

# The effects of nutrient enrichment and insecticide application on planktonic and benthic algal biomass in Delta Marsh

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## Introduction

Prairie wetlands have been extensively altered, both physically and chemically, due to the impacts of intensive agriculture. Many wetlands have been drained and cultivated, while those that remain intact are subject to the chemical applications used to boost crop productivity: fertilizers and pesticides. Wetlands are often viewed as natural filters for such anthropogenic discharges. Therefore, it is important to develop an understanding of the way in which these ecosystems respond to such inputs. A study to investigate the impacts of nutrient addition and insecticide application on the algal communities in Delta Marsh, Manitoba was initiated in 1997.

Two views of the controls on aquatic community structure and production predominate in the ecological literature. The "top-down" theory proposes that animal predation regulates the structure of trophic levels below them. Therefore, suppression of algal production occurs when herbivorous zooplankton is abundant (e.g., Carpenter *et al.*, 1985). On the other hand, the "bottom-up" theory proposes that ecosystem production is controlled by the availability of nutrients and resulting primary production (e.g., Schindler, 1978).

The objective of this study was to elucidate the effects of inorganic nutrient addition (N and P) or insecticide application (chlorpyrifos, an arthropod-specific organophosphorus pesticide) on the algal communities using large, fishless enclosures in Delta Marsh. We hypothesized that control of algal biomass would be released by these treatments; insecticide application would remove grazers (zooplankton and other microinvertebrates) and nutrient addition would stimulate algal growth directly.

## Materials and Methods

### *Experimental Treatments*

Twelve floating enclosures were deployed in the center of Blind Channel on 27 May 1997. Each enclosure (5 m x 5 m) consisted of translucent plastic curtain extended from a floating wooden frame into the

sediments, and therefore was isolated from the surrounding water. Water depth in each enclosure was approximately 100 cm throughout the study period (ending 29 August). Minnow traps were used to exclude fish from the enclosures. This was done in an attempt to prevent the occurrence of unnaturally high fish densities, which would exert strong top-down effects. The experiment began on 23 June (week one) after a three-week pre-treatment period to allow for recovery from the disturbance caused by installation of the enclosures.

Three replicated treatments were studied: (1) procedural control (no manipulation), (2) inorganic N and P addition, and (3) chlorpyrifos addition. Nine enclosures were used in this project, allowing for three replicate enclosures for each treatment. Enclosures 4, 9, and 12 (numbered clockwise from the northwest corner of the complex of 12 enclosures) were controls. N and P were added to enclosures 3, 5, and 10 three times weekly beginning on 23 June and ending 27 August (week 10). These nutrients were dissolved in 1 L of carbon-filtered water in the laboratory, transported to the enclosures, mixed with approximately 10 L of enclosure water, and sprinkled evenly over the surface of the enclosure. Each enclosure received a cumulative load of 23.9 g/m<sup>2</sup> N and 3.2 g/m<sup>2</sup> P. The insecticide Lorsban™ 4E (an emulsifiable liquid formulation made by DowElanco Ltd.) was added to enclosures 1, 6, and 8 once on 14 July to achieve a nominal chlorpyrifos concentration of 10 µg/L. A known amount of the insecticide was mixed in 250 mL of distilled water in the laboratory, then mixed with about 20 L of enclosure water and sprinkled over the surface of each treated enclosure. See Zrum and Hann (1998) for details on chlorpyrifos sampling and analysis.

### *Sampling and analyses*

Water samples were collected twice weekly, beginning on 10 June (2 weeks prior to manipulation), from each enclosure and analyzed for pH, NH<sub>3</sub>-N, NO<sub>3</sub>+NO<sub>2</sub>-N, soluble reactive P (SRP), and titratable alkalinity (Stainton *et al.* 1977, APHA 1992). Temperature and oxygen profiles with depth in each enclosure were measured weekly starting 19 June.

Phytoplankton and epiphyton biomass was estimated using chlorophyll measurements and submersed macrophyte biomass was determined using dry weight.

Two random samples of submersed macrophytes, their associated epiphytic algae and phytophilous invertebrates were taken weekly from each enclosure using a Downing box sampler (Downing 1986). This sampling regime began later than that of other parameters (9 July; week 3). Prior to this time, macrophytes were observed to be growing actively but they were not large enough to be sampled quantitatively. Epiphytes were removed from the macrophyte samples by vigorous shaking in carbon filtered water. Algae in the wash solution was collected on Whatman GF/C filters under vacuum. The filters were frozen for a minimum of 24 hours to lyse algal membranes, then placed in 90% methanol and stored in the dark for 24 hours to extract chlorophyll pigments. Chlorophyll concentration was measured spectrophotometrically and calculated using the formulae of Marker *et al.* (1980). At occasional intervals, additional epiphyton samples were retained for determination of dry weight. A sample of the algal suspension was collected onto pre-tared GF/C filters which were then dried for 24 hours at 100°C and reweighed.

