

Final report to the St. Lawrence River Research and Education Fund

***Plankton Dynamics in a Large River System: Investigating Plankton Growth and Grazing Rates in the International Section of the St. Lawrence River***

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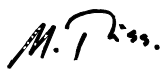
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**Students involved:** One graduate student (Derek E. Smith) assisted with the sampling along the river from June to December 2009. The student was responsible for assisting in conducting the dilution assays, data synthesis, and extra experimentation to confirm methodology. The student will be the junior co-author on any publication that comes from this report.

**Significance of this work:** The results from this study are the first growth and grazing rates ever produced for the International Section of the St. Lawrence River (ISSLR). The data base covers three ecologically important size fractions of phytoplankton (0.2-2  $\mu\text{m}$ , 2-20  $\mu\text{m}$ , and 20-153  $\mu\text{m}$ ), a 180 km section of the river from the headwaters at Lake Ontario to the power dam in Massena NY, and a seasonal aspect from late spring to early winter. In conjunction with a data set established earlier on plankton and limnological state variables (e.g. nutrient concentrations, phytoplankton and zooplankton community structure; Twiss et al. 2010), the data rate variables reported herein provides an important step to establishing a mathematical model of phytoplankton dynamics in the ISSLR. Such a model will provide a useful tool to predicting how changing water levels could impact plankton production, and accordingly fish production in this section of the river.

**Publications:** Since this work was completed in December 2009, there has been no opportunity to present these results, to date, at a research conference. It is anticipated that a manuscript will be submitted to a peer reviewed journal, e.g. *Journal of Great Lakes Research*, based on the results from this experiment, which are described briefly in this report.



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## Introduction

Recent investigation has provide a high resolution data set showing a pronounced decrease in phytoplankton biomass, inferred from decreases in chlorophyll-*a* (Chl-*a*) concentrations, in water of the St. Lawrence River following outflow from Lake Ontario (Twiss et al. 2010) despite an increase in nutrients, such as phosphorus, with water transit downstream. Results from that study propose that grazing is an important loss term that causes phytoplankton biomass to decrease. Grazing can occur in two ways in river water: 1) losses due to grazers (protozoan, metazoan and crustacean) present in the water column, and 2) losses due to phytoplankton contact with sessile grazers (benthic organisms present on the sediment and attached to submerged aquatic vegetation). The Twiss et al. (2010) study showed up to an order of magnitude decrease in crustacean zooplankton density from Lake Ontario to fluvial Lake St. Lawrence, approximately 180 km downstream; hence, losses due to crustacean zooplankton can be discounted. However, Basu et al. (2000) showed marked increases in rotifer density from Lake Ontario downstream in this section of the river; thus, a shift from crustacean to rotifers as the dominant grazers in the water column is possible. To test the impact of rotifers and protozoa on phytoplankton grazing we conducted 43 dilution assays in the International Section of the St. Lawrence River (Fig. 1) during 2009. Dilution assays (Landry and Hassett 1982) are designed to measure Chl-*a* specific growth and grazing rates in phytoplankton. Our objective was to conduct dilution assays in water sampled from throughout the International Section of the St. Lawrence River from late spring to winter in order to reveal how growth and grazing rates differ among seasons and different geomorphological reaches in the river. We test the hypothesis that there is no difference in growth and grazing rates as a function of season or location.

## Methods

Stations selected were those among the set used to assess the status of phytoplankton and nutrients in the International Section of the St. Lawrence River (Twiss et al. 2010), with the exception of November and December sampling, which took place from a pier at Waddington, NY. Sampling was conducted in a way to provide both spatial comparisons along the entire reach of the International Section of the St. Lawrence River as well as to show any temporal differences at a given station, with a focus on stations located in fluvial Lake St. Lawrence (Fig. 2). Water collection from one to three stations occurred between 06h:30 and 09h:30 (local time) on each sampling date. Water was collected from a depth of 0.5 m using either a clean food-grade polyethylene bucket attached to a nylon rope, or via an in hull pump onboard the RV *Lavinia*. Twenty liters of water from each station collected and passed through a screen (153- $\mu$ m mesh opening) and collected in opaque polyethylene containers. Water temperature was determined at each station and the average temperature (Fig. 3) on each sampling date used to set the incubation conditions (see below).

