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IRON-ALGAE INTERACTIONS AS A FACTOR  
IN LAKE ERIE WATER QUALITY

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ABSTRACT

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Nutrient enrichment analyses revealed that both chelated and unchelated iron affect the photosynthetic rate of natural phytoplankton communities in eastern Lake Erie. Depending on the form, the concentration, and the time of year, iron additions may stimulate or inhibit algal photosynthesis. However, iron discharges from wastewater treatment plants using iron for phosphorus removal may not significantly affect algal growth in Lake Erie because of the high dilution effect.

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## INTRODUCTION

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Iron is necessary in algae growth for a variety of cellular processes, including photosynthesis, respiration, and nitrogen fixation (Burris 1965; Bidwell 1974). Several investigators have also shown that iron concentrations may occasionally limit the growth rate of algae in some freshwater environments (Schelske 1962; Sakamoto 1971; Murphy and Lean 1975).

Despite its important role in algal metabolism, iron is usually not considered to be a primary nutrient responsible for controlling eutrophication in aquatic environments. This is in part due to the fact that non-point sources do not generally discharge large concentrations of iron into aquatic environments, nor do domestic and industrial wastewaters usually contain significantly large concentrations. In the near future, however, as the number of wastewater treatment plants using iron to remove phosphorus increases, more iron will enter aquatic environments via their discharges. As a result, iron may cause changes in the phytoplankton communities of some of these aquatic ecosystems.

Precipitating phosphorus from wastewaters with iron has become a common method of treatment plants to reduce its input to natural waters (Leary et al 1974; Ockershausen 1975; Braasch and Joeres 1976). While iron effectively removes large quantities of phosphorus, some remains in the treated waters. The iron may so enter the aquatic environment receiving the treatment plant discharges. Preliminary analyses of a few treatment plants using iron

salts indicate that approximately 20 to 850  $\mu\text{g liter}^{-1}$  total iron might remain in the treated effluent (A. Bennice, K. Wuhrmann, and A. Bernhardt, personal communication). Because iron is important in algal metabolism, this addition might affect the growth of phytoplankton in the receiving aquatic environments.

This investigation studied the effects of iron additions on the growth of natural communities of Lake Erie phytoplankton. The purpose for studying these effects was to determine whether or not iron discharge from wastewater treatment plants would significantly affect the growth of phytoplankton in Lake Erie. Although similar investigations in Europe have suspended iron use at some wastewater treatment plants, the U.S. generally lacks such studies.

#### Materials and Methods

Using natural communities of Lake Erie phytoplankton as the bioassay organisms, we conducted seven iron enrichment analyses between September 1975 and August 1976. We sampled surface water for these analyses from the eastern basin of Lake Erie usually from a boat off Point Gratiot near Dunkirk, NY, or from the break-wall at Barcelona, NY. On two occasions, we obtained water samples from the intake pipe of the water filtration plant at Dunkirk.

For each enrichment analysis, we collected lake water samples in four, 20-liter polyethylene containers and transported them to the laboratory at Fredonia, NY. Within two hours of sampling, we transferred the water to a series of 4-liter glass culture bottles. Iron was then added to duplicate or triplicate samples either

singularly as  $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$  (unchelated iron) or in combination with ethylenediamine tetraacetic acid (EDTA). Iron concentrations of the treated samples ranged between 50 and 2000  $\mu\text{g liter}^{-1}$ . Following enrichment, we incubated the samples for nine to 17 days at lake water temperature ( $\pm 2^\circ\text{C}$ ), either in a growth chamber with fluorescent lights (approximately  $150 \mu \text{Einsteins m}^{-2} \text{sec}^{-1}$ ) or in a greenhouse under natural solar radiation conditions.

During the incubation period, we monitored the responses of the algae to the iron additions in terms of the effect of the iron on algal photosynthetic rates. To measure these rates, we removed 300-ml subsamples from each 4-liter culture bottle and placed them in 300-ml Biochemical Oxygen Demand (BOD) bottles. After adding  $\text{NaH}^{14}\text{CO}_3$  (10  $\mu\text{Ci}$ ) to each 300-ml subsample, the BOD bottles were incubated for four hours under the same light and temperature conditions to which the corresponding 4-liter samples were exposed. After four hours of incubation, we filtered the algae in the BOD bottles onto membrane filters and measured their radioactivity with a liquid scintillation counter. To convert the  $^{14}\text{C}$  radioactivity counts to absolute values of photosynthetic  $^{12}\text{C}$  fixation, we also monitored the alkalinity and pH of the sample in each 4-liter bottle (Saunders, Trama, and Bachmann 1962).

The figures presented in this paper plot the results of the enrichment analyses as relative photosynthetic rates versus time. That is, the rate of photosynthesis in an iron enriched sample was divided by the rate of photosynthesis in the control sample to determine a relative rate of photosynthesis. Using this method to present the data, a relative photosynthetic rate above 1.0 indicates a stimulation of algal growth and a relative rate below 1.0 indicates an inhibition.



Lake water phytoplankton and iron concentrations were monitored during the 12-month investigation. We measured total iron concentrations in filtered and unfiltered samples with either the bathophenanthroline technique (Strickland and Parsons 1968) or atomic absorption spectrophotometry. We identified and counted the phytoplankton by the inverted microscope technique, and determined phytoplankton chlorophyll a spectrophotometrically (Strickland and Parsons 1968).

## RESULTS

### Unchelated Iron

The most commonly observed effect of unchelated iron additions ( $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ ) was a general inhibition of algal photosynthesis within the first seven to ten days of each enrichment analysis. The photosynthetic inhibition occurred in the five analyses conducted between October 1975 and July 1976 (Figures 1,2,3,4, and 5). Generally, the degree of inhibition increased the greater the iron concentration between 50 and 1,000  $\mu\text{g liter}^{-1}$  (figures 1,2, and 3). Over the range of iron concentrations (50 to 2,000  $\mu\text{g liter}^{-1}$ ) used in the five analyses where iron did inhibit, algal photosynthetic rates decreased approximately 10 to 70 percent relative to the controls.

