Spring isothermal mixing in the Great Lakes: evidence of nutrient limitation and nutrient–light interactions in a suboptimal light environment

G.L. Fahnenstiel, R.A. Stone, M.J. McCormick, C.L. Schelske, and S.E. Lohrenz

Abstract: During the spring isothermal mixing period (April–May) in 1993–1995, photosynthesis–irradiance and growth–irradiance experiments were conducted in Lakes Erie, Huron, Michigan, and Ontario to assess light limitation. Additionally, nutrient enrichment experiments were conducted in Lake Ontario. Results from the photosynthesis–irradiance experiments suggested that phytoplankton communities in all the lakes can be either light limited or light saturated, as the threshold parameter (I_k) was similar to mean water column irradiances ($\bar{I}_{wc}$, ratio = 1.0). Growth–irradiance experiments also suggested the potential for light saturation; mean daily irradiance exceeded the threshold growth irradiance ($I_{k,g}$) in 95% of cases. Growth rates became light saturated at lower irradiances than photosynthetic rates. Evidence for a nutrient–light interaction in controlling in situ growth rates was also found in the nutrient enrichment experiments at incubation irradiances $\geq \bar{I}_{wc}$. Our results suggest that an interaction between nutrients and light is often controlling phytoplankton growth during spring mixing in the Great Lakes. The role of these nutrient–light interactions has increased in the past decade due to increased light availability in the lower lakes caused by phosphorus load reductions and the filtering activities of nonindigenous mussels.

Résumé : Pendant la période printanière de brassage isotherme (avril–mai), en 1993–1995, nous avons réalisé dans les lacs Érié, Huron, Michigan et Ontario des expériences sur les rapports photosynthèse – éclairement énergétique et croissance – éclairement énergétique pour évaluer la limitation par l'éclairement. De plus, nous avons mené des expériences d'enrichissement en nutriants dans le lac Ontario. Les résultats des expériences photosynthèse – éclairement énergétique permettent de penser que les communautés phytoplanctoniques de tous les lacs peuvent subir soit une limitation soit une saturation par la lumière, car le paramètre seuil (I_k) était semblable à l'éclairement énergétique moyen de la colonne d'eau ($\bar{I}_{wc}$, rapport = 1,0). Les expériences croissance – éclairement énergétique semblaient aussi indiquer un potentiel de saturation par la lumière; l'éclairement énergétique moyen dépassait le seuil de croissance ($I_{k,g}$) dans 95% des cas. La saturation par la lumière était atteinte pour les taux de croissance à des niveaux d'éclairement énergétique plus bas que pour la photosynthèse. Nous avons aussi constaté dans les expériences sur l'enrichissement que l'interaction nutriants–lumière régit les taux de croissance in situ à des niveaux d'éclairement énergétique $\geq \bar{I}_{wc}$ pendant l'incubation. Nos résultats permettent de penser qu'une interaction entre les nutriants et la lumière régit souvent la croissance du phytoplancton pendant le brassage printanier dans les Grands Lacs. Le rôle de ces interactions nutriants–lumière a augmenté au cours de la dernière décennie à cause de la hausse de l'éclairement dans les lacs d'aval liée à la baisse des charges en phosphore et aux activités de filtrage des moules exotiques.

[Traduit par la Rédaction]

Introduction

In many environments, the availability of light controls phytoplankton photosynthesis and growth. The environments where light availability may limit growth are highly diverse, ranging from turbid estuaries (Pennock 1985; Alpine and Cloern 1988) to deep clear-water lakes (Brooks and Torke 1977; Nalewajko and Voltolina 1986). The St. Lawrence Great Lakes are one such environment. Because of the depth of these lakes, during periods of complete water column mixing, phytoplankton communities spend most of their time in the aphotic zone. In the spring isothermal mixing period, the ratio of the euphotic zone to water column depth typically is less than 0.5 in all the lakes (Table 1). During this period, light has been suggested as limiting phytoplankton growth in Lakes Superior (Nalewajko and Voltolina

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G.L. Fahnenstiel[1] and R.A. Stone. Lake Michigan Field Station, GLERL/NOAA, 1431 Beach Street, Muskegon, MI 49441, U.S.A.
M.J. McCormick. Great Lakes Environmental Research Laboratory, GLERL/NOAA, 2205 Commonwealth Boulevard, Ann Arbor, MI 48105, U.S.A.
C.L. Schelske. Department of Fisheries and Aquatic Sciences, University of Florida, Gainesville, FL 32653, U.S.A.
S.E. Lohrenz. Institute of Marine Science, University of Southern Mississippi, Stennis Space Center, MS 39529, U.S.A.

[1]Author to whom all correspondence should be addressed. e-mail: fahnenstiel@glerl.noaa.gov

Table 1. Station depth (SD), euphotic zone depth (EZD), EZD:SD ratio, mean irradiance in the mixed layer expressed as a percentage of surface incident irradiation (% $\bar{I}_{Z_m}$), and summary photosynthetic characteristics (P_{max} (mg C·mg Chl^{-1}·h^{-1}), α (mg C·mg Chl^{-1}·mol quanta^{-1}·m^{-2}), and I_k (mol quanta·m^{-2}·h^{-1})) (all mean values) at sampling stations.

Station	SD (m)	EZD (m)	EZD:SD	% $\bar{I}_{Z_m}$	P_{max}	α	I_k
Lake Michigan south	107	21	0.20	4.3	1.32	10.0	0.16
Lake Michigan north	144	24	0.17	3.6	1.70	11.9	0.16
Lake Huron north	64	26	0.41	8.8	0.95	7.2	0.14
Lake Huron central	68	27	0.40	8.6	1.26	8.6	0.15
Lake Huron south	37	25	0.68	14.7	1.39	8.8	0.16
Lake Erie east	57	23	0.40	8.8	2.51	12.0	0.22
Lake Ontario west	133	25	0.19	4.1	1.60	12.2	0.14
Lake Ontario east	159	26	0.16	3.6	1.51	9.0	0.19

1986), Ontario (Lean et al. 1987; Millard et al. 1996), Erie (Lean et al. 1983), and Michigan (Brooks and Torke 1977). Light limitation may even occur in the surface mixed layer during thermal stratification (Nalewajko et al. 1981). Because of the primary importance of light to phytoplankton growth, the value of phosphorus control in maintaining and controlling phytoplankton biomass has been questioned (Nalewajko et al. 1981).

