Estuarine phytoplankton group-specific responses to sublethal concentrations of the agricultural herbicide, atrazine

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Abstract

Atrazine is a common agricultural herbicide that is readily transported into estuaries through surface water runoff. In this study, we determined the short-term (24-48 h) sublethal effects of atrazine on estuarine phytoplankton biomass and community composition. Phytoplankton group-specific responses to atrazine exposure (25 µg l⁻¹) were measured using natural water samples collected from two locations in Galveston Bay, Texas. Addition bioassays, coupled with HPLC pigment analysis, were used to quantify changes in the relative abundances of algal groups. For all algal groups except prasinophytes, the addition of atrazine in combination with nitrate was not significantly different from nitrate additions alone. These results suggest no significant negative effect of atrazine on phytoplankton under the specified environmental conditions for the bioassays. Although low concentrations of atrazine may have minimal impacts on phytoplankton, herbicide loadings need to be further characterized before generalizations can be applied to estuarine and coastal ecosystems.

Keywords: Atrazine; Texas; Phytoplankton; Estuary; Gulf of Mexico; Nutrient; Agriculture; Herbicide

1. Introduction

Estuarine phytoplankton are exposed to a variety of anthropogenic contaminants that may have negative impacts on both biomass and community composition. Herbicides, which are often found in rivers draining agricultural watersheds, may affect estuarine phytoplankton communities. Triazine compounds, including atrazine (2-chloro-4-ethylamino-6-isopropylamino-s-triazine), are some of the most commonly used agricultural herbicides in the United States (Moody and Goolsby, 1993; Solomon et al., 1996). The relatively high solubility of atrazine (33 µg ml⁻¹ at 22 °C) facilitates export from agricultural fields to receiving waters through surface runoff and sediment transport (Richard et al., 1975; Frank et al., 1979; Goolsby et al., 1991; Goolsby and Battaglin, 1993; Goolsby et al., 1993; Moody and Goolsby, 1993). Widespread use of atrazine promotes this herbicide as a common contaminant in streams, rivers, and lakes (Solomon et al., 1996; Clark et al., 1999). In microcosm studies, the reported environmental half-lives of atrazine range from 30 to 120 days in aqueous solutions exposed to natural conditions (Cunningham et al., 1984; Glotfelty et al., 1984; Kemp et al., 1985). Biodegradation is negligible due to the s-triazine ring, which is resistant to microbial attack (Howard, 1991). Estuaries can accumulate atrazine from both acute and chronic loading events (Jones et al., 1982; Glotfelty et al., 1984; Isensee, 1987; Kemp et al., 1985). The long water residence time in estuaries coupled with high atrazine loading rates may result in prolonged exposure of phytoplankton communities to atrazine. Although concentrations may not be high enough to cause direct mortality of algal species, they may impair phytoplankton photosynthesis and growth over ecologically-relevant time scales (hours to days) (Millie and Hersh, 1987; Hersh and Crumpton, 1989; Hoagland et al., 1996; Tang et al., 1997; Carder and Hoagland, 1998; Fairchild et al., 1998).

Galveston Bay, Texas is the second largest estuary in the Gulf of Mexico region. The watershed for this estuary has a drainage area of 46,100 km² and includes urban, industrial, and agricultural land uses. Herbicides (particularly triazines) are routinely applied to agricultural lands within the drainage basin during spring planting (Brown, 1996; Baumann and Ketchersid, 1998).
In a recent study, atrazine, metolachlor, and molinate were detected in more than 75% of the 20 samples collected in the lower segment of the Trinity River (Brown, 1996), which is the primary source of freshwater input into Galveston Bay. The maximum concentration of atrazine (4.0 \(\mu g\) l\(^{-1}\)) occurred in the spring (Brown, 1996). In a survey of the entire Trinity River Basin, atrazine was the most commonly detected herbicide in both winter and spring sampling periods (USGS, 1998). Concentrations were as high as 4.0 \(\mu g\) l\(^{-1}\) in the winter and 4.2 \(\mu g\) l\(^{-1}\) in the spring. However, these values, which are based on single point-in-time samples, may not reflect the true variability in concentrations, episodic pulses, or potential loading rates into Galveston Bay. Given the previous measurements of 4 \(\mu g\) atrazine l\(^{-1}\) in the waters entering Galveston Bay, the assumption that phytoplankton in the bay are exposed to concentrations less than 4 \(\mu g\) l\(^{-1}\) is reasonable. Therefore assessments of phytoplankton responses to atrazine concentrations greater than 4 \(\mu g\) l\(^{-1}\) would offer insights into the potential responses of the phytoplankton community to increased atrazine concentrations in Galveston Bay. In addition, an increase in atrazine concentrations would likely accompany an increase in agricultural fertilizer (particularly nitrate and phosphate) due to the interrelationship between fertilizer use, herbicide applications, and runoff. The widespread use of atrazine in this watershed in combination with spring rains, chronic leaching, and long half-life of the compound provides a clear justification for examining the potential impacts of atrazine runoff in this estuary.

