±108.5	198.4±40.5	319.2±70.4	317.6±39.9
g/m^2 ash free dry wt.[b]	16.6	38.7	62.2	61.9
<u>Phytoplankton</u>				
ug/l chlorophyll <u>a</u>	31.2±29.8	15.9±10.6	6.8±4.3	4.2±3.1
g/m^2 ash free dry wt.[b]	2.40	1.22	0.52	0.32
<u>Periphyton</u>				
ug/cm^2 chlorophyll <u>a</u>	7.5±2.9	7.6±4.68	5.0±4.3	6.87±0.56
g/m^2 ash free dry wt.[b]	5.77	5.85	3.85	0.67
Total	24.8	45.8	66.6	62.9

[a]± one standard deviation.

[b]Estimated ash free dry weight.

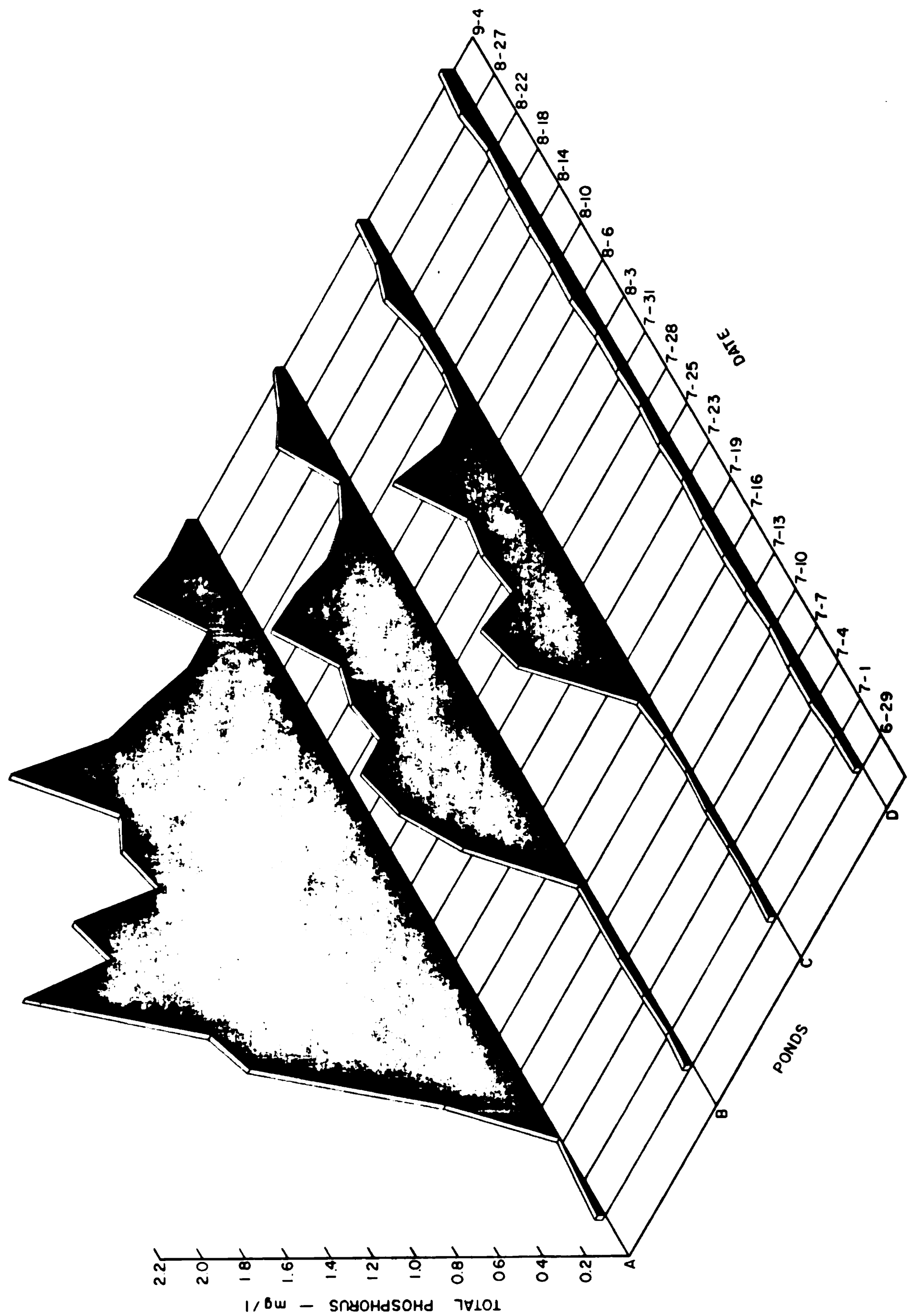
disappear between sampling dates. Phytoplankton standing crop may appear stable between sampling dates, yet this may be an entirely new population. Production of phytoplankton can be high with little change in standing crop. Periphytic growth in the form of filamentous algae which was removed during a bloom from Ponds B and C was not added to the estimates. This would add to the total standing crop for Ponds B and C, but insufficient comparable data were collected for quantification.