During the past decade, significant changes have occurred in the water quality of the Great Lakes due to phosphorus load reductions and the filtering activities of nonindigenous mussels (Johengen et al. 1994; Fahnenstiel et al. 1998). Large increases in transparency have been noted in the lower Great Lakes (Fahnenstiel et al. 1998). This increased light availability would not only affect phytoplankton growth rates during periods of light limitation, but may even produce nutrient limitation, or a nutrient–light interaction (Rhee and Gotham 1981; Fahnenstiel et al. 1984) where light limitation existed in the past. Thus, simple light limitation during spring isothermal mixing may not be as common in the 1990s, particularly the lower Great Lakes, as it was in the 1980s. The intent of this research was to examine the roles of nutrients and light during spring isothermal mixing in the Great Lakes in the early to mid-1990s, with particular emphasis on the potential for nutrient limitation.

In this study, as in all studies of nutrient and light limitation, the distinction between rate and biomass limitation must be emphasized. In laboratory chemostats of algae, the distinction between growth rate and biomass limitation are clear, but for field communities of phytoplankton, it is more difficult. Rate limitation is easier to document for field communities than biomass limitation. Nutrients can limit biomass and rates. Light is primarily a rate-limiting factor but can limit biomass when light levels are very low and self-shading becomes an important concern (i.e., turbid lake). Temperature is also primarily a rate-limiting factor. In natural communities, light and temperature can indirectly limit biomass through their control over growth rates, as the balance between growth and loss rates is an important factor controlling in situ phytoplankton biomass (Wofsy 1983). In our experiments, we focused on examining the role of light in limiting rates of photosynthesis and growth. If phytoplankton were found to be light saturated at in situ light levels, then other factors (nutrients, temperature, etc.) may potentially be limiting. Additionally, nutrient enrichment experiments were conducted to examine the potential for nutrients to control both rates and biomass, as the large increase in nutrient concentration added as part of the enrichment can increase both rates and biomass. While biomass limitation can be elucidated with nutrient enrichment experiments, the large nutrient perturbation sometimes reduces the applicability to in situ conditions.

Methods

Sampling was conducted at eight stations in the St. Lawrence Great Lakes aboard the RV *Laurentian* (Fig. 1). Two or three stations in the offshore region of each lake were sampled during the spring isothermal period (April–May) of each year from 1993 to 1995. Each station was occupied for 3–24 h.

At each station, a Seabird CTD (conductivity, temperature, and depth meter equipped with Sea-Tech fluorometer and transmissometer, 25-cm beam path) cast was made from the surface to just above the bottom at a 2-Hz sampling rate. The CTD profiles were used to determine the discrete sampling depths for water collections. Secchi disk transparency was measured with a black/white 25-cm disk. Underwater light extinction of photosynthetically active irradiation (k_{PAR}) was measured with a LICOR 193SB scalar (4π) light sensor and LICOR 1000 data logger and (or) a Biospherical integrating natural fluorometer (INF-3000) equipped with a downwelling scalar PAR sensor located on the ship. Surface incident irradiation was measured on the ship using a LICOR sensor and data logger. At stations sampled in darkness, beam attenuation values were converted to extinction coefficients using the conversion of Fahnenstiel et al. (1995), developed in Saginaw Bay with the same transmissometer. These k_{PAR} values were used to calculate the depth of the euphotic zone (1% isolume).

Discrete samples were taken from three to six depths in the euphotic zone using a modified Niskin bottle (all rubber parts were replaced with silicone- or teflon-coated parts) and poured into a 24-L carboy (one carboy for each depth). All water samples were taken from these carboys. Chlorophyll samples were filtered onto Whatman GF/F filters, extracted with *N*,*N*-dimethylformamide, and analyzed fluorometrically (Speziale et al. 1984).

Phytoplankton samples were preserved in amber bottles with 0.5% Lugol's solution. These samples were filtered onto microscope slides according to the procedure of Dozier and Richerson (1975). A minimum of 300 phytoplankton entities were enumerated under both high (1000–1200×) and low magnification (200×). Cell volumes were estimated by determining average cell dimensions of a minimum of 100 cells for each dominant taxon and at least 10 cells for rare taxa and then applying these dimensions to appropriate geometric shapes. The cell volumes used for this study were from a compilation of values from this and previous studies

Fig. 1. Location of sampling stations.

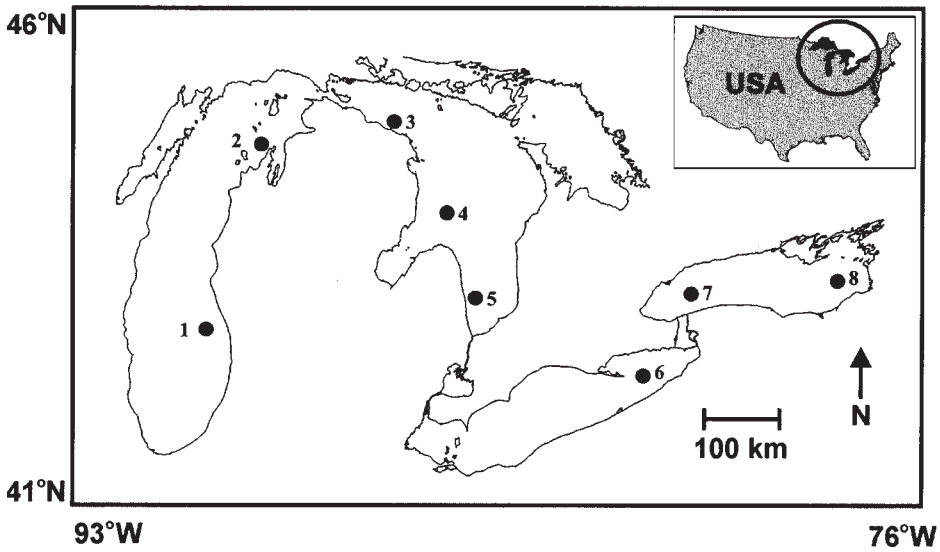
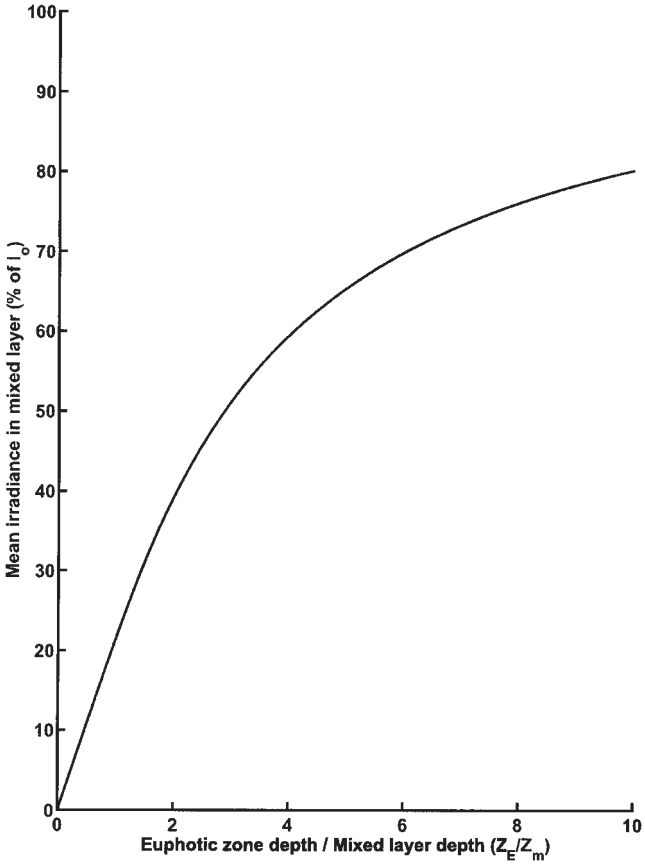