Dilution assays (Landry and Hassett 1982) were used to measure size-specific growth and grazing rates of phytoplankton. Upon return to the laboratory (maximum 2 h following sampling) river water was filtered

through a 0.2- $\mu\text{m}$  pore size cartridge filter (MilliPore Corp., Billerica, MA) to achieve a series of six dilutions (100%, 66%, 41%, 33%, 19% and 8% river water) in 1.215 L polycarbonate bottles. An enrichment of 30 nM  $\text{PO}_4\text{-P}$  was added to account for nutrient regeneration resulting from grazing (Landry and Hassett 1980) or viral and fungal induced lysis; periodically assays were conducted with a negative control in which 100% river water was incubated that had no added phosphorus.

Size-fractionated Chl-*a* concentrations (153-20  $\mu\text{m}$ , 20-2  $\mu\text{m}$ , 2-0.2  $\mu\text{m}$ ) were determined in triplicate at the beginning of the assays and a single size-fractionated measurement made in each bottle after approximately a 24-36 hour period. Bottles were incubated in a temperature and light-controlled incubator (Percival model 130BLL) set to ambient water temperature and day length. Bottles were arranged in a random order and then rearranged in random order after approximately 24 hours. Light was provided by seven fluorescent bulbs (4  $\times$  FT20T12-CW and 3  $\times$  F15T8-PL/AQ). Light was attenuated using neutral density screens to provide a photon flux field of 40  $\mu\text{mol}/\text{m}^2/\text{s}$ . Experiments conducted on x-y August 2009 were incubated in situ at a depth of 2.3 m by attachment to a pier on the east end of a small island; shading occurred from approximately 14h:00 onward.

Chl-*a*-specific growth rates ( $\mu$ ) were calculated in each dilution as follows:

$$\mu = (\ln[\text{Chl-}a]_{\text{end}} - \ln[\text{Chl-}a]_{\text{start}})/t \quad (1)$$

where  $\ln[\text{Chl-}a]$  is the natural logarithm of the Chl-*a* concentration in a given size fraction at the beginning or end of the incubation period, and  $t$  the duration of the incubation. The intrinsic growth rate was determined as the intercept with the ordinate of a linear least squares regression of specific growth rate in each dilution versus the respective dilution factor; grazing rate ( $g$ ) was determined as the slope ( $\times -1$ ) of this regression (Landry and Hassett 1982).

The following rules were applied to accepting estimated rates. In cases where there was a regression that had a significance ( $P$ )  $>0.10$ , the growth rates in all dilutions was averaged rather than using the intercept to predict the specific growth rate (per Chen et al. 2009) and no grazing rate was determined, and grazing rates were considered to be undeterminable in assays that resulted in a positive slope (theoretically impossible), in which case growth rates in all dilutions were averaged.

Experiments conducted in May and June used a 0.45-  $\mu\text{m}$  pore size high capacity, groundwater capsule filter (MilliPore Corp., Billerica, MA) to produce water for dilutions. A test of this water contained Chl-*a* (approx. 1% of the total) that increased in concentration following P enrichment and incubation over 3 days. Thus, a series of experiments were conducted to examine the filtration efficiency of the 0.45- $\mu\text{m}$  cartridge filters compared to the 0.2- $\mu\text{m}$  cartridge filters, which included re-filtration of 0.45- $\mu\text{m}$  filtrate and comparison of growth and grazing rates using river water  $<0.45\text{-}\mu\text{m}$ , re-filtered (0.45- $\mu\text{m}$  pre-size filter), and river water  $<0.2\text{-}\mu\text{m}$  as diluents.