In these analyses where iron was generally inhibiting, the extended period of inhibition was preceded by a period of stimulation that occurred within 24 to 48 hours after iron was first added to the samples (Figures 1,2,3,4 and 5). Also, the degree of inhibition in some analyses decreased with incubation time to the extent that algal photosynthetic rates in samples with 50  $\mu\text{g liter}^{-1}$  added iron were equivalent to or greater than those in the control samples after 10 to 16 days (Figures 1,4, and 5).

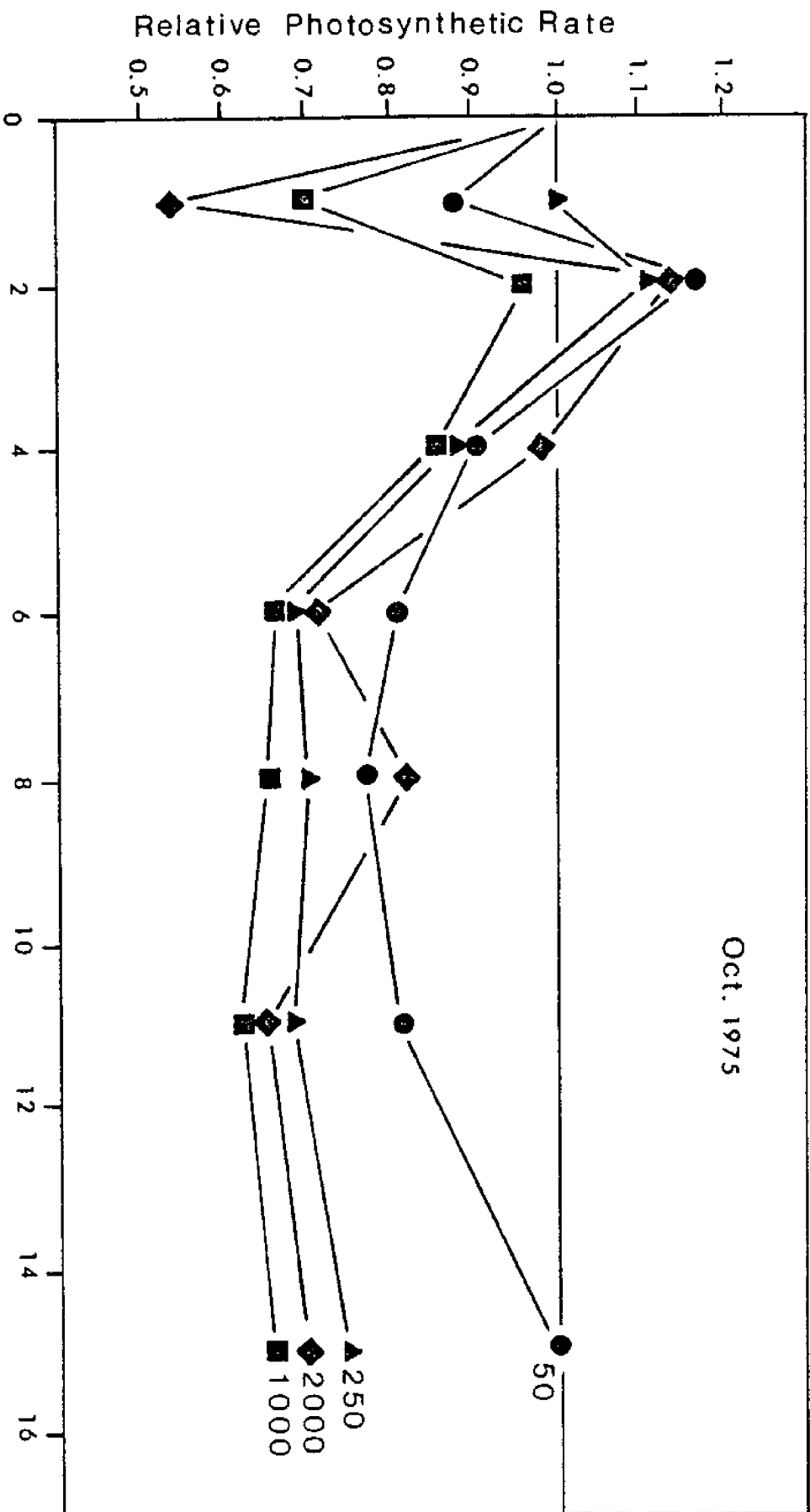


FIGURE 1. Photosynthetic rates of natural phytoplankton communities in samples spiked with iron relative to control: 50 µg/l (●), 250 µg/l (▲), 1000 µg/l (■), 2000 µg/l (◆), and 2000 µg/l (◆). 22 October to 7 November 1975. Lake Erie water sampled off Point Gratiot, New York.