Three random phytoplankton samples were taken weekly from each enclosure beginning on 4 June (3 weeks prior to manipulation) using a cylindrical integrated water column sampler. Each 4 L sample was passed through a 53 µm mesh net to remove zooplankton (Zrum and Hann 1998) then filtered (Whatman GF/C) to collect algae. The filters were analyzed for chlorophyll concentration (µg/L) and dry weight (mg/L) as for epiphyton samples.

Macrophytes were sampled monthly from each enclosure using a 0.55 m diameter cylinder. All macrophyte biomass in the cylinder was harvested, dried at 105°C for 24 hours, and weighed. These data were used to interpolate macrophyte dry weight for each epiphyton sampling date so that epiphyton data could be expressed per unit of macrophyte dry weight (µg/g and mg/g for algal chlorophyll and dry weight, respectively) and, using areal measurements of macrophyte abundance, per unit of marsh bottom area (mg/m<sup>2</sup> for chlorophyll data).

## Results and Discussion

Levels of NO<sub>3</sub>-N, NH<sub>3</sub>-N, and SRP in control and insecticide-treated enclosures remained low throughout the study (Fig. 1). Nutrient addition enclosures showed a pattern of increasing NO<sub>3</sub>-N and SRP concentrations throughout the summer, whereas NH<sub>3</sub>-N levels increased only slightly in July and August, probably due to

reduction of added nitrate. By the end of August, the NO<sub>3</sub>-N concentration in the water column represented < 4% of the amount added cumulatively during the experiment; the SRP concentration at the same time represented 47.5% of the cumulative input load. Therefore, ambient N and P levels diverged over time from those added to the enclosures because the input weight ratio was 7.5 whereas the ratio of N to P at the end of the experiment was about 0.6. Similar results have been found in past experiments, suggesting that the marsh has a greater assimilative or metabolic capacity for inorganic N than for inorganic P. However, we have no information on the magnitude of N volatilization from these enclosures which may explain some of the unaccounted N in this system.

Phytoplankton abundance in control enclosures remained low (< 20 mg/m<sup>2</sup>; mean 6.5 mg/m<sup>2</sup>) throughout the study (Fig. 2). Chlorophyll levels in nutrient-treated enclosures increased briefly in late June and early July, after which they dropped off to those similar to controls (overall mean 18 mg/m<sup>2</sup>). Insecticide treatment did not have a long-lasting effect on phytoplankton abundance, as chlorophyll levels increased slightly above control levels within a week of insecticide application (14 July), and dropped off again for the remainder of the study (overall mean 12 mg/m<sup>2</sup>). Water column grazers such as cladocerans and copepods would have been eliminated immediately after exposure to the pesticide, allowing phytoplankton to proliferate during the following week. The short duration of this response by the phytoplankton can probably be explained by the resurgence of calanoid copepods and rotifers (Zrum and Hann 1998). Calanoid copepods, known to filter phytoplankton as a food source, are thought to be more tolerant to chlorpyrifos than other filter feeders (specifically, cladocerans) which may explain their dominance in the weeks following chlorpyrifos addition, and therefore, the reduction in chlorophyll to pre-insecticide levels.

Epiphyton abundance did not exhibit a clear trend with time (Fig. 2). Epiphyton abundance was low in control enclosures throughout the experiment, hovering around 50 mg/m<sup>2</sup> (but still about 20-fold higher than the biomass of phytoplankton at corresponding times). Insecticide application increased epiphyton biomass slightly, but only in the last three weeks of the experiment (60-203 mg/m<sup>2</sup>). With a few exceptions, epiphyton biomass in nutrient-treated enclosures was generally higher than in controls (overall mean 108 mg/m<sup>2</sup>).