Fig. 2. Relationship between mean irradiance in the mixed layer expressed as percentage of incident irradiation (I_0) and the ratio of euphotic zone depth to mixed layer depth.



of the Great Lakes. Phytoplankton volumes were converted to carbon units using the respective equations from Strathman (1967) for diatoms and from Verity et al. (1992) for nondiatoms.

Phytoplankton photosynthesis was measured with the ^{14}C technique in a photosynthetron (Fahnenstiel et al. 1995). Water samples were inoculated with ^{14}C, and subsamples of 3 mL were incubated in acid-rinsed scintillation vials for 40 min. Eighteen light levels from <1 to 1800 mol quanta·m^{-2}·s^{-1} were used. After incubation, subsamples were acidified and sparged with air for 15 min. Time-zero blanks were taken and subtracted from all light values. Total carbon dioxide was determined from alkalinity and pH measurements.

Photosynthetic rates, normalized to chlorophyll, were used to construct a single photosynthesis–irradiance curve using the methods outlined in Fahnenstiel et al. (1989). Three parameters were determined from this model: P_{max}, maximum photosynthetic rate at light saturation (milligrams carbon per milligram chlorophyll per hour); α, initial linear slope at low irradiances (milligrams carbon per milligram chlorophyll per mole quanta per square metre); β, negative slope at high irradiance (same units as α). If the 95% confidence interval of β included zero, a simple two-parameter model was used (Fahnenstiel et al. 1989). The light saturation parameter (I_k), which has been used to differentiate between the light-limited and light-saturated regions of photosynthesis (Millard et al. 1996), was determined as the ratio P_{max}:α.

Phytoplankton growth rates were determined by ^{14}C labeling into chlorophyll *a* (Redalje and Laws 1981; Goericke and Welschmeyer 1993). This technique is basically an extension of the traditional ^{14}C experiment except that the end-point is labeling into chlorophyll *a*. The chlorophyll-specific growth rate equals the carbon-specific growth rate when growth is balanced. Significant photoadaption may decouple carbon and chlorophyll synthesis. To minimize photoadaptation, all experiments were conducted for 24 h (dawn to dawn) and incubated at as close to in situ irradiances as possible with static bottle incubations. These type of precautions should minimize photoadaptation artifacts and provide reasonable estimates of growth based on chlorophyll synthesis (Goericke and Welschmeyer 1993; Gallegos and Vant 1996). For our growth rate experiments, water samples were collected 1–4 h before dawn. Clean 2-L samples were incubated with 400 μCi of ^{14}C (1 μCi = 37 GBq) and placed in an in situ simulated incubator where the spectral quality of light was similar to in situ conditions. Six incubators with different light levels (50, 25, 12, 6, 3, and 1% of surface irradiance) were used to simulate the quantity of light received at depth. All growth samples were incubated at the irradiance that most closely approximated in situ irradiance. In most cases (57%), the samples were collected from the depth that received mean water column irradiance (Fig. 2) and then incubated at this mean water column irradiance. These incubations will be referred to as simulated in situ incubations. The other cases (43%) are where samples were collected at specific depths and then incubated at the irradiances received at those specific depths. Chlorophyll *a* was ex-

tracted and analyzed for radiochemical activity using the techniques of Redalje (1990). Dark and time-zero controls were performed. Growth rates were calculated from the specific activity of chlorophyll *a* using eq. 6 from Goericke and Welschmeyer (1993). By combining growth rates from all experiments over a 3-year period as incident irradiation varied, a growth–irradiance curve was constructed for each lake. These growth–irradiance curves were then fit with a nonlinear model similar to that used for photosynthesis–irradiance curves described above. Two parameters were determined from these models: μ_{max}, maximum growth rate at light saturation; α_g, initial linear slope at low irradiances. From these two parameters, a light saturation parameter ($I_{k,g}$) was calculated as the ratio μ_{max}:α_g.

On eight occasions, phytoplankton growth rates were also estimated with the dilution technique (Landry and Hassett 1982). These experiments consist of a series of incubations in which raw lake water (unfiltered) is diluted with varying amounts of filtered water; the phytoplankton growth rate is the intercept of the growth rate from each dilution regressed against the fraction of raw lake water. Diluted samples (2- to 20-fold) were prepared by mixing appropriate volumes of raw lake water with filter sterilized lake water (<0.2 μm) and incubating in 4-L polycarbonate bottles. Because only growth rates were determined, more highly diluted samples (8- to 20-fold dilutions) were used. Water for these dilution experiments was taken from the depth at which the mean irradiance in the water column was received (Fig. 2). All dilution samples were incubated in the simulated in situ incubator described above at the mean irradiance in the mixed layer and at lake water temperature. Samples for phytoplankton counts were removed at 0, 48, and 72 h. Growth rates were calculated from changes in phytoplankton carbon determined from microscopic counts.