Estuarine phytoplankton communities are composed of diverse species assemblages. The species-specific physiological responses of phytoplankton to even trace concentrations of atrazine range from impaired physiological function to rapid cell mortality (Solomon et al., 1996). Due to the rapid growth rates of estuarine phytoplankton (\(\approx 0.5-1.0\) d\(^{-1}\)), the physiological effects of atrazine will likely be expressed as alterations in the relative abundance of algal groups over time scales from hours to days. Atrazine concentrations as low as 100 \(\mu g\) l\(^{-1}\) may be sufficient to impair photosynthesis and induce physiological stress in some phytoplankton taxa (Solomon et al., 1996). Previous studies using monocultures under controlled laboratory conditions have established toxicities, but few studies have attempted an assessment using natural communities incubated in situ light and temperature.

Differential species responses and susceptibilities may have profound implications for phytoplankton community structure and dynamics in Galveston Bay. If differential responses and/or sensitivity occurs in a large enough proportion of an assemblage, noticeable changes in phytoplankton community structure and temporal periodicity within Galveston Bay would be expected. Therefore, we hypothesized that inputs of sublethal concentrations of agricultural herbicides into Galveston Bay have a significant impact on phytoplankton community composition and growth. The primary objective of this research was to determine the short-term (24-48 h) sublethal effects of realistic concentrations of atrazine on estuarine phytoplankton community structure. Although longer time scales of exposure to atrazine may be necessary to elicit a change in community composition, short-term bioassays provide insights into the immediate effects of atrazine on algal groups within the community.

2. Materials and methods

2.1. Study site

Galveston Bay, Texas is a shallow (~2 m depth) estuary that encompasses 1554 km\(^2\) of water surrounded by 526 km\(^2\) of marshland. Freshwater drainage from the Trinity (83%) and San Jacinto (8%) Rivers transports terrestrial organic and inorganic materials with moderate to high dissolved organic matter (5-8 mg C l\(^{-1}\)) and suspended particulate (4-200 mg l\(^{-1}\)) concentrations into the Bay (Guo and Santschi, 1997). The tidal range in the bay averages 40 cm, is primarily diurnal, and fosters the long hydraulic residence time of the estuary (40-88 days) (Santschi, 1995). Winds are more important than tides for circulation in Galveston Bay.

The estuarine phytoplankton community in Galveston Bay is diverse and varies with season and nutrient inputs. The most common algal groups in Galveston Bay are diatoms, cyanobacteria, chrysophytes, and cryptophytes (Sheridan et al., 1988; Ornólsdóttir et al., personal observation). Dinoflagellates, chlorophytes, and euglenoids are occasionally abundant, but on an annual basis are minor components in the phytoplankton community. Freshwater and nutrient inputs from the Trinity River extend well into Trinity Bay and Galveston Bay, especially during periods of high river discharge in the spring (Santschi, 1997). Nitrate concentrations are inversely correlated with salinity and benthic regeneration of phosphorus leads to a phosphate maximum in late summer (Santschi, 1995; Twilley et al., 1999).

In May 1999, we instituted a biweekly sampling program to collect data on water quality parameters, nutrient concentrations, and phytoplankton dynamics at seven locations in Galveston and Trinity Bays. Twelve nutrient (\(\text{NO}_3^-\), \(\text{PO}_4^{3-}\)) addition bioassays were conducted from May 1999 to July 2000 and all indicated that the phytoplankton community is consistently N-limited (Ornólsdóttir, 2002). All major algal groups of the community showed significant increases in biomass in response to the addition of \(\text{NO}_3^-\) (10 \(\mu M\) final concentration), sometimes resulting in a doubling in biomass (relative to controls) over a 24 h incubation period. Evidence of phosphate or silicate limitation was not
detected. In terms of size distributions, the picoplankton (<2 µm), nanoplanckton (2–20 µm), and microplankton (20–200 µm) fractions constituted 18%, 50%, and 32% of the total community biomass, respectively. All size ranges had representatives from most of the major algal groups and the nanoplanckton had the highest diversity.

Bioassays were used to measure initial phytoplankton responses to atrazine exposure. Water for the bioassays was collected on 11 March and 13 March 2000 from a depth of 0.5 m at two locations in Galveston Bay (Fig. 1). In the first bioassay (Bioassay 1), water was dispensed into eight flasks (2.5 l PTFE) and nitrate (10 µM final concentration as KNO$_3$) and phosphate (3 µM P final concentration as KH$_2$PO$_4$) was added to all containers. Atrazine was added (25 µg l$^{-1}$, 0.116 µmol l$^{-1}$ final concentration) to four of the flasks. There were four replicates for each factor level (N + P and N + P + atrazine). In the second bioassay (Bioassay 2), we expanded the experimental treatments to assess the individual effects of nitrate (10 µM) and phosphate (3 µM) and a control (without nutrient additions). Each factor level (control, N only, P only, N + P, and N + P + atrazine) consisted of three replicates in this experiment. The flasks were incubated in the boat basin at Texas A&M University at Galveston to simulate in situ temperatures, sunlight, and photoperiod for Galveston Bay.