The relationship of epiphyton chlorophyll to dry weight was linear and positive ( $r^2 = 0.70$ ; Fig. 3) although the corresponding relationship for phytoplankton was more variable. This may indicate

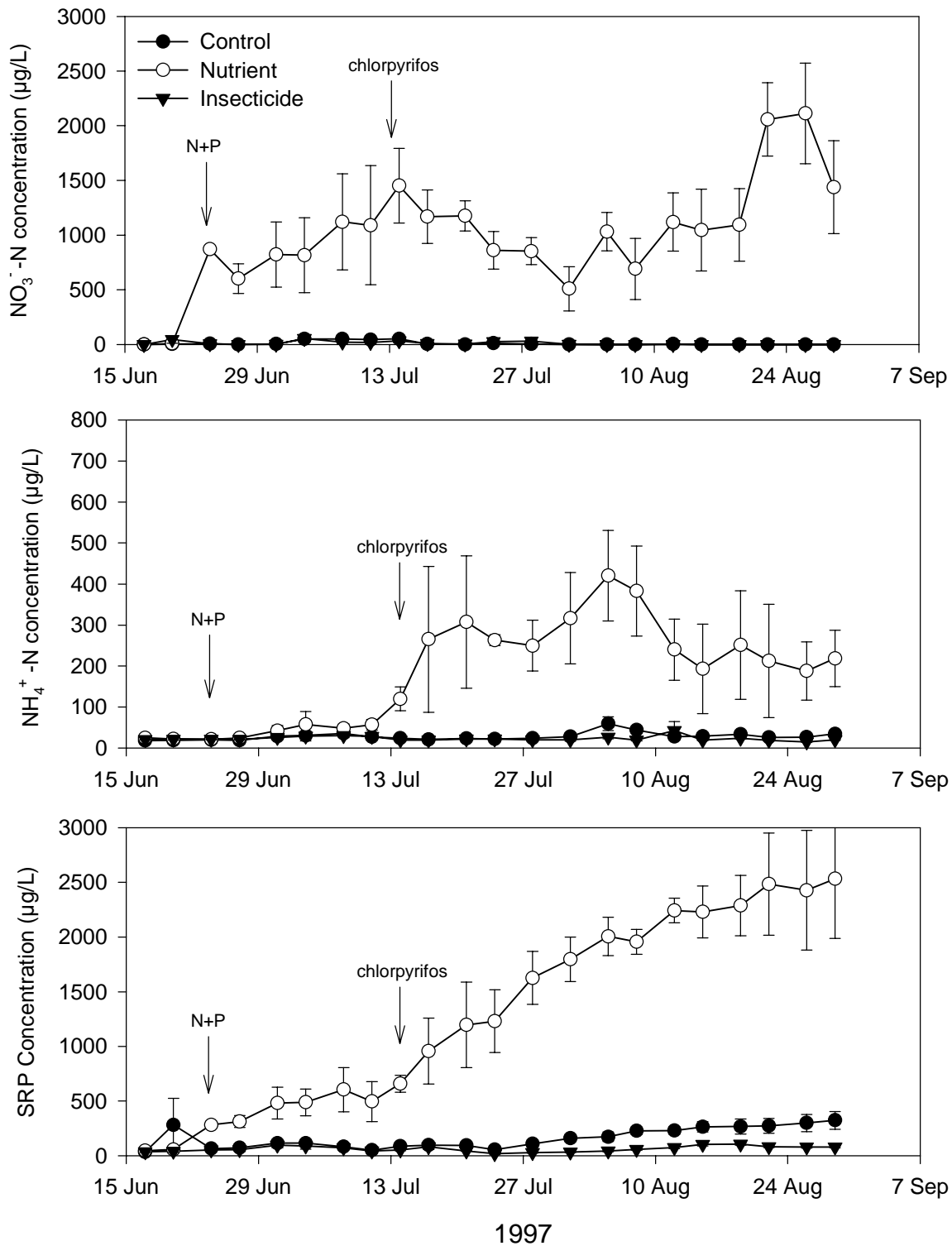


Figure 1. Nitrate+nitrite-N, ammonium-N, and soluble reactive P concentrations (mean ± SE, n=3) in control, nutrient addition, and insecticide addition treatments in marsh enclosures over an 11-week experimental period. Thrice-weekly nutrient additions began on 23 June and insecticide was added once on 14 July.