Nutrient enrichment experiments were conducted with water samples from Lake Ontario (Station 7 or 8). For these experiments, water was incubated in 20-L carboys in a simulated in situ incubator to assess the potential for nutrient stimulation of phytoplankton growth and biomass. These samples were maintained at ambient temperature and light level (2–12% of incident irradiation). Incubator temperature was maintained within 1°C of lake water temperature. Two experiments were conducted, and for each experiment, two light levels were used. For the spring 1993 experiment, light levels were 2 and 6% of incident, whereas for the spring 1994 experiment, light levels were 4 and 12% of incident. This range of light intensities allowed us to examine the potential for light limitation in a range of naturally occurring irradiances during the spring isothermal period. A complete set of macronutrients (phosphorus, nitrogen, and silicon) and micronutrients (vitamins and trace metals) were added in order to alleviate nutrient limitation. The concentrations of added macronutrients were 0.5 μM phosphorus, 8 μM nitrogen, and 25 μM silicon. The vitamins and trace metals were supplied in proportion to phosphorus based on WC media (Guillard and Lorenzen 1972).

Calculations for light parameters were as follows. Irradiance in the water column at a specific depth (I_Z) can be determined by

$$(1) \qquad I_z = I_0 e^{-kZ}$$

where k is the extinction coefficient of PAR, I_0 is surface irradiance, and Z is depth. Mean irradiance in the mixed layer ($\bar{I}_{Z_m}$) can be calculated by integrating eq. 1 over the mixed layer:

$$(2) \qquad \bar{I}_{Z_m} = \frac{I_0}{kZ_m}[1 - e^{-kZ_m}]$$

where Z_m is the depth of the mixed layer. For the spring isothermal mixing period in the Great Lakes, the mixing depth equals the station depth or water column depth ($\bar{I}_{Z_m} = \bar{I}_{we}$). Assuming that the euphotic zone depth (Z_E) equals the depth of 1% of I_0, then eq. 2 can be solved for the mean irradiance in the mixed layer as a percentage of surface incident irradiance ($\%\bar{I}_{Z_m}$):

$$(3) \qquad \%\bar{I}_{Z_m} = 21.7156\left(\frac{Z_E}{Z_m}\right)\left[1 - e^{-4.605\left(\frac{Z_m}{Z_E}\right)}\right].$$

Using this equation or Fig. 2, the mean irradiance in the mixed layer as a percentage of surface irradiance can be related to the ratio of the euphotic zone depth to mixed layer depth. Even when the euphotic zone equals the mixed layer, phytoplankton receive approximately 22% of surface irradiance.

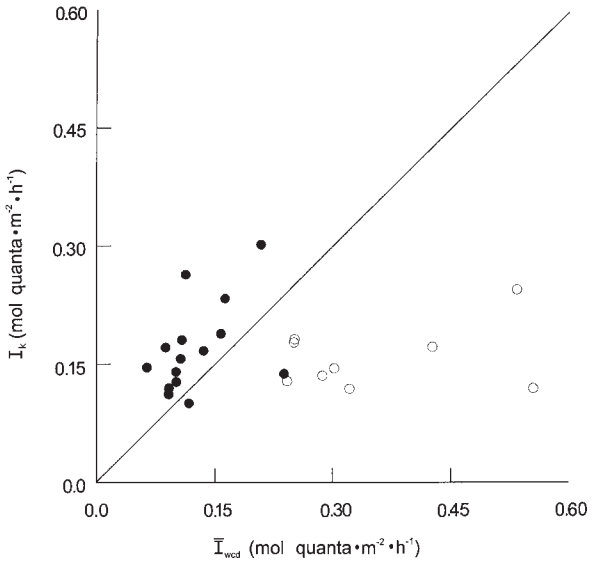
Results

The sampling periods during each year corresponded to spring isothermal mixing (late April – May). For spring 1993 and 1994, all stations exhibited isothermal conditions with temperatures between 1 and 4°C. Sampling occurred 2 weeks later in May of 1995 than in 1993 and 1994, and slight stratification was found at two stations that were isothermal in previous years. Surface temperatures at the southern Lake Huron station and eastern basin of Lake Erie were 5°C, whereas those at all other stations in Lakes Huron, Michigan, and Ontario ranged between 2.5 and 4°C. The results from these two stations were not included in any analysis.

The euphotic zone was relatively similar among all stations sampled, ranging from 21 to 27 m (Table 1). The ratios of euphotic zone depth (EZD) to station depth (SD) were more variable, ranging from 0.16 to 0.68. These differences were strongly related to the station depth, as deeper stations (Lake Ontario and northen Lake Michigan) had the lowest ratios. Using eq. 3 or Fig. 2 and the EZD:SD ratios, the percentage of surface irradiance received in the water column during spring isothermal mixing ranged from 4 to 15% (Table 1).

Isothermal conditions do not necessarily imply vertically homogeneous distributions of phytoplankton, and this condition is important for our analysis. For all of our isothermal samplings, phytoplankton carbon and chlorophyll concentrations were uniform with depth ($p > 0.05$). In all cases, chlorophyll concentrations varied by <10% throughout the water column and phytoplankton carbon by <20%. Moreover, the physiological condition of phytoplankton communities in the isothermal water column was also similar with depth. Photosynthesis–irradiance parameters were measured with depth at several stations and no significant differences were noted ($p < 0.05$). In vivo fluorescence profiles from the CTD casts can also be used to examine the homogenous distributions of phytoplankton, and possibly even their physiological conditions. Because fluorescence yields can vary over short time periods (minutes), the fluorescence profile represents phytoplankton abundance and photoacclimation. In most cases, in vivo chlorophyll fluorescence was uniform with depth (82% of profiles). Even for profiles where statistical differences were noted (18%), chlorophyll fluorescence varied by <10% in the water column. Thus, during all of our samplings, phytoplankton biomass and physiological condition were vertically uniform.

Fig. 3. Relationship between the light saturation parameter for photosynthesis (I_k) and mean daylight water column irradiance ($\bar{I}_{wcd}$) calculated for the preceeding 3 days. Open circles indicate Lake Huron data.