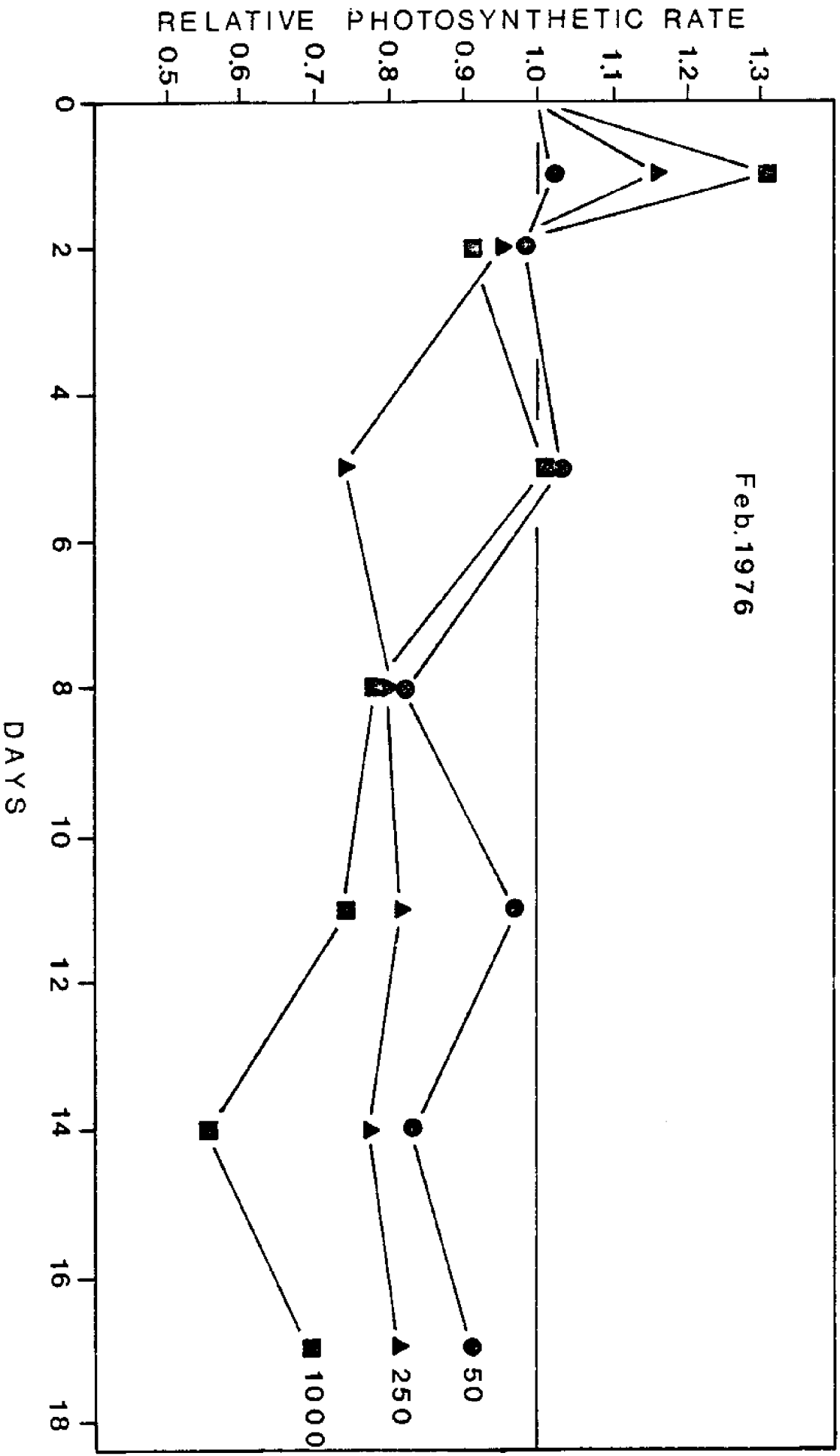


FIGURE 2. Photosynthetic rates of natural phytoplankton communities in samples spiked with iron relative to control: 50 µg/l (●—●), 250 µg/l (▲—▲) and 1000 µg/l (■—■). 4 to 21 February 1976. Lake Erie water sampled at Dunkirk, New York Water Filtration plant.