## Results

*Phytoplankton biomass and size structure:* A summary representation of total Chl-*a* (a proxy of phytoplankton biomass) and size fractionated Chl-*a* shows a decrease in phytoplankton biomass upon entry of lake Ontario waters into the St. Lawrence River (Fig. 4A). The proportion of picoplanktonic phytoplankton (0.2-2  $\mu\text{m}$ ) increased with distance water flowed downstream (Fig. 4B), with picoplankton comprising approximately 60% of the total Chl-*a*, regardless of season, by the time water enters fluvial Lake St. Lawrence.

*Phytoplankton growth and grazing rates:* Growth and grazing rates expressed as a function of temperature reveal closely matched rates of production (Fig. 5A) and consumption (Fig. 5B) of phytoplankton (Chl-*a*) across the range of temperatures assayed (Fig. 5) with a few notable exceptions. There was an apparent disconnect between growth and grazing rates at river water temperatures greater than 20°C with growth rates increasing with increasing temperature (Fig. 5A) whereas grazing rates of phytoplankton did not apparently increase with increasing temperature (Fig. 5B). In addition, a grazing rate determined at a water temperature of 3°C was  $2.90 \cdot \text{d}^{-1}$ , which coincided with a growth rate of  $0.87 \cdot \text{d}^{-1}$ , values among the highest but at the lowest water temperature sampled.

Size fractionated growth and grazing rates show the microplankton and picoplankton size fractions had slight increases in growth rates over grazing rates with increasing temperature, whereas the nanoplankton remained balanced between these estimates of production and consumption rates (Fig. 6).

*Methodological considerations:* There was no discernible difference in growth and grazing rates measured if river water <0.4  $\mu\text{m}$  or river water <0.2  $\mu\text{m}$  was used as diluent (data not shown). Re-filtering water that had passed through a 0.45- $\mu\text{m}$  pore size cartridge filter reduced Chl-*a* that passed through the filter but using this water for dilution assays caused spurious results – for some unknown reason. Using All dilution assays were enriched with 30 nM phosphate. In some cases, additional controls (no dilution water added) had no phosphorus added in order to test if nutrient enrichment enhanced growth or grazing; the results from the control bottles showed no impact of phosphorus over the course of the dilution assay (data not shown).

## Discussion

The data collected support the hypothesis that there is no difference in growth and grazing rates in the river with respect to spatial but not temporal considerations. An examination on the database shows no apparent trend with location in the river but temperature has a marked effect on growth and grazing rates, as described above. Based on examples from published literature, Peters (1994) showed that grazing rates increase exponentially with increasing temperature, in contrast to what was observed here for water sampled from the International Section of the St. Lawrence River. One possible cause for this discrepancy is the fact that only phytoplankton grazing was followed. Increasing water temperatures in rivers will increase the growth rate of bacteria (White et al. 1991). Increasing availability of bacteria will relieve grazing stresses on phytoplankton by microzooplankton grazers as they switch to more abundant prey. Accordingly, high grazing rates observed at