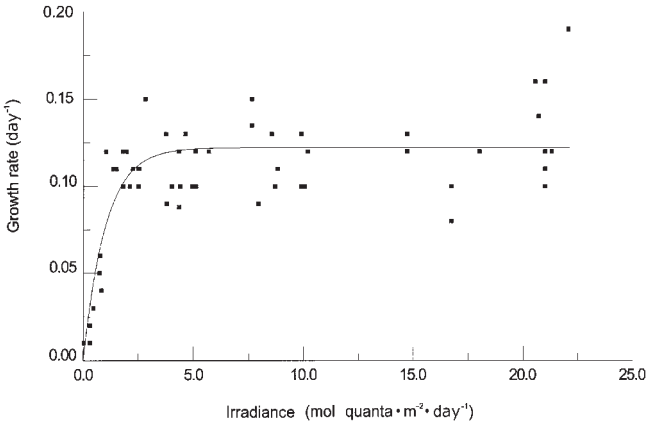


Photosynthetic rate parameters were relatively similar among stations (Table 1). The P_{max} values ranged from 0.95 to 2.51 mg C·mg Chl^{-1}·h^{-1}, with significantly lower values associated with Lake Huron stations ($p < 0.05$). The α values were similar across all stations, ranging from 7.2 to 12.2 mg C·mg Chl^{-1}·mol quanta^{-1}·m^{-2} ($p > 0.05$). The threshold parameter I_k was also similar among all stations, ranging from 0.14 to 0.22 mol quanta·m^{-2}·h^{-1} ($p > 0.05$).

In our study, comparisons of I_k and mean in situ irradiance ($\bar{I}_{wcd}$, calculated as mean water column irradiance during the daylight period) were used to determine light-limited conditions for spring phytoplankton (Fig. 3). A 3-day mean $\bar{I}_{wcd}$ was used to average out any single-day variation and to be more consistent with the generation times of the phytoplankton. Photosynthesis for natural phytoplankton communities was found to be both light limited and light saturated in the spring, as the ratio I_k:$\bar{I}_{wcd}$ was both above and below 1 but strongly dependent on the EZD:SD ratios. Those communities with relatively high EZD:SD ratios (all Lake Huron values and one Lake Erie value) exhibited $\bar{I}_{wcd}$ values greater than I_k (right-hand side of plot). On the other hand, phytoplankton communities with lower EZD:SD ratios (all Lakes Michigan and Ontario values and two Lake Erie values) exhibited I_k values that were higher than $\bar{I}_{wcd}$ values, thus suggesting light limitation. Because many values from all lakes were close to 1, the potential for both light limitation and saturation cannot be dismissed. For example, even though all Lake Michigan ratios were >1, the mean of all ratios was only 1.3.

Growth rates determined from the synthesis of chlorophyll *a* were relatively similar among the lakes, ranging from 0.01 to 0.19·day^{-1} (Figs. 4 and 5). Maximum growth rates (μ_{max}) were similar for each lake, ranging from 0.12 to 0.16·day^{-1} ($p > 0.05$). Even though the initial linear slopes ($I_{k,g}$) were also similar among lakes ($p > 0.05$, range 0.08–0.15 m^2·mol quanta^{-1}), limited sampling in the low-light region prevented any thorough comparison.

Fig. 4. Growth rate versus incubation irradiance for all experiments.



Precautions were taken to ensure that our chlorophyll synthesis growth rates were similar to carbon-based growth rates: 24-h incubations started at dawn, incubation at in situ irradiances, etc. Because the generation time of Great Lakes phytoplankton was long (mean = 6.3 days) relative to the incubation duration (24 h), it is likely that phytoplankton were adapted to a mean water column irradiance and that significant photoadaption did not occur during our relatively short incubations. To evaluate the effectiveness of these precautions and whether our growth rates were similar to carbon-based growth rates, on eight occasions, we compared chlorophyll synthesis growth rates with dilution growth rates calculated from changes in phytoplankton carbon. No significant differences were found between chlorophyll and dilution growth rates ($p > 0.05$; mean dilution = 0.14·day^{-1}, mean chlorophyll = 0.12·day^{-1}), suggesting that the assumption of balanced growth was met and that our chlorophyll synthesis growth rates are reasonable measures of the growth rate. These dilution growth rates were used only for comparative purposes and are not included in any further analysis.

The potential for light limitation was also examined in relation to phytoplankton growth rates (Figs. 4 and 5). The light saturation parameter for all growth rates, $I_{k,g}$, was 1 mol quanta·m^{-2}·day^{-1} (μ_{max} = 0.12·day^{-1}, α_g = 0.12 m^2·mol quanta^{-1}; R^2 = 0.66, $p < 0.05$) (Fig. 4). For the individual lakes, $I_{k,g}$ values were higher for Lakes Erie and Huron (1.6 and 1.5 mol quanta·m^{-2}·day^{-1}, respectively) than for Lakes Michigan and Ontario (1.0 and 0.9 mol quanta·m^{-2}·day^{-1}), respectively. Growth rates became light saturated at irradiance of around 3–4 mol quanta·m^{-2}·day^{-1}. Mean water column irradiance ($\bar{I}_{wc}$, calculated for a 24-h period) exceeded $I_{k,g}$ values in almost all cases (95% of days) and the light saturation level (4 mol quanta·m^{-2}·day^{-1}) on 26% of days. These data suggest light saturation of growth was common at in situ irradiances. Growth versus simulated irradiance curves for the individual lakes also demonstrate the potential for light saturation (Fig. 5). The majority of simulated in situ growth rates were near μ_{max}, and simulated irradiances were greater than $I_{k,g}$ values. However, because many of the growth rates were close to $I_{k,g}$, the possibility of either light limitation or light saturation exists (Fig. 5).

Fig. 5. Growth rate versus incubation irradiance for (*a*) Lake Michigan, (*b*) Lake Huron, (*c*) Lake Erie, and (*d*) Lake Ontario. Open squares are for growth experiments incubated at mean water column irradiance.