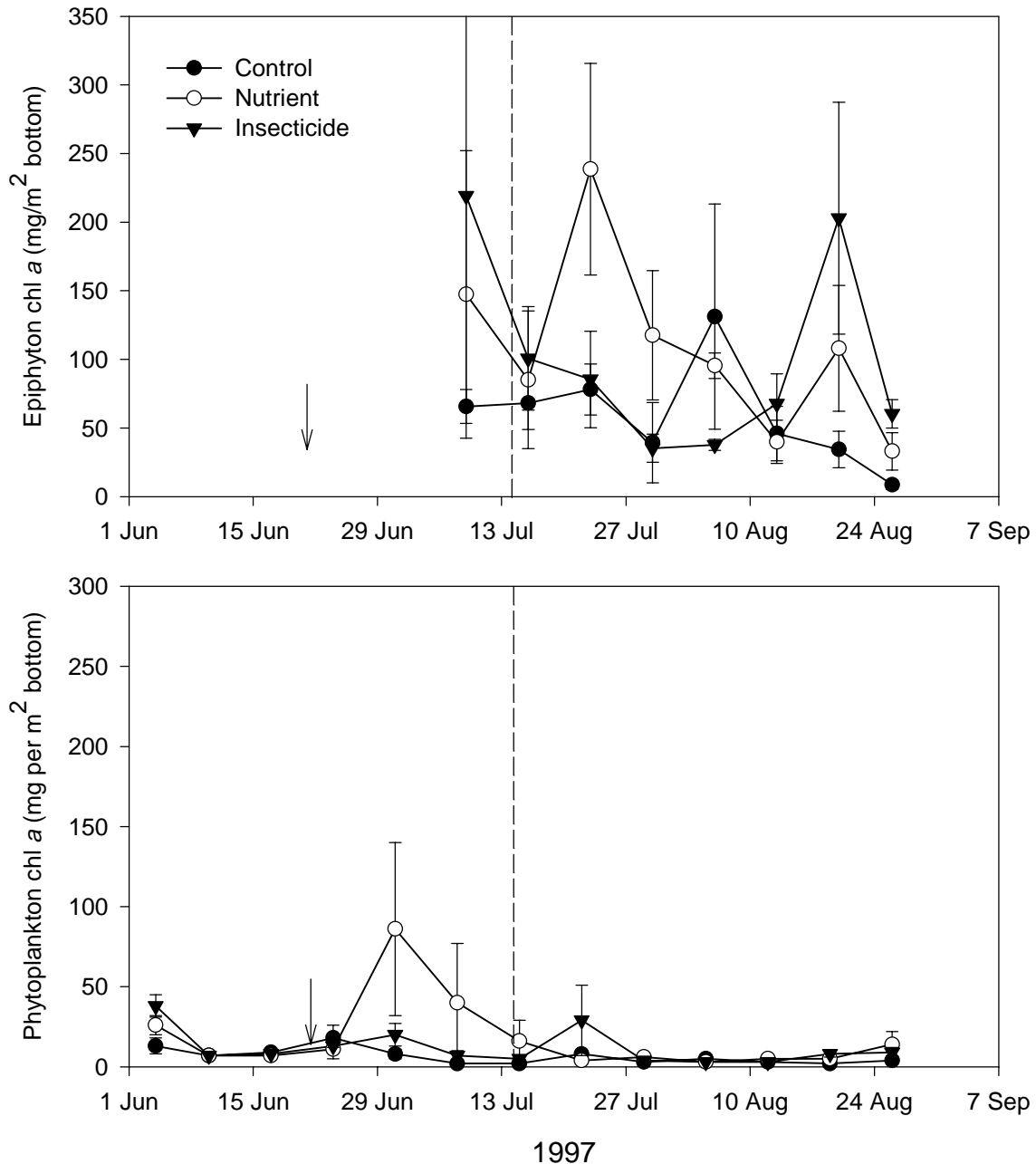


Figure 2. Phytoplankton and epiphyton chlorophyll per unit bottom area ( $\text{mg}/\text{m}^2 \pm \text{SE}$ ,  $n=3$ ) in control, nutrient addition, and insecticide addition treatments in marsh enclosures over an 11-week experimental period. Thrice-weekly nutrient additions began on 23 June and insecticide was added once on 14 July.

that plankton samples were more likely to contain suspended materials other than algae (e.g., suspended sediments) whereas epiphyton samples, having had zoobenthos removed, were largely comprised of algae. The ratio of chlorophyll to dry weight, a useful metric in converting algal chlorophyll data into units comparable to those of other wetland plants (dry weight), was variable for both datasets. The median value was 0.23% for phytoplankton and 0.24% for epiphyton but

the means were higher (Fig. 3). However, these values agreed closely with those calculated from other data collected at Delta Marsh (mean = 0.25%, Goldsborough, unpublished).