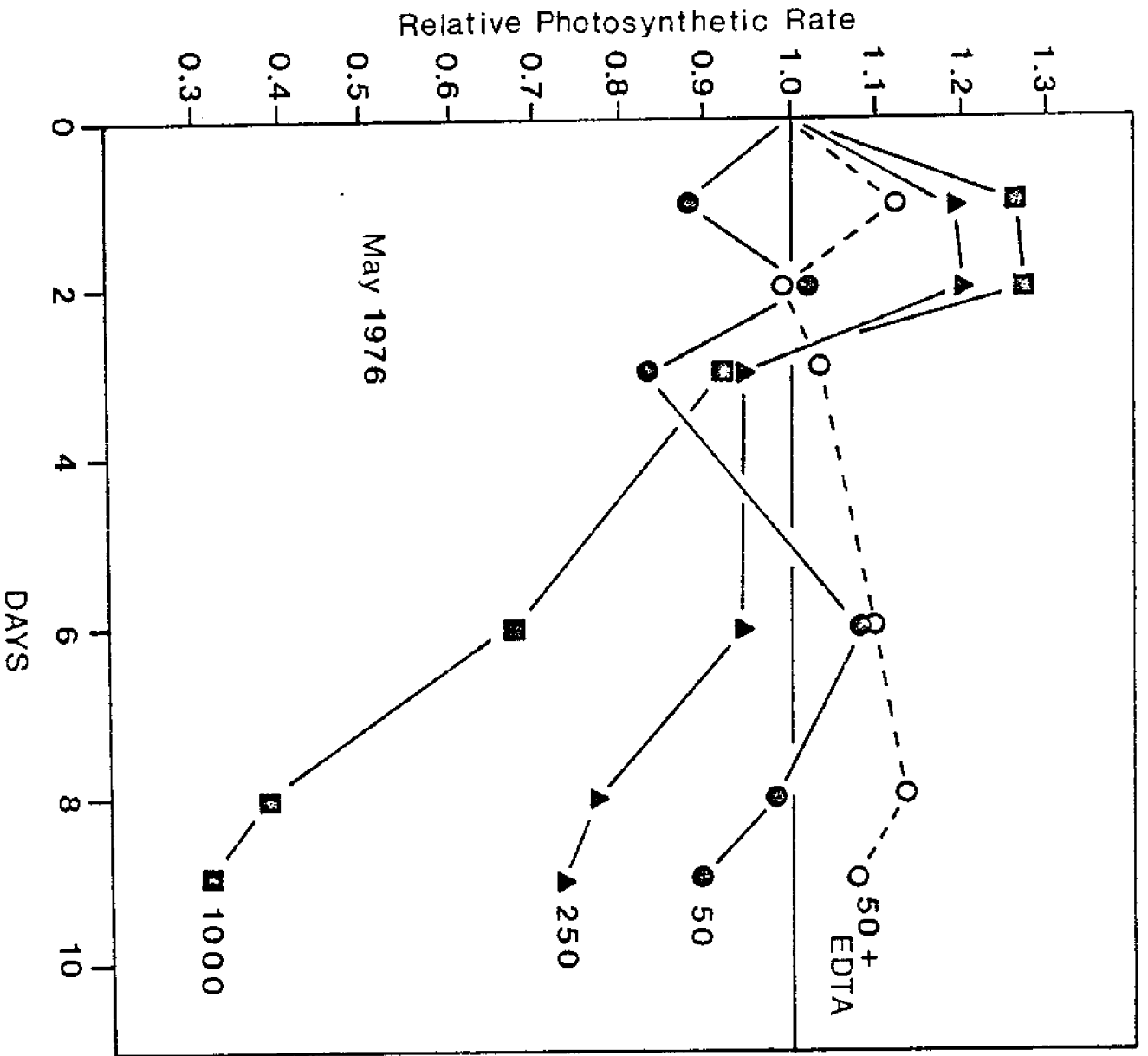


FIGURE 3. Photosynthetic rates of natural phytoplankton communities in samples spiked with iron relative to control: 50 µg/l (●—●), 250 µg/l (▲—▲), 1000 µg/l (■—■), and 50 µg/l EDTA (0--0). 25 May to 4 June 1976. Lake Erie water sampled off Barcelona, New York.

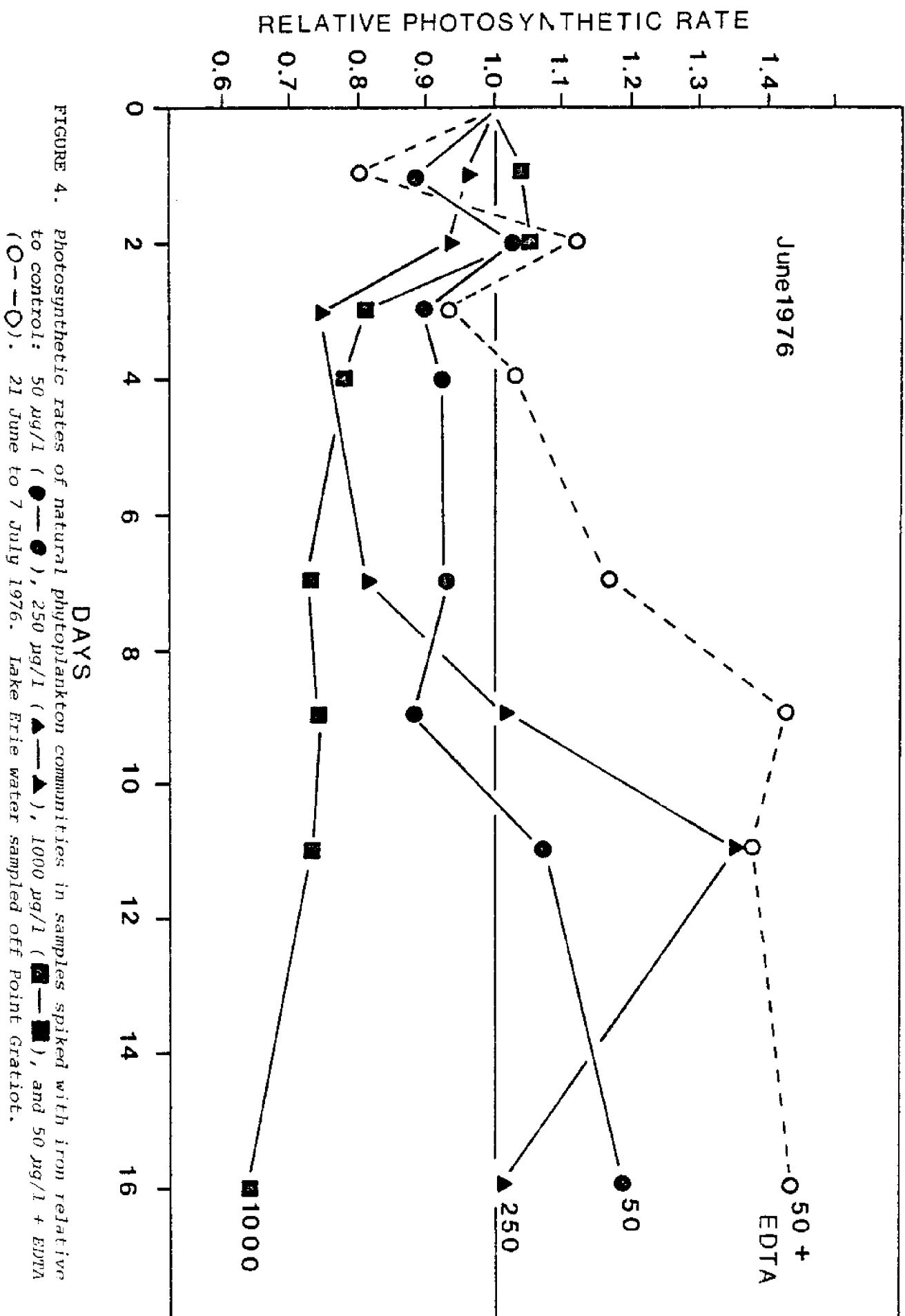


FIGURE 4. Photosynthetic rates of natural phytoplankton communities in samples spiked with iron relative to control: 50 µg/l (●-●), 250 µg/l (▲-▲), 1000 µg/l (■-■), and 50 µg/l + EDTA (○-○). 21 June to 7 July 1976. Lake Erie water sampled off Point Gratiot.