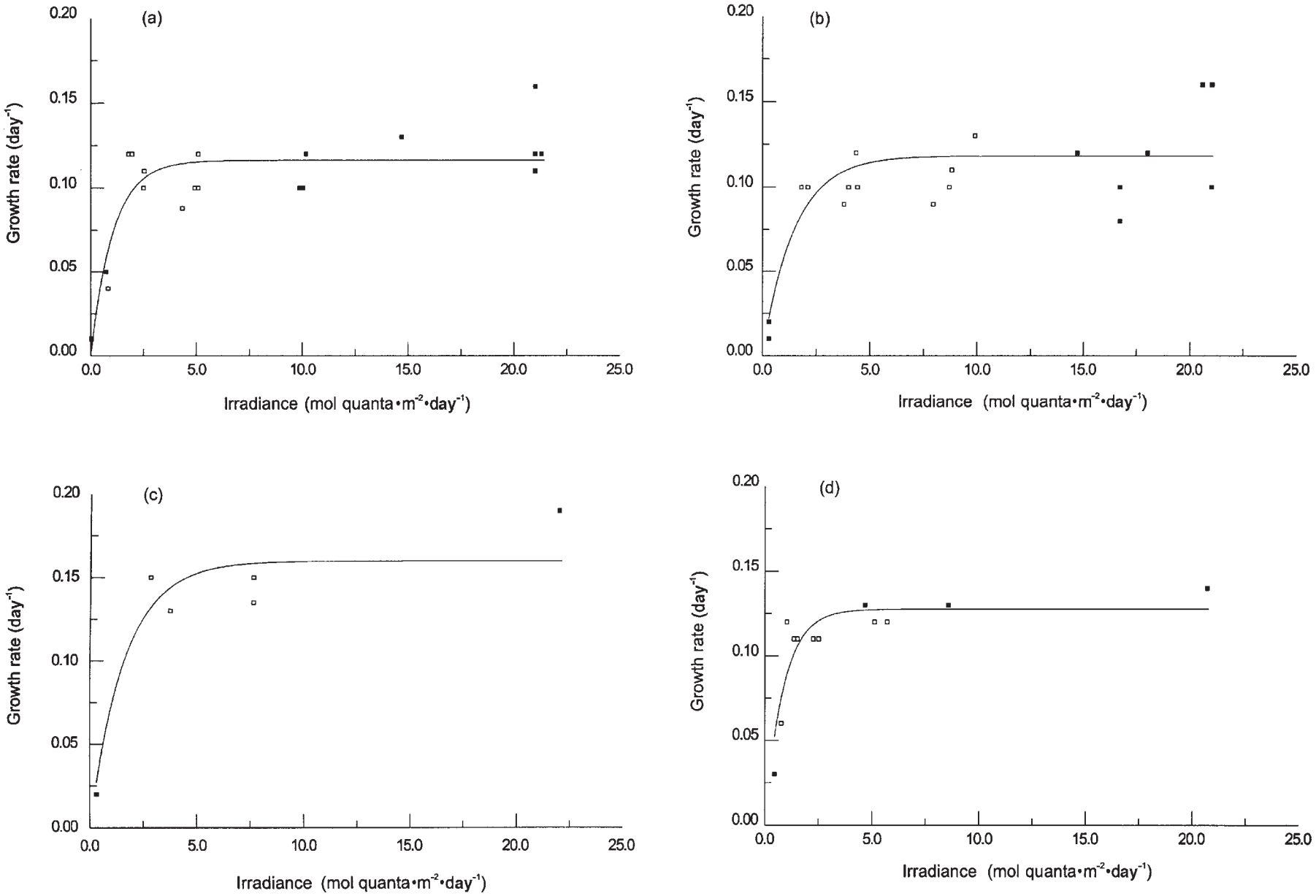


Fig. 6. Chlorophyll concentrations from May 1993 nutrient enrichment experiment. Experiments were performed at 2% (circles) and 6% (squares) of surface irradiance. Solid symbols indicate nutrient treatments.

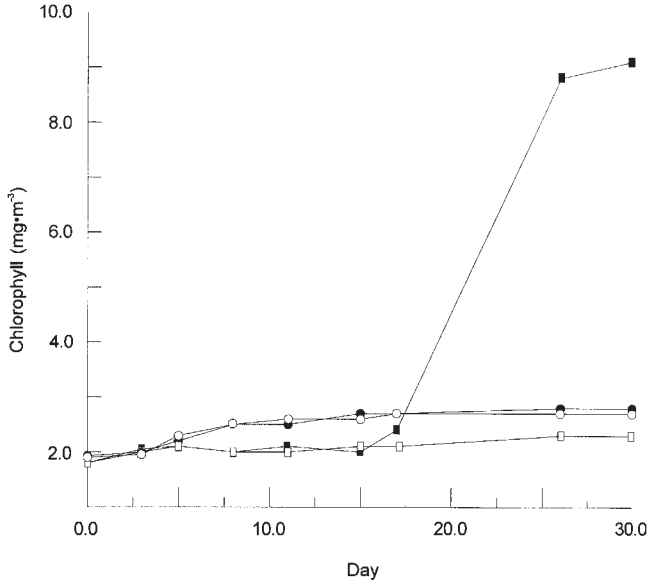
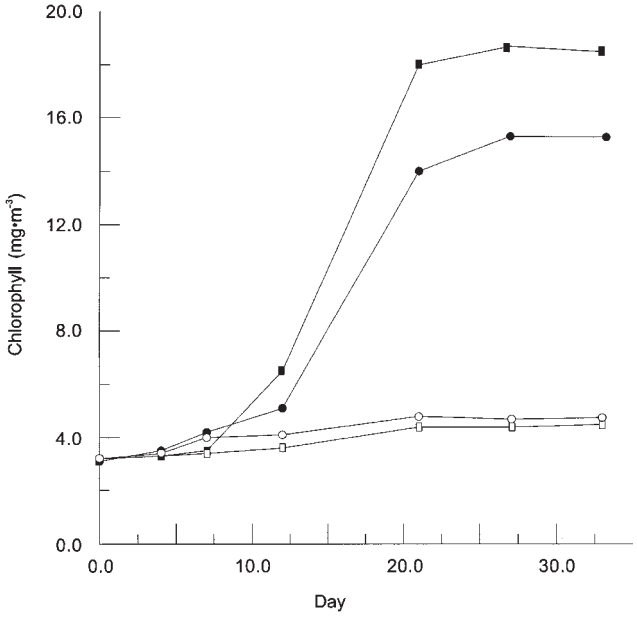


Fig. 7. Chlorophyll concentrations from May 1994 nutrient enrichment experiments. Experiments were performed at 4% (circles) and 12% (squares) of surface irradiance. Solid symbols indicate nutrient treatments.



Lakes Ontario and Michigan had a greater percentage of simulated in situ growth rates in the light-limited region of the curve than did Lake Erie or Lake Huron.

Nutrient enrichment experiments demonstrated the potential for nutrient limitation in Lake Ontario. In the May 1993 experiment, at the 2% of surface irradiance treatment, both control and nutrient treatments exhibited similar biomass throughout the experiment (Fig. 6). For the 6% light treatment, a significant increase in biomass was noted at day 26 ($p < 0.05$), as chlorophyll concentration increased almost four times higher than controls. Even though biomass increased only slightly by day 17, growth rates and P_{max} values for the nutrient treatment were significantly greater (about 50%, $p < 0.05$) than control values on day 17. In the May 1994 experiment, a significant nutrient effect was noted for nutrient treatments at both the 4 and 12% light levels ($p < 0.05$) (Fig. 7). On day 7, significant increases in growth rates for nutrient treatments were noted at the 12% (40% increase, $p < 0.05$) and 4% (30% increase, $p < 0.05$) light levels. The results from these experiments suggest that the threshold for nutrient limitation was >2 but <4% of surface irradiation. The actual incident surface irradiation during these two experiments was 38 and 39 mol quanta·m^{-2}·day^{-1}, respectively. Thus, the absolute threshold for these two experiments was between 0.8 and 1.6 mol quanta·m^{-2}·day^{-1}, which is very similar to the light saturation parameter value for growth of 1 mol quanta·m^{-2}·day^{-1}.