Macrophyte dry weight was low in the spring, increased until late August then decreased (Fig. 4). The overlap of error bars between treatments throughout the study indicates that no treatment affected macrophyte biomass significantly. Peak biomass was about 200 g/

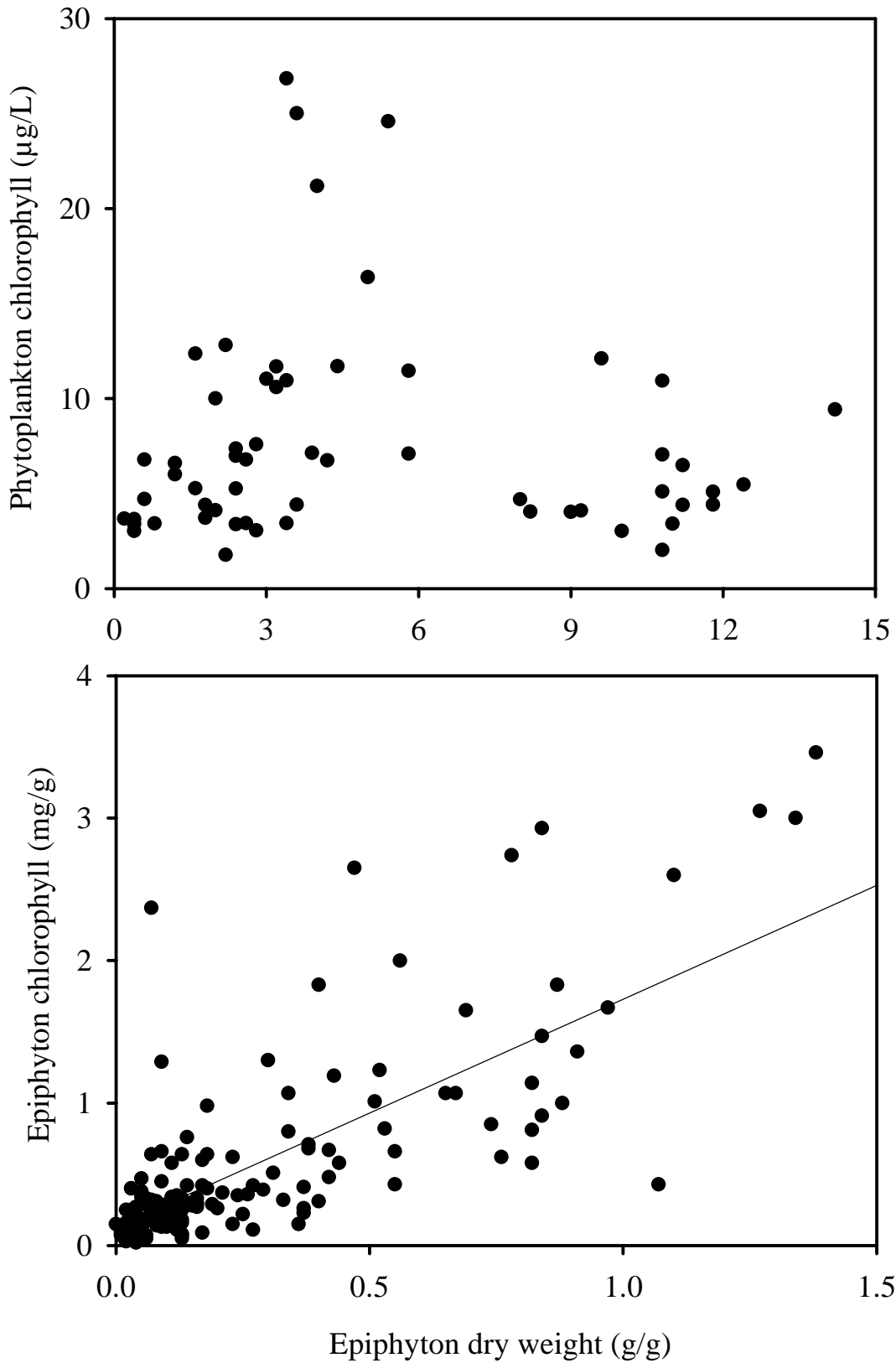


Figure 3. Relationship between the dry weight and total chlorophyll of phytoplankton (top) and epiphyton (bottom) collected during the experiment in 1997. The  $r^2$  values for the two relationships were 0.70 and 0.01, respectively. The mean and median values were 0.36 and 0.23 for phytoplankton, and 0.36 and 0.24 for epiphyton.