The physical removal of the floating filamentous algae from Ponds B and C during the bloom (August 5) may have had a substantial yet undetermined effect on the ponds. If this material had been allowed to remain in the ponds, it may have shaded out the macrophytes and algae, caused the ponds to go anaerobic, or the filamentous algae may have had an antagonistic effect on the plants and phytoplankton. The mean standing crop of primary producers as discussed in the previous section, as well as the individual results in Ponds B and C, may have seriously been affected by this manipulation.

The response of the primary producers in each pond has already been discussed. The results of each fraction (i.e., macrophytes, phytoplankton and periphyton) in the four ponds will now be compared.

The response of the macrophyte community in the four ponds varied with the nutrient loading (Figure 8). Pond A which received the highest nutrient loading had the lowest standing crop of macrophytes at the end of the sampling season. Smith (1969), while

Figure 8. Standing crop of macrophytes (biomass) observed in Lake City, Michigan Experimental Ponds, 1970.


TOTAL PHOSPHORUS — mg/l
2.2
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
A
B
C
D
PONDS
DATE
6-29
7-1
7-4
7-7
7-10
7-13
7-16
7-19
7-23
7-25
7-28
7-31
8-3
8-6
8-10
8-14
8-18
8-22
8-27
9-4

studying changes in the biota of a lake after fertilization, reported that healthy macrophytes increased in abundance after years when algae blooms had occurred as a result of fertilization. However, during the fertilization and algal blooms, macrophytes died or appeared moribund.

Ponds B and C which received lower nutrient loading had higher standing crops of macrophytes. Pond D which was not enriched contained the largest standing crop and highest production rate of any study pond. Phytoplankton populations in the four ponds did not follow a similar pattern.

Phytoplankton standing crop, as determined by chlorophyll a concentrations, appeared to be directly related to the rate of nutrient loading. Standing crop in Pond A was greater than Pond B > Pond C > Pond D (Figure 9).

These findings are in agreement with those of Mulligan and Baranowski (1969). While conducting bioassays on the effects of various phosphate and nitrate concentrations on primary production, they reported the amount of chlorophyll a in the water was highest in heavily fertilized treatments, lowest in control treatments and slightly higher in low fertilizer treatments. They also describe a pattern for periphyton development which was observed at the Lake City study.

The periphyton community of the ponds responded quickly to enrichment. Highest average rate of accrual occurred in Pond B, which received an intermediate rate of enrichment (Figure 10). Pond A had the next highest mean accrual rate. Much of the

Figure 9. Standing crop of phytoplankton (biomass) observed in Lake City, Michigan Experimental Ponds, 1970.