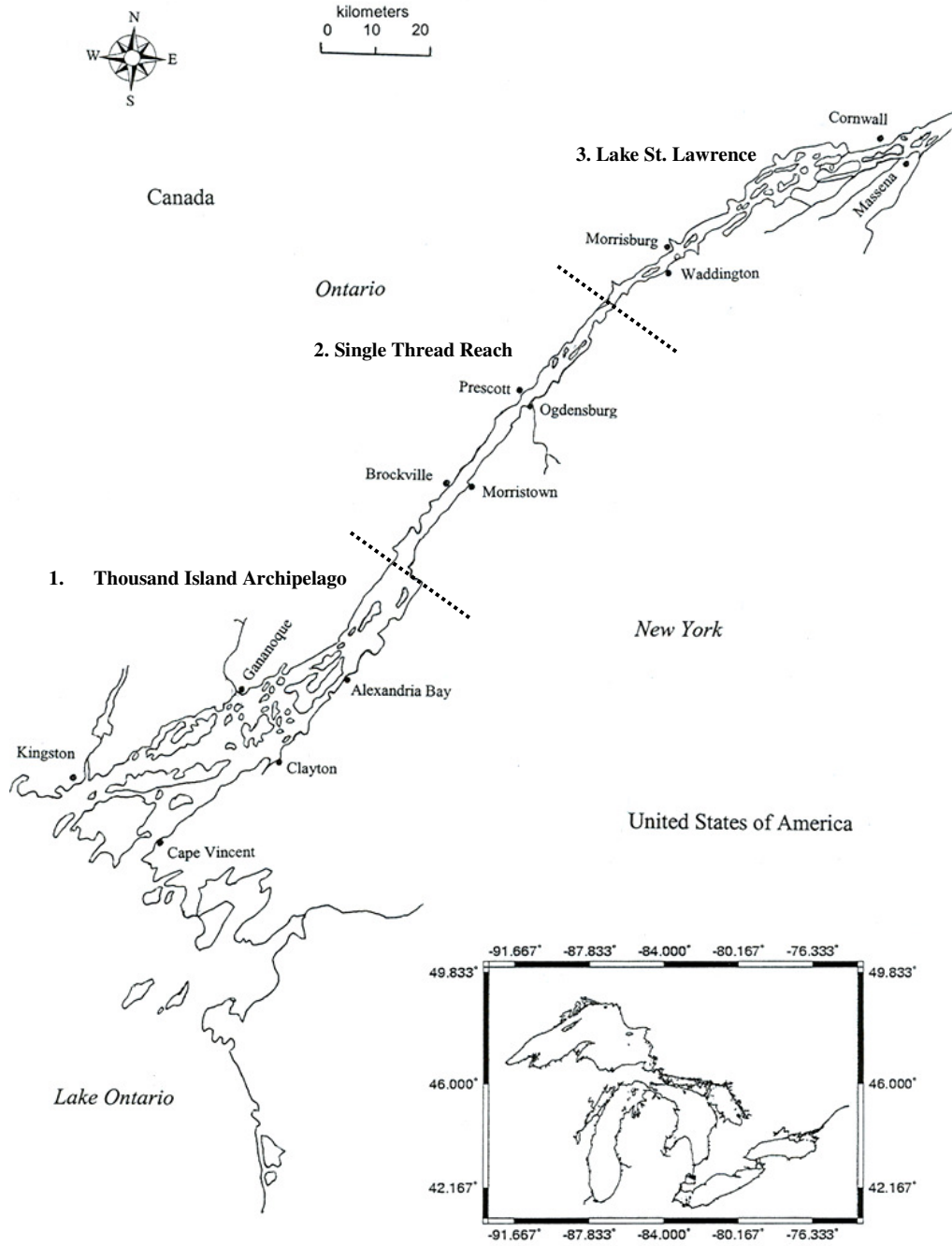
3°C may be related to expected low bacterial growth rates (White et al. 1991) and thus, greater observed grazing pressure on phytoplankton. The observed high growth rate of phytoplankton at low river water temperatures are consistent with rates observed during February 2008 and 2009 in Lake Erie using dilution assays (Twiss, M.R., unpublished data) and reflect the presence of a cryophilic phytoplankton community.

The tight coupling of growth and grazing rates, as well as times at which growth exceeds grazing, suggest that grazing due to benthic organisms (mussels, insect larvae, sessile rotifers and protists) are the primary source of phytoplankton loss along this stretch of the St. Lawrence River. Even during periods of warm river water temperature (> 20°C) where growth rates exceed grazing rates approximately  $0.5 \cdot d^{-1}$ , decreases in phytoplankton are observed in the river with transit from Lake Ontario and the expected bloom of phytoplankton does not materialize as predicted (Fig. 7).