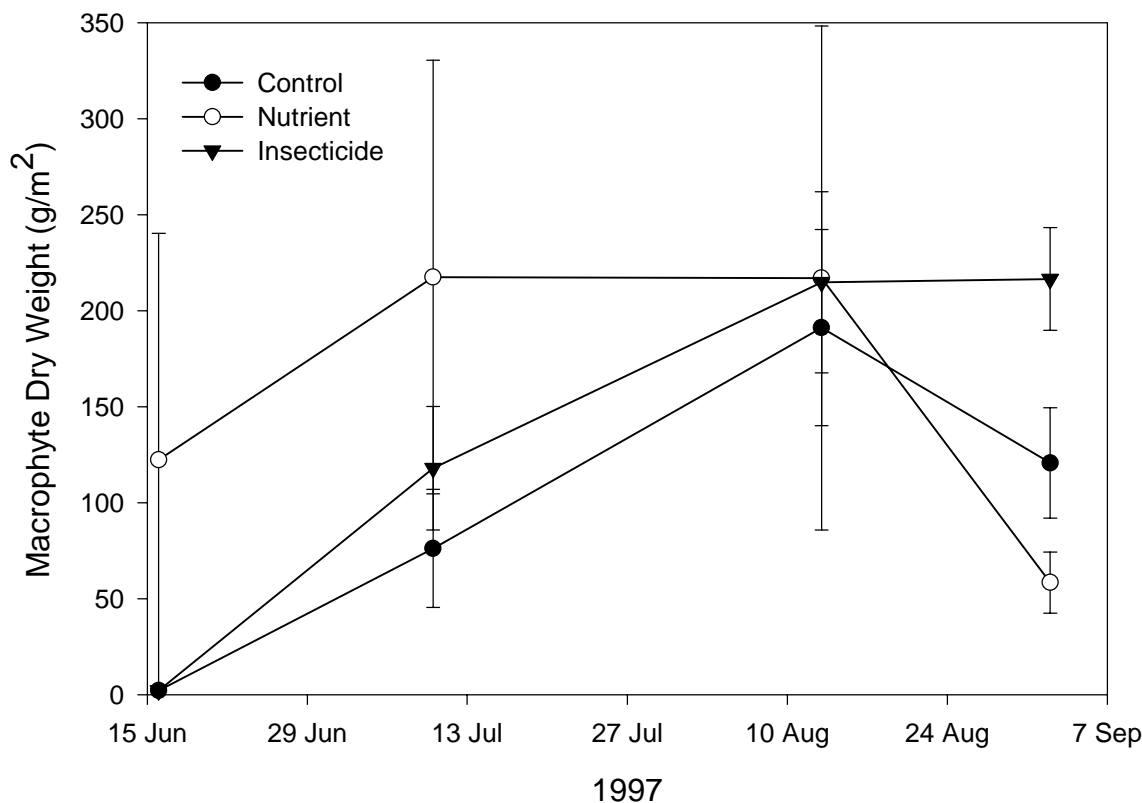


Figure 3. Submersed macrophyte biomass ( $\text{g/m}^2 \pm \text{SE}$ ,  $n=3$ ) in control, nutrient addition, and insecticide addition treatments in marsh enclosures over an 11-week experimental period.

$\text{m}^2$  in all cases but variation between replicate enclosures was extremely high, with patches of individual species occurring irregularly in some enclosures but not others. Submersed macrophytes clearly contributed the majority of total primary production (algal plus plant biomass) during the peak period of August (Table 1) although the contribution by algae increased slightly with nutrient or insecticide addition.

Planktonic and epiphytic algae responded positively to inorganic N and P enrichment of the water column but their increases in biomass, particularly those for phytoplankton, were generally short-lived. Insecticide application reduced the abundance of water column grazers for a short time (2 weeks; Zrum and Hann 1998), relieving grazing pressure and resulting in a transient phytoplankton bloom. Collectively, these results indicate that nutrient supply and herbivory are both important controls on algal production in Delta Marsh. However, we were surprised that the algal response to the manipulations was less marked than we had expected based on the high level of nutrient input (cf. McDougal *et al.* 1997) and the expected severity of invertebrate inhibition. It would be interesting to see whether the increase in algal abundance would be larger and more prolonged with a combined treatment of inorganic

nutrients and insecticide. A follow-up experiment, including this combined treatment, is planned for the 1998 field season.

	Control	Nutrients	Insecticide
Phytoplankton	1	3	2
Epiphyton	22	28	37
Macrophytes	156	138	216
% algal	13	18	15

nutrients and insecticide. A follow-up experiment, including this combined treatment, is planned for the 1998 field season.

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