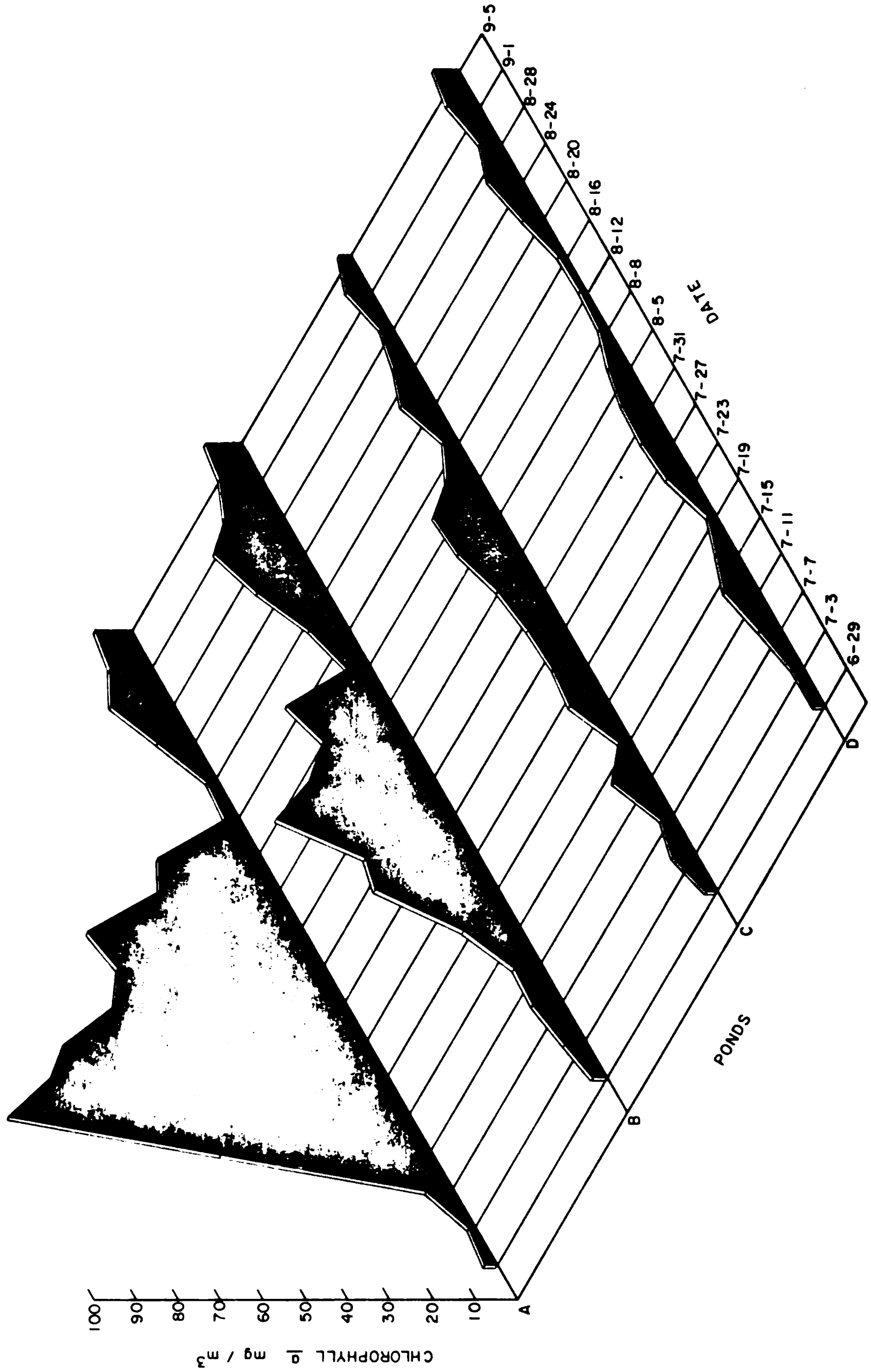
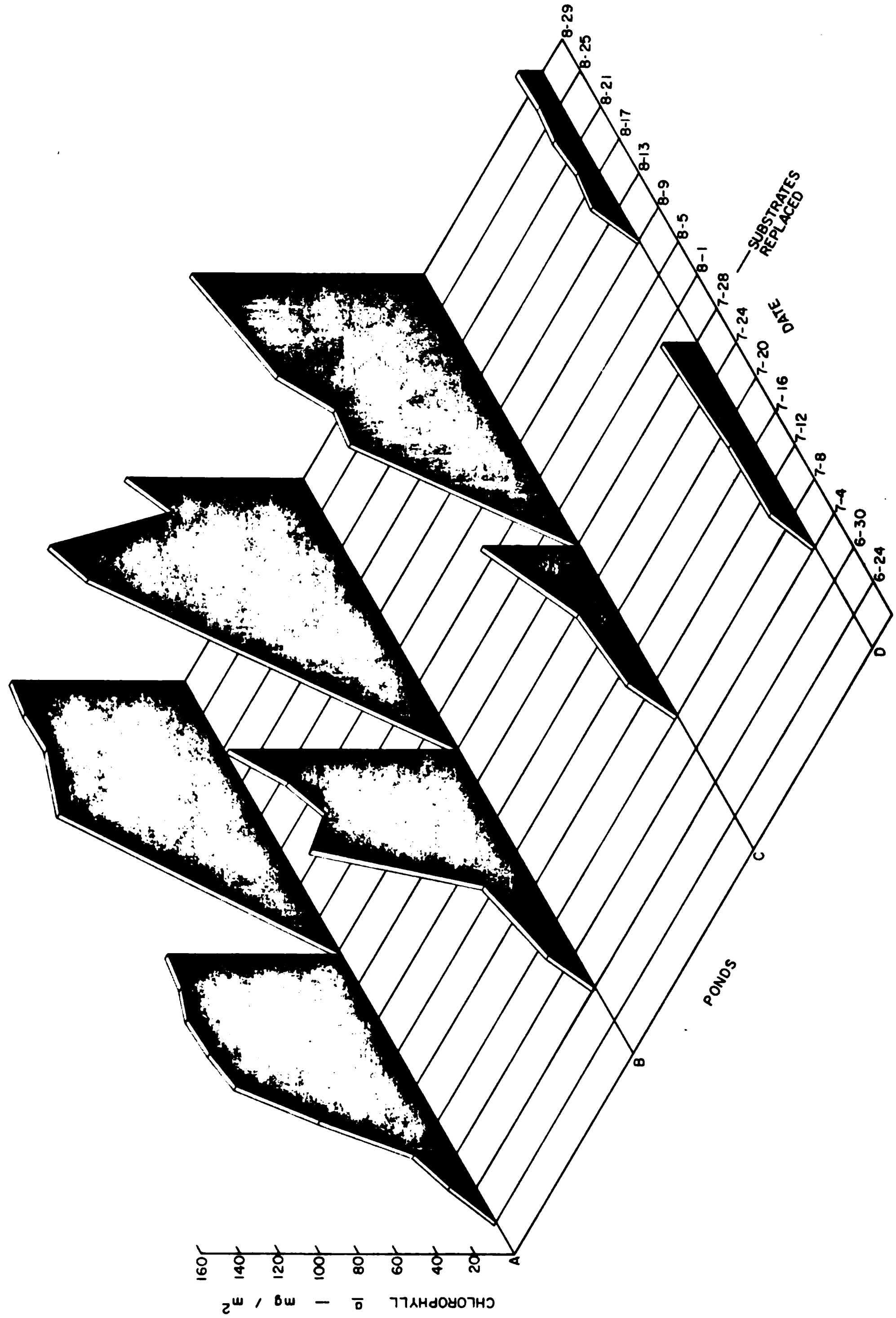