The consistency of phytoplankton biomass in the control treatments is noteworthy and suggests that the results may be applicable to in situ conditions. In the May 1993 experiment, chlorophyll concentrations in the controls changed only 25 and 40% after 30 days (Fig. 6). Moreover, phytoplankton growth rates in the controls were not significantly different after 27 days ($p < 0.05$). In the May 1994 experiment, chlorophyll concentrations changed only 30–40% after 33 days (Fig. 7). Also, photosynthetic and growth parameters were similar in controls after 21 days ($p < 0.05$).

Discussion

Phytoplankton photosynthesis and growth during spring isothermal mixing in the Great Lakes are not strictly controlled by the availability of light. In many cases, phytoplankton growth and, to a lesser extent, photosynthesis rates were at light saturation, suggesting that factors other than light (nutrients, temperature, etc.) may be important in controlling rates. Although we did not sample Lake Superior, the similarity of EZD:SD ratios between Lake Superior and the other lakes suggests that other factors may also be important in Lake Superior. Lake Superior has a greater euphotic zone (about 42–46 m; Fahnenstiel et al. 1998) and greater depths (mean depth = 149 m) than the other Great Lakes, but the EZD:SD ratio is about 0.29, which is slightly higher than the mean ratio for Lakes Michigan and Ontario but lower than the ratio for Lake Huron. The potential limiting factor(s) for each lake can vary depending on EZD:SD ratios, with Lakes Michigan, Ontario, and Superior more likely to be light limited than Lakes Erie and Huron.

Despite large differences in geographic position among stations, growth rate parameters were very similar among stations. Maximum growth rates and growth efficiency (α) were not significantly different among any of the lakes ($p < 0.05$). Thus, constraints on growth rates may be similar among all the lakes, and experimental evidence from one lake, e.g., Lake Ontario, may be applicable to all the lakes during the spring period. Moreover, similarity in growth rates across all of these stations may be used to suggest that regional climatic factors such as light and temperature provided important constraints on growth rates. While these factors are likely important, nutrient concentrations and food

web structure are also similar across these lakes during spring mixing (Fahnenstiel et al. 1998). Total phosphorus concentrations ranged from 0.1 to 0.2 nM. Food web characteristics, i.e., autotrophic to heterotrophic, picoplankton to nannoplankton and microplankton ratios, etc., were also similar among lakes (Fahnenstiel et al. 1998). Thus, given the similarity of primary controlling factors, it is not surprising that growth rates are similar across all of the lakes.

Phytoplankton growth in the Laurentian Great Lakes may often be controlled by a complex interaction of light, nutrients, and possibly even temperature. All three of these environmental factors can be or are suboptimal in all of the Great Lakes during the spring isothermal mixing period. During spring mixing, both light availability and nutrient availability in the lakes are often near the threshold values for both nutrient and light limitation; neither light nor nutrients are in abundant supply. Soluble phosphorus concentrations during this period were typically <50 nM (G.L. Fahnenstiel, unpublished data). Moreover, the low temperatures (1–4°C) found at this time of year are often suboptimal for growth (Stoermer and Ladewski 1976; Reynolds 1984). Historically, light has been suggested as limiting phytoplankton growth in the Great Lakes during spring isothermal mixing (Nalewajko and Voltolina 1986; Scavia and Fahnenstiel 1987; Millard et al. 1996). The important role that light plays in controlling phytoplankton photosynthesis and growth was evident in our study. In situ light conditions were suboptimal for photosynthesis and growth during many of the sampling periods. Only in Lake Huron were ambient light levels consistently higher than threshold values for photosynthesis. However, the potential that other factors may be important in controlling phytoplankton growth during this period is a relatively new idea.

Another important factor controlling phytoplankton growth during spring isothermal mixing is nutrient availability. In Lake Ontario, which had the lowest EZD:SD ratios and thus the most potential for strict light limitation, nutrient additions at simulated in situ irradiances produced significant increases in phytoplankton growth rates and biomass. These results, combined with observation of light saturation at in situ irradiances, suggest that nutrients can limit phytoplankton growth. This nutrient role is consistent with earlier work in the upper Great Lakes (Superior and Michigan). Previous work has documented that phytoplankton from the upper Great Lakes exhibit low to moderate phosphorus deficiency during spring isothermal mixing (Nalewajko and Voltolina 1986; G.L. Fahnenstiel, unpublished data). This lack of strong phosphorus, or nutrient, limitation is consistent with our nutrient enrichment experiments, in which only a slight increase in growth rate occurred with nutrient additions. Only in the lower Great Lakes (Ontario and Erie) has no evidence of nutrient limitation been found during spring isothermal mixing (Lean et al. 1983, 1987; Millard et al. 1996).

Although nutrients may be important in controlling phytoplankton growth rates, they likely do not act alone but rather in combination with light and possibly even temperature. Evidence for this conclusion is based on several factors, including direct experimental evidence from the growth and nutrient enrichment experiments. In most instances, neither light nor nutrient availability alone constrained growth rates, for if either limitation was alleviated, only slight increases in growth rate were found, and rates remained relatively low. In situ growth rates were low, ranging from 0.04 to 0.15·day^{-1}, and the mean light-saturated rate μ_{max} was only 0.12·day^{-1}. Thus, under light-saturating conditions, growth rates would still be <0.2·day^{-1}. Similarly, alleviating nutrient limitation by adding a saturating nutrient concentration (0.5 μmol additions of phosphorus usually produce V_{max} in the Great Lakes; G.L. Fahnenstiel, unpublished data), as was done in nutrient enrichment experiments, increased growth rates by only 30–50%, leaving them still <0.3·day^{-1} in most cases. However, when both nutrient availability and light availability were increased, the highest growth rates (about 0.3·day^{-1}) and final biomass were found (12% light level with nutrient addition), suggesting that nutrients and light interact in controlling in situ rates. Because growth rates were only 0.3·day^{-1} when both light and nutrients were saturating, suboptimal temperatures (1–4°C) likely constrain maximum growth rates and possibly even in situ rates. Temperature has been thought to limit the maximum growth rate, whereas other factors (e.g., nutrients and light) were considered more important in controlling in situ rates (Epply 1972). However, at suboptimal temperatures during spring mixing, temperature may also interact with nutrient availability and constrain in situ growth rates. Under nutrient-limited conditions, suboptimal temperatures interact with nutrients and light in a complex manner and likely aggravate nutrient limitation (Rhee 1982). Also, temperature influences light requirements for growth (Morgan and Kalff 1979).