Unchelated iron in only two analyses had a significant stimulating effect on the photosynthetic rate of Lake Erie phytoplankton communities over the initial seven to fourteen days of an enrichment analysis. This stimulation occurred in late summer (August 1976) and early fall (September 1975) when  $50 \mu\text{g liter}^{-1}$  iron were added (Figures 6 and 7). The maximum degree of stimulation was low and ranged only between 10 and 18 percent relative to the controls. In both analyses, after an initial seven to fourteen days of stimulation the relative photosynthetic rates of the algae tended to decrease to the extent that rates in the iron-spiked samples were equal to or lower than those in the controls.

Figure 8 presents a summary of the results of the enrichment analyses with unchelated iron ( $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ ). It also includes data on phytoplankton chlorophyll a and the algae dominating the phytoplankton community over the 12-month period of investigation. Unchelated iron inhibited algal photosynthesis except in August and September. During the short stimulation period for these samples, algal chlorophyll concentrations were high, and blue-green algae were either dominant or codominant with green algae. This suggests that a relationship between the stimulatory effect of iron and the high densities of blue-green algae might have existed during these months.

#### Chelated Iron

The four enrichment analyses conducted between May and August 1976 employed chelated iron ( $\text{EDTA} + \text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ ). In all of these analyses,  $50 \mu\text{g liter}^{-1}$  of chelated iron stimulated algal growth (Figures 3, 4, 5 and 6). Maximum stimulation occurred in late June when algal growth in the presence of chelated iron was

1.4 times higher than the controls. Generally, relative algal growth rates increased only about 10% when  $50 \mu\text{g liter}^{-1}$  of chelated iron were added. In the one analysis with  $1,000 \mu\text{g liter}^{-1}$  chelated iron, iron inhibited algal growth (Figure 5). This suggests that the response of algae to chelated iron is a function of both the presence and concentration of iron in the chelated state. Thus, while low concentrations ( $50 \mu\text{g liter}^{-1}$ ) of chelated iron stimulated algal growth, higher concentrations ( $1,000 \mu\text{g liter}^{-1}$ ) inhibited the growth of Lake Erie phytoplankton.

#### Sampling Site Comparison

When this investigation began in September 1975, we made plans to run iron enrichment studies during the period of ice cover. Since we could not obtain samples by boat during the winter, water samples were to be taken from the intake pipe of the Dunkirk water filtration plant. However, subsequent comparisons of water taken directly from Lake Erie and from the water filtration plant indicated significantly higher iron concentrations in samples taken from the water filtration plant (Table 1). We suspect that the water accumulated additional iron as it passed through a cast iron pipe from the lake to the filtration plant.

Because of the high iron concentrations, the results of the enrichment analyses using the filtration plant water might not always represent effects of iron additions on samples taken directly from the lake. We confirmed this statement with a comparative analysis run in September 1975 (Figure 7). As a consequence, monitoring of the responses of winter and early spring phytoplankton communities to iron additions was limited to one analysis in February 1976

(Figure 2). While this one analysis indicated that iron inhibits algal growth during periods of ice cover, the results must be interpreted in light of the fact that the water was obtained from the intake pipe of the Dunkirk filtration plant.