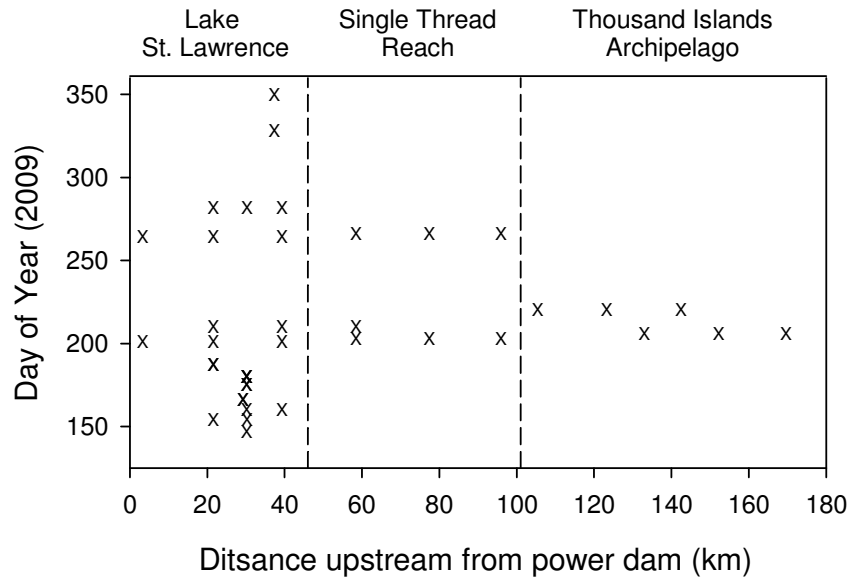
In conjunction with a data set established earlier on plankton and limnological state variables (e.g. nutrient concentrations, phytoplankton and zooplankton community structure; Twiss et al. 2010), the data rate variables reported herein provides an important step to establishing a sophisticated mathematical model of phytoplankton dynamics in the International Section of the St. Lawrence River. Such a model will provide a useful tool to predicting how changing water levels could impact plankton production, and accordingly fish production in this section of the river.