Two layers of fiberglass neutral density screen were used to approximate in situ irradiances (30% of incident light) at the time and depth of collection.

High performance liquid chromatography (HPLC) was used to determine chemosystematic photosynthetic pigments at the initiation of the experiment and after 24 h (Bioassay 1) or 48 h (Bioassay 2). Aliquots (0.5 l) of bioassay water were filtered under a gentle vacuum (<50 kPa) onto 2.5 cm diameter glass fiber filters (Whatman GF/F), immediately frozen, and stored at $-80\,^\circ$C. For analyses, frozen filters were placed in 100% acetone (1 ml), sonicated, and extracted at $-20\,^\circ$C for 12-20 h. Filtered extracts (300 µl) were injected into a Shimadzu HPLC equipped with a monomeric (Rainin Microorb-MV, 0.46 × 10 cm, 3 mm) and a polymeric (Vydac 201TP, 0.46 × 25 cm, 5 mm) reverse-phase C$_{18}$ columns in series. A nonlinear binary gradient, adapted from Van Heukelem et al. (1994), was used for pigment separations (for details, see Pinckney et al., 1996). Absorption spectra and chromatograms (440 nm) were acquired using a Shimadzu SPD-M10av photodiode array detector. Pigment peaks were identified by comparison of retention times and absorption spectra with pure crystalline standards, including chlorophylls a, b, β-carotene (Sigma Chemical Company), fucoxanthin, and zeaxanthin (Hoffman–LaRoche and Company). Other pigments were identified by comparison to extracts from phytoplankton cultures (Wright et al., 1991) and quantified using the appropriate extinction coefficients (Rowan, 1989; Jeffrey et al., 1997).

The abundance of algal groups was determined using CHEMTAX (CHEMical TAXonomy), a matrix factorization routine for calculating algal class abundances based on the concentrations of diagnostic chlorophyll and carotenoid phot pigments (Mackey et al., 1996; Wright et al., 1996; Pinckney et al., 1998). The program uses a steepest descent algorithm to determine the best fit based on an initial estimate of pigment ratios for algal classes. Input for the program consists of a raw data matrix of photopigment concentrations obtained by HPLC analyses and an initial pigment ratio file. The data matrix is subjected to a factor minimization algorithm that calculates a best fit pigment ratio matrix and a final phytoplankton class composition matrix. The class composition matrix can be expressed as relative or absolute values for specified photopigments. The absolute chlorophyll a (Chl a) contribution of each class is particularly useful because it partitions the total Chl a into major phytoplankton groups. Full discussions, validation, and sensitivity analyses of CHEMTAX are provided in Mackey et al. (1996). Previous studies comparing CHEMTAX and microscopy have consistently demonstrated the reliability of CHEMTAX for quantifying the relative abundances of major phytoplankton groups (for recent summaries see Jeffrey et al., 1999, Schluter et al., 2000, Wright and van den Enden, 2000).

For nutrient measurements, water samples (50 ml) were filtered through pre-combusted (500 C, 16 h) 25 mm Whatman GF/F glass-fiber filters before chemical analyses. Samples were analyzed for concentrations of nitrite, nitrate, ammonium, urea, phosphate, and silicate by the Marine Technical Services Group at Texas A&M University using standard auto analyzer techniques.

Fig. 1. Sample collection sites in Galveston Bay, Texas.
Bioassay responses (algal group abundances) to the manipulated factors were analyzed using a general linear model (GLM) multivariate analysis of variance (MANOVA) with two fixed factors (Neter et al., 1985; Scheiner and Gurevitch, 1993). Factor 1 was the bioassay experiment (2 levels; bioassay 1 and bioassay 2) and Factor 2 was the experimental treatment (5 levels; no addition control, atrazine + N + P, N alone, P alone, and N + P). The dependent variables were the CHEMTAX-derived abundances for 11 algal groups. The between-subjects effects for Factor 2 were used to test the null hypotheses that algal group-specific biomass was not significantly different between control and atrazine treatments. All data were ln-transformed before analysis to satisfy the normality assumption. Equality of error variances was checked using Levene’s Test (Neter et al., 1985). A Dunnett’s T3 test, which does not assume homogeneous variances, was used for post hoc multiple comparisons of treatment means.