Relative  
Photosynthetic Rate

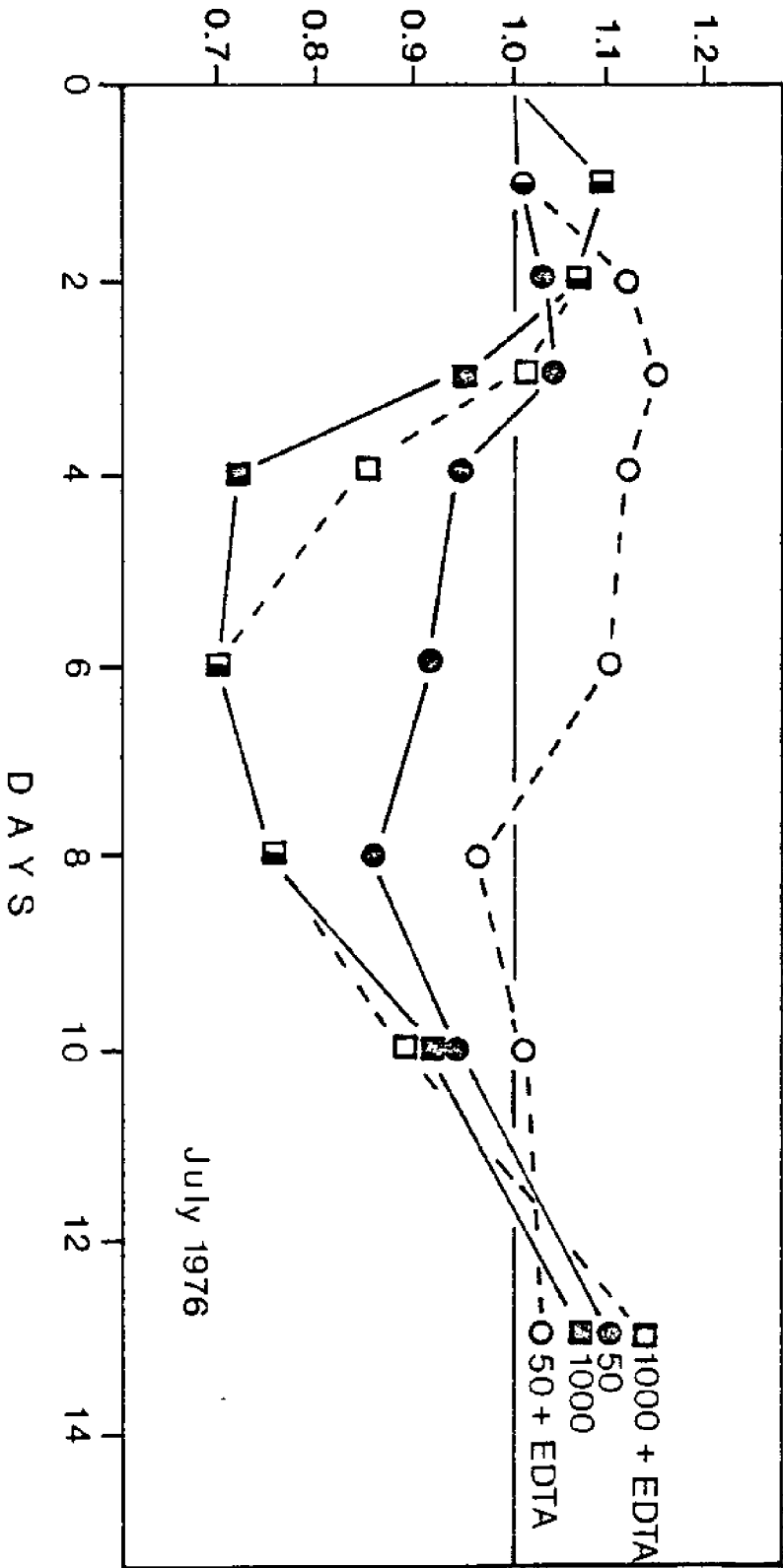


FIGURE 5. Photosynthetic rates of natural phytoplankton communities in samples spiked with iron relative to control: 50 µg/l (●—●), 1000 µg/l (■—■), 50 µg/l + EDTA (○—○), and 1000 µg/l + EDTA (□—□). 13 to 26 July 1976. Lake Erie water sampled off Point Gratiot.

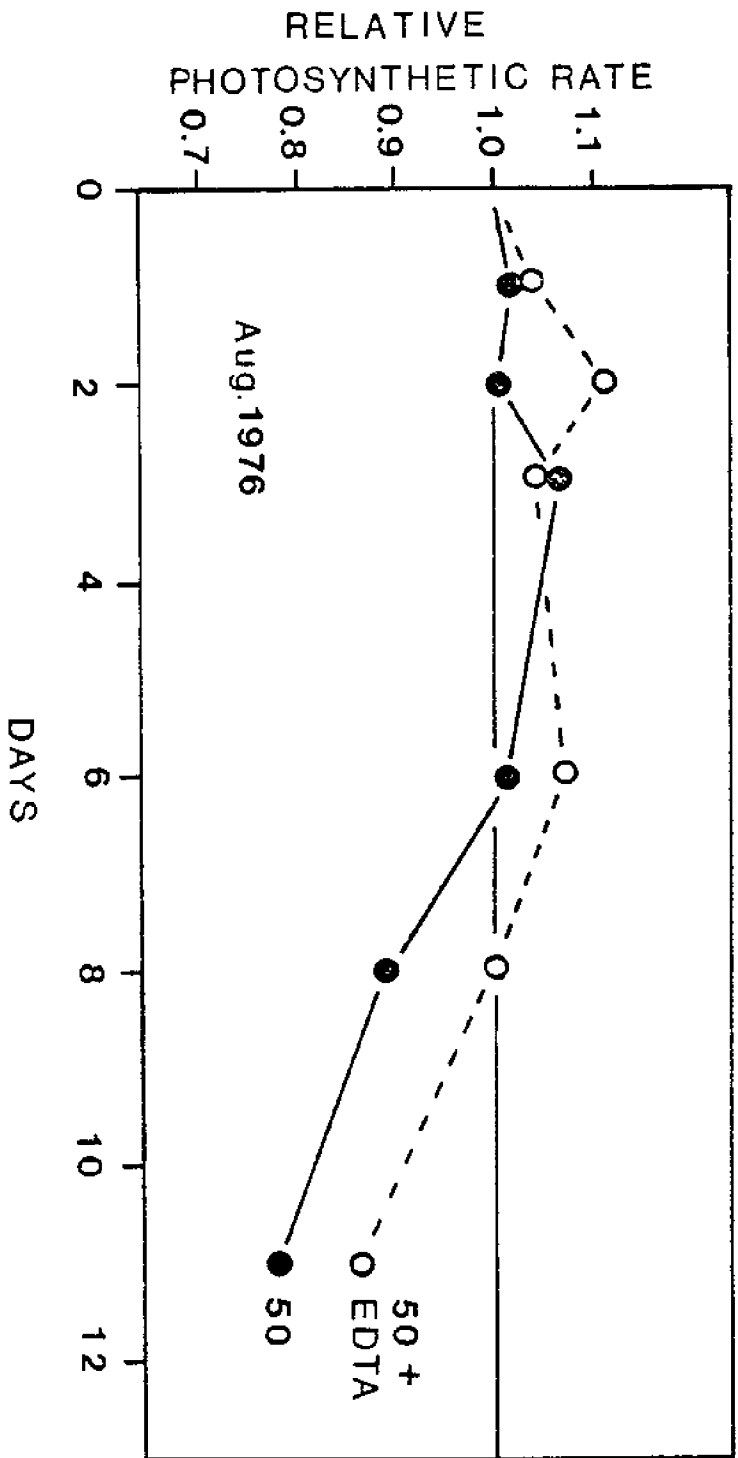


FIGURE 6. Photosynthetic rates of natural phytoplankton communities in samples spiked with iron relative to controls: 50 µg/l (●) and 50 µg/l + EDTA (○). 3 to 14 August 1976. Lake Erie water sampled off Point Gratiot.