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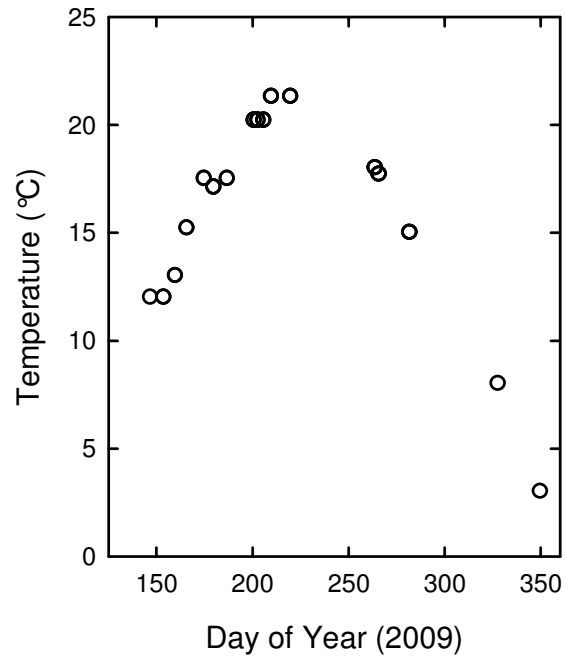
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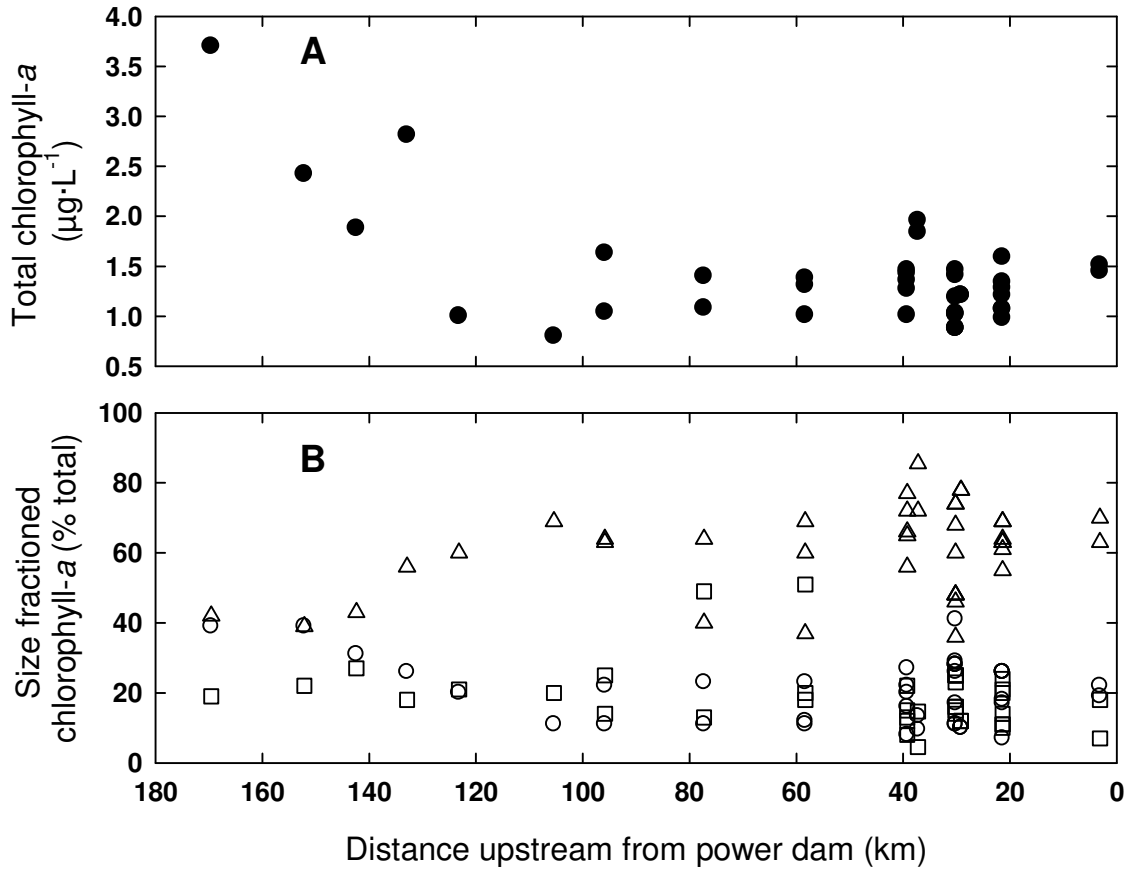
**Figure 1.** International Section of the St. Lawrence River showing the three geomorphic regions along this reach of the river after its outflow from Lake Ontario.



**Figure 2.** Sampling strategy showing the spatial extent of sampling over the reach of the International Section of the St. Lawrence River and temporal frequency of sampling in the river regions.