CHLOROPHYLL a mg / m³
100
90
80
70
60
50
40
30
20
10
A
B
C
D
PONDS
DATE
6-29
7-3
7-7
7-11
7-15
7-19
7-23
7-27
7-31
8-5
8-8
8-12
8-16
8-20
8-24
8-28
9-1
9-5

Figure 10. Standing crop of periphyton observed in Lake City, Michigan Experimental Ponds, 1970.



CHLOROPHYLL a — mg / m²
160
140
120
100
80
60
40
20
A
B
C
D
PONDS
DATE
6-24
6-30
7-4
7-8
7-12
7-16
7-20
7-24
7-28
8-1
8-5
8-9
8-13
8-17
8-21
8-25
8-29
SUBSTRATES REPLACED

periphyton accrual occurred after the phytoplankton standing crop had begun to decline. Smith (1969) described similar results. He reported that immediately after fertilization there was a pulse of phytoplankton. However, after a time blooms of filamentous algae appeared.

In conclusion, the results of this study found the highest loading of phosphorus and nitrogen used favored the phytoplankton community to the detriment of the macrophyte community. Filamentous algae appears to have been most abundant at the intermediate loading rates. The natural macrophyte community of the ponds produced the greatest biomass under "natural" or control conditions.

These findings are supported by Mulligan and Baranowski (1969) in their field and laboratory studies. Wetzel and Hough (1973) in an assessment of the role of macrophytes in lakes suggest a similar relationship. They suggest that as nutrient limitations are removed, phytoplankton and periphytic algae are capable of responding quickly and increasing primary production. However, certain types of macrophytes can respond at high nutrient levels.

McNabb (1975) presented a similar theory. He observed that the initial response to increasing nutrient levels was an increase in macrophyte biomass and changes in the macrophyte community. As nutrient levels increase, he observed an increase in the importance of the periphyton community. At the highest nutrient levels he suggests an increase in the dominance of the phytoplankton community.

SUMMARY

The primary production was measured on four farm ponds from June 26, 1970, through September 3, 1970, at the Michigan State University, Lake City, Michigan experimental field station. Three ponds were enriched with different concentrations of phosphorus and nitrogen. The fourth pond served as a control.

Chlorophyll a was used to estimate standing crop of phytoplankton and periphyton. In addition, it was used to determine rate of colonization of the periphyton community. Macrophyte standing crop was determined gravimetrically. Water chemistry was monitored for pH, alkalinity, soluble and total phosphate, nitrate and dissolved oxygen.

An investigation of the data showed that the phosphorus and nitrogen added to the ponds did not result in the theoretical concentration of these constituents. Due to chemical and biological conditions operating in the ponds, the nutrients did not remain in solution for any length of time.

Under the environmental conditions operating in the ponds during the study, it appeared as if high nutrient loading rates favored the production of phytoplankton and periphyton. However, the results of the project may have been an artifact caused by the manipulation of the primary producers. The physical removal of a portion of the periphyton community during the filamentous algae

bloom placed an undetermined influence on the other primary producers in the pond. If left in the pond, the macrophyte and phytoplankton populations may have responded in a manner other than the one observed.

Macrophyte production was highest in the control pond, and was adversely affected in the other ponds by the conditions imposed by the experiment.

Additional research is needed to determine the exact set of physical, chemical and biological variables operating at the time of this investigation. Insufficient data was collected to significantly quantify the effect of each of these parameters.

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