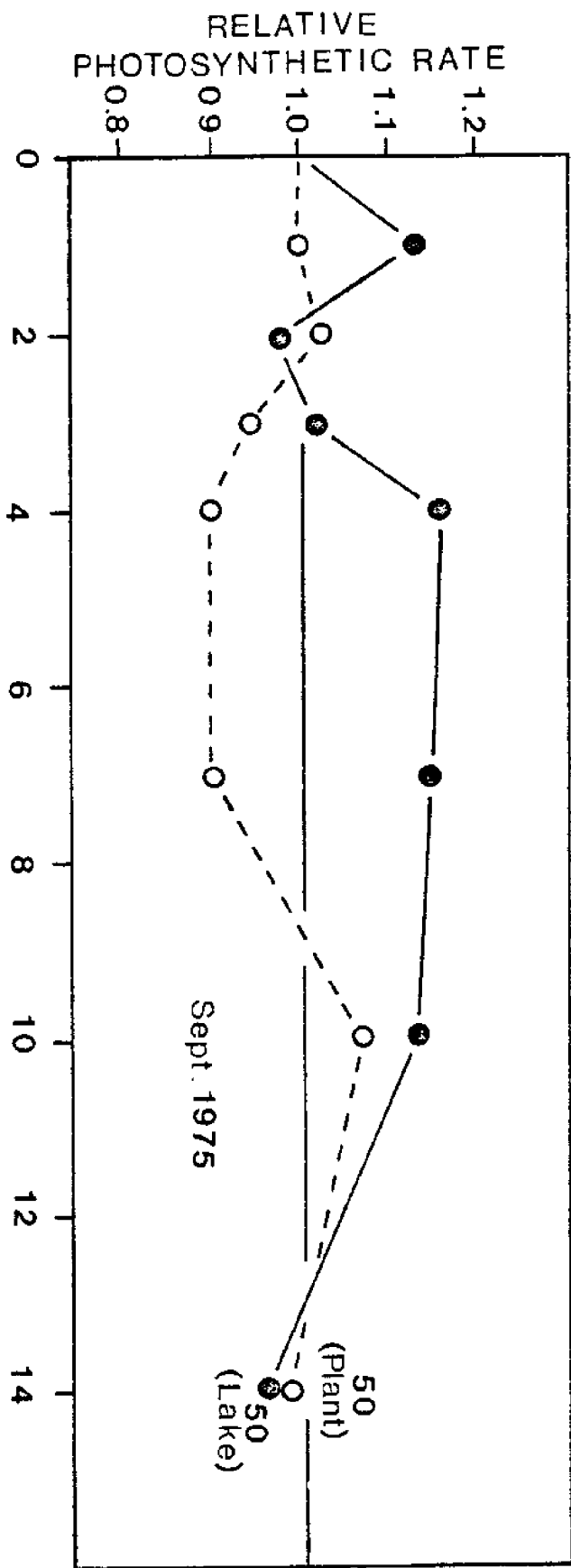


FIGURE 7. Photosynthetic rates of natural phytoplankton communities in samples spiked with 50 µg/l iron relative to controls: Lake Erie water sampled off Point Gratiot, New York (○—○) and at the Dunkirk water filtration plant. (●—●), 15 to 29 September 1975.