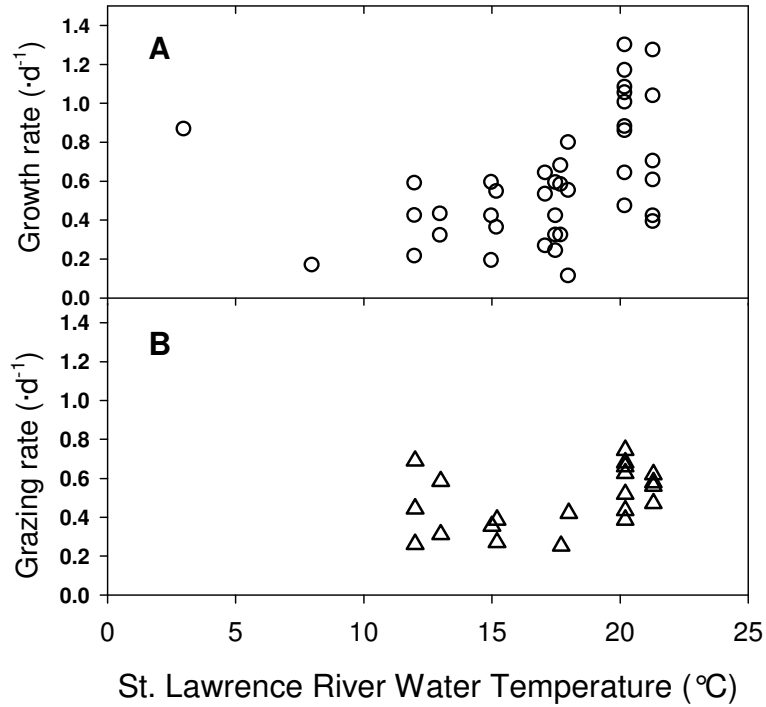


**Figure 3.** Ambient St. Lawrence River water temperature observed during the sampling period (May 27 to December 17, 2009).