The likelihood that an interaction of light and nutrients controls phytoplankton growth in the Great Lakes during spring isothermal mixing is supported by a comparison of previous work documenting nutrient–light interactions and present conditions in the Great Lakes. Several authors have used laboratory cultures to evaluate interactions of nutrients and light in controlling growth rates when light and temperature are at suboptimal levels (Rhee and Gotham 1981; Fahnenstiel et al. 1984; Healey 1985). The combined effects of nutrient and light limitation are greater than the sum of individual effects, and at suboptimal irradiances, nutrients and light can compensate for each other in maintaining a given growth rate (Rhee and Gotham 1981). Rhee (1982) suggested that this compensatory relationship would make it difficult to separate the effects of light and nutrient limitation in the euphotic zone. Healy (1985) noted that over a narrow range of low irradiances, simultaneous limitation of light and nutrients can be found. Using laboratory cultures from the deep chlorophyll layer in Lake Michigan, Fahnenstiel et al. (1984) described a similar nutrient–light interaction, where the maximum steady-state yield was determined by the combined effects of nutrients and light. These nutrient–light interactions occurred at irradiances ranging from 8 to 78 μmol quanta·m^{-2}·s^{-1}. For unialgal cultures of *Synechococcus*, Healy (1985) reported interactions between 10 and 20 μmol quanta·m^{-2}·s^{-1}, and for cultures of *Scenedesmus*, Rhee and Gotham (1981) reported interactions between 7 and 17 W·m^{-2} or approximately 32–78 μmol quanta·m^{-2}·s^{-1} (J. Cullen, Dalhousie University, Halifax, N.S., personal communication). For natural assemblages of Lake Michigan phytoplankton, Fahnenstiel et al. (1984) found interactions to occur between 8 and 57 μmol quanta·m^{-2}·s^{-1}. These values for laboratory cultures are simi-

lar to mean water column irradiances that exist in the Great Lakes during spring isothermal mixing. Mean water column irradiances in the Great Lakes ranged from 11 to 101 μmol quanta·m^{-2}·s^{-1} with a mean of 36 μmol quanta·m^{-2}·s^{-1}. Thus, nutrient–light interactions should occur in the Great Lakes during spring isothermal mixing.

The previously noted absence of nutrient limitation in Lakes Erie and Ontario during the spring period (Lean et al. 1983, 1987) and the evidence for strict light limitation during this period (Millard et al. 1996) may seem contradictory to our conclusions, but this apparent contradiction is easily explained. First, and most important, are the dramatic changes in the light climate that have occurred in the lower Great Lakes during the past two decades due to phosphorus load reductions and the impact of nonindigenous mussels (e.g., Holland 1993; Fahnenstiel et al. 1998). These large increases in light penetration would decrease the potential for light limitation and increase the potential for nutrient limitation and nutrient–light interactions. In the early 1970s, Thomson et al. (1974) reported k_{PAR} values from the spring mixing period of 0.57 and 0.34·m^{-1} in Lakes Erie and Ontario, respectively. In the late 1980s, k_{PAR} values from two stations in Lake Ontario during the April–May period were 0.26 and 0.32·m^{-1} (Millard et al. 1996). In 1993–1995, we reported a mean k_{PAR} value for stations in Lake Ontario of only 0.18·m^{-1}. This large increase in transparency in the 1990s produced an increase in the mean water column irradiance in Lake Ontario of 43–76% from the late 1980s and an 87% increase from the early 1970s. Assuming a k_{PAR} value of 0.29·m^{-1} for the late 1980s and 0.34·m^{-1} for the early 1970s (and similar physiological characteristics for phytoplankton communities), the increase in transparency in Lake Ontario would change the ratio $I_{k,g}$:$\bar{I}_{wc}$ from 0.6 in the early to mid-1990s to 1.0 in the late 1980s and to 1.1 in the early 1970s, respectively. Moreover, for photosynthesis, the ratio I_k:$\bar{I}_{wcd}$ would change from 1.3 in the 1990s to 2.2 in the 1980s and to 2.6 in the 1970s. Thus, at the time that Lean et al. (1983, 1987) and Millard et al. (1996) performed their experiments, the phytoplankton communities were likely light limited.

A second factor that needs consideration when comparing our results to those of Millard et al. (1996) are the differences between the photosynthetic and growth light saturation parameters (I_k versus $I_{k,g}$). Photosynthesis–irradiance relationships can be very different from growth–irradiance relationships, and it is not uncommon for growth to saturate at much lower irradiances than photosynthesis (Beardall and Morris 1976; Morris and Glover 1981). We reported a saturation irradiance for growth of 0.07 mol quanta·m^{-2}·h^{-1} (calculated for photoperiod only), whereas the threshold for photosynthesis was 0.16 mol quanta·m^{-2}·h^{-1}. These differences are important not only for the Great Lakes but also for all studies where photosynthesis–irradiance parameters are extrapolated to growth. The relationship between growth and photosynthesis will not be similar for all communities (Cullen 1990), and thus, extrapolating photosynthesis–irradiance parameters may produce erroneous conclusions.

Finally, nutrients appear to limit the maximum spring phytoplankton biomass across the Great Lakes. Carbon to chlorophyll ratios of spring phytoplankton and in situ chlorophyll concentrations suggest that light is not likely limiting biomass during late April – May (Parker et al. 1977; Fahnenstiel and Scavia 1987). However, prior to the development of this phytoplankton maximum, environmental variables that limit growth not only control the rate of biomass increase but potentially also biomass. In this study, we demonstrated that important rate processes such as growth and photosynthesis were controlled by nutrients and light. Thus, these same two factors are likely important factors controlling in situ biomass prior to the spring maximum. Temperature, acting through an interaction with nutrients, may also be important but clearly sets a range within which nutrient availability and light availability provide more proximate control.

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