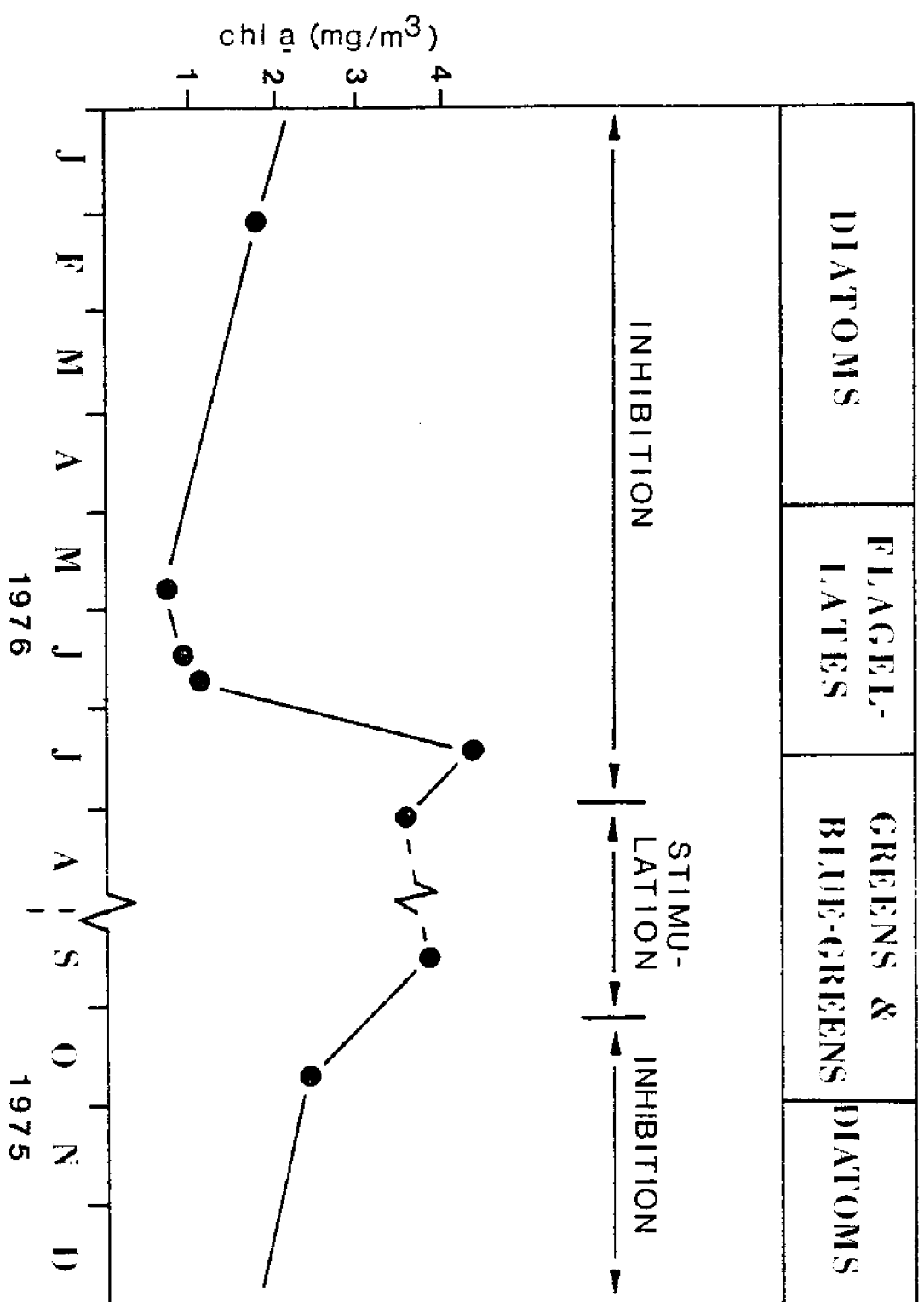


FIGURE 8. Summary of phytoplankton chlorophyll a, dominant phytoplankton types, and the effects of 50 µg/l unchelated iron on phytoplankton photosynthesis in Lake Erie, September 1975 through August 1976.

TABLE 1. Concentration ( $\mu\text{g}/\text{l}$ ) of iron in unfiltered and membrane filtered samples taken directly from the surface of Lake Erie and from the Dunkirk water filtration plant.

<u>Date</u>	Lake Water		Plant Water	
	<u>Unfiltered</u>	<u>Filtered</u>	<u>Unfiltered</u>	<u>Filtered</u>
15 September 1975	90	-	447	-
22 October 1975	79	18	408	13
16 November 1975	-	-	462	26
15 December 1975	-	-	424	58
4 February 1976	-	-	652	42
25 May 1976	192	10	-	-
14 June 1976	120	24	-	-
21 June 1976	-	4	-	-
3 August 1976	47	1	-	-

## DISCUSSION

The results of this investigation indicate that the addition of both chelated and unchelated iron affects the photosynthetic rate of the phytoplankton communities in Lake Erie. However, the degree of stimulation and inhibition by either form of iron does not appear to be extensive at low concentrations ( $50 \mu\text{g liter}^{-1}$ ). Photosynthetic rates of natural algal communities in the presence of  $50 \mu\text{g liter}^{-1}$  unchelated iron generally remained within  $\pm 10\%$  of the controls and additions of  $50 \mu\text{g liter}^{-1}$  chelated iron produced at best a 10% to 40% stimulation. At concentrations greater than  $50 \mu\text{g liter}^{-1}$ , both the chelated and unchelated iron tended to inhibit algal photosynthesis.

The response of algae to iron discharged from wastewater treatment plants depends on several factors: these include the concentration of iron in the effluent, the volume of the effluent, the volume of the receiving water, and the degree of mixing of the effluent with the receiving water. When small bodies of water receive large amounts of iron, one might expect that the iron will significantly affect the growth rate of the algae. However, considering specifically the iron discharged from wastewater treatment plants into Lake Erie such as in Dunkirk, we assume that the discharge iron will have little effect on the productivity of the phytoplankton.

The Dunkirk wastewater treatment plant, designed to treat an average flow of  $23 \times 10^6$  liters (6 million gallons) per day, discharges an effluent with an average of approximately  $600 \mu\text{g liter}^{-1}$  iron to Lake Erie (A. Bennice, personal communication). Thus, it discharges less than 14 kilograms (approximately 30 standard US lbs.) of iron per day. Though we did not monitor iron concentrations in

the discharge area during this investigation, we assume that the lake currents rapidly dilute the effluent (Niagara Mohawk Power Corporation 1976) and prevent significant increases in lake water iron concentrations. In addition, release of iron from the sediments following long term accumulation of iron in the bottom sediments should not be a problem, because the lake water remains well oxygenated throughout the zone between the shoreline and the ten meter contour where the effluent is discharged (Niagara Mohawk Power Corporation 1976). It seems, therefore, that the high dilution factor, the low concentration of iron in the effluent, and the small effects of iron on algal photosynthetic rates all indicate that the iron discharged from wastewater treatment plants such as the one in Dunkirk probably has no significant effect on the photosynthetic rates of Lake Erie phytoplankton communities.

When assessing the impact of iron additions to aquatic environments, one must also consider the effects that iron may have on the composition of the phytoplankton community. That is, iron affects the photosynthetic rate as well as the species composition of the phytoplankton community. This investigation did not examine the influence of iron additions on community structure, but work by other researchers suggests that small additions of iron may provide blue-green algae with a competitive advantage over other algal taxa.

Blue-green algae, the only group of algae known to fix nitrogen, require iron for nitrogen fixation. The concentration of available iron is therefore believed to control blue-green algal blooms in some lakes (Horne 1975). More recently, evidence supports the concept that blue-green algae release strong iron chelators (hydroxamates) that not only aid in the uptake of iron by the blue-green algae, but also suppress the growth of other algal species (Murphy, Lean and Nalewajko 1976). This

apparently accounts for results of experiments conducted in Lake Ontario, where iron additions increased the abundance of blue-green algae (Murphy and Lean 1975; Murphy et al. 1976).

Thus, iron addition to aquatic environments may change the composition of the natural community by increasing the relative abundance of blue-green algae. Since blue-green algae are often responsible for problems involving taste, odor, and surface scums in natural waters, such enhancement could, in turn, lead to water quality problems (Lin 1976).

Presently, there are insufficient data to determine if the iron in the discharge from wastewater treatment plants changes the composition of Lake Erie phytoplankton communities. However, if the dilution of wastewater effluents is rapid and extensive in Lake Erie, iron in the effluent of wastewater treatment plants should bring about no significant effect on either the composition or the photosynthetic rates of the phytoplankton communities in Lake Erie.



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