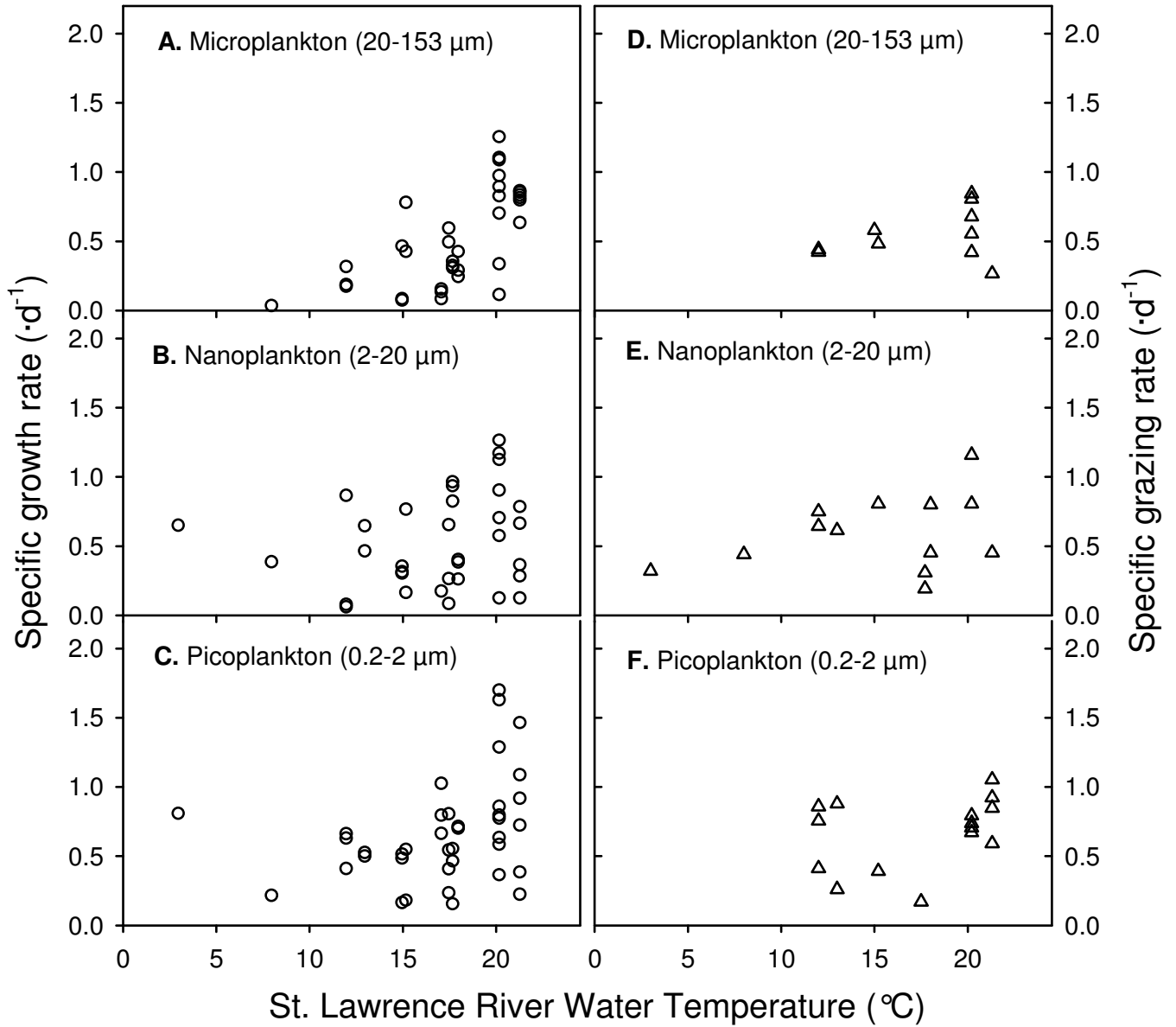


**Figure 4.** Phytoplankton biomass estimates in the International Section of the St. Lawrence River using chlorophyll-*a* as a surrogate of phytoplankton during May to December 2009 sampling period. A. Total chlorophyll-*a* (0.2-153  $\mu\text{m}$ ); B. *circles* microplankton (20-153  $\mu\text{m}$ ), *squares* nanoplankton (2-20  $\mu\text{m}$ ), *triangles* picoplankton (0.2-2  $\mu\text{m}$ ).

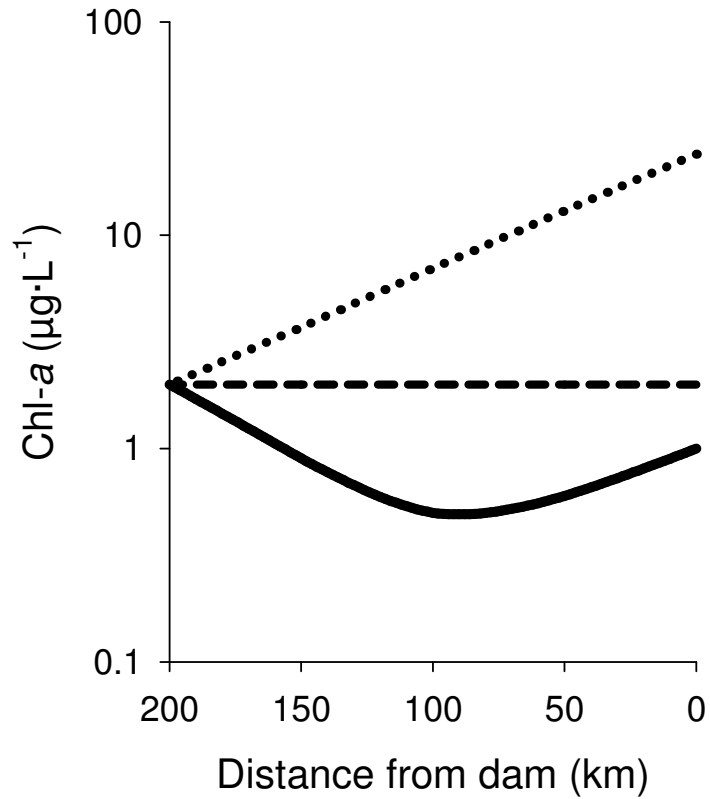




**Figure 5.** Estimated chlorophyll-*a*-specific growth and grazing rates of phytoplankton (>0.2-153  $\mu\text{m}$ ) in the International Section of the St. Lawrence River as a function of river water temperature. Note: not all dilution assays used to derive rates supplied data that could provide a valid estimate of grazing rate. A grazing rate determined at a water temperature of 3°C was  $2.90 \cdot d^{-1}$  (datum not plotted on panel B).



**Figure 6.** Chlorophyll-a-specific size fractionated rates of phytoplankton growth (A-C) and grazing (D-F) in the International Section of the St. Lawrence River as a function of river water temperature.



**Figure 7.** A simple model of phytoplankton (Chl-*a*) concentrations from Lake Ontario to the Moses-Saunders Power Dam. *Dotted line*, growth rate > grazing rate by  $0.5\cdot\text{d}^{-1}$ ; *dashed line*, growth rate = grazing rate; and *solid line*, observed trend in chl-*a* concentrations in this section of the St. Lawrence River (per Twiss et al. 2010).