3. Results

The first bioassay, Bioassay 1, was conducted on 11–12 March 2000 using water collected from Galveston Channel (Fig. 1). The salinity was 31.0 psu, water temperature was 18.0 °C, and nitrate and ammonium were present in approximately equimolar concentrations (Table 1). The addition of nitrate (N) and phosphate (P) at the beginning of the incubation resulted in a fourfold increase in the ambient dissolved inorganic nitrogen (DIN) concentration and a decrease in the DIN:P ratio from 5.29 to 3.64. The initial phytoplankton biomass (all algal groups) was 7.29 μg Chl a l⁻¹ (Fig. 2). HPLC photopigment and CHEMTAX analyses indicated that the community was composed primarily of diatoms, which represented 54% of the total algal biomass (as Chl a). Cryptophytes, chrysophytes, and prasinophytes comprised 21%, 10%, and 11% of the total community, respectively. Both N + P (no atrazine added) and atrazine treatments showed large increases in phytoplankton biomass, from 7.29 to nearly 25 μg Chl a l⁻¹, after 24 h incubation. Nearly all algal groups exhibited marked increases in biomass, with diatoms showing the greatest increase.

The second bioassay was conducted over 48 h using water collected from the central region of Galveston Bay (Fig. 1). In Bioassay 2 (13–15 March 2000), the salinity was lower (24.2 psu) and the temperature was the same as in Bioassay 1. The total DIN concentration (2.95 μM N) in the incubation water was nearly identical to Bioassay 1, but the relative proportions of the three N species differed from the previous experiment (Table 1). The addition of N and P resulted in a minor shift in the N:P ratio from 3.35 to 3.34. A comparison of the nutrient concentrations at the beginning and end of the incubation shows that nitrate and ammonium concentrations were reduced to <0.5 μM in all treatments (Fig. 3). Phosphate concentrations were also reduced (except in the control) while silicate was lower in the N and N + P treatments. The N:P ratio changed from an initial value (before nutrient additions) of 3.35 to <0.50 in all treatments except the nitrate addition. However, the change in the N:P ratio in the nitrate treatment (after nutrients were added) was larger than for all the other treatment groups.

Table 1

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>Bioassay 1</th>
<th>Bioassay 2</th>
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<tbody>
<tr>
<td></td>
<td>Initial concentration (μM)</td>
<td>Initial concentration (μM)</td>
</tr>
<tr>
<td>NH₄⁺</td>
<td>1.38</td>
<td>1.87</td>
</tr>
<tr>
<td>NO₃⁻</td>
<td>1.33</td>
<td>0.85</td>
</tr>
<tr>
<td>NO₂⁻</td>
<td>0.25</td>
<td>0.23</td>
</tr>
<tr>
<td>Urea</td>
<td>0.32</td>
<td>0.57</td>
</tr>
<tr>
<td>PO₄³⁻</td>
<td>0.56</td>
<td>0.88</td>
</tr>
<tr>
<td>SiO₃²⁻</td>
<td>12.34</td>
<td>7.77</td>
</tr>
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</table>
The initial phytoplankton community composition was also similar to Bioassay 1. Diatom, chrysophyte, cryptophyte, and chlorophyte contributions to total phytoplankton biomass ($15.44 \mu g Chl \alpha l^{-1}$) were 47, 22, 16, and 1%, respectively (Fig. 4). Compared to Bioassay 1, the initial biomass of all algal groups (except prasinophytes) was higher but the magnitude of the biomass increase during the incubation was lower (329% in Bioassay 1 vs. 112% in Bioassay 2). In the treatments that did not receive N additions (i.e., control and phosphate additions), the biomass of all algal groups declined during the incubation. The algal responses to N and atrazine additions varied among the different groups. Diatom biomass increased while other groups, such as the cryptophytes and chrysophytes, declined or were nearly constant in comparison with the initial biomass concentrations.

### Table 2

Results of MANOVA testing for significant treatment effects in the bioassays. The results of a posteriori comparisons of means are shown under conclusions. Treatments are ordered from highest to lowest mean value. The abbreviation Atr denotes the addition of atrazine + N + P. The underline indicates that means were not significantly different ($p < 0.05$).

<table>
<thead>
<tr>
<th>Algal group</th>
<th>$P$-value</th>
<th>Power</th>
<th>Conclusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>All groups</td>
<td>$&lt;0.001$</td>
<td>1.000</td>
<td>$Atr \ N \ NP \ P \ C$</td>
</tr>
<tr>
<td>Chlorophytes</td>
<td>0.015</td>
<td>0.828</td>
<td>$Atr \ N \ P \ C \ NP$</td>
</tr>
<tr>
<td>Chrysophytes</td>
<td>0.001</td>
<td>0.975</td>
<td>$Atr \ N \ P \ C$</td>
</tr>
<tr>
<td>Cryptophytes</td>
<td>$&lt;0.001$</td>
<td>1.000</td>
<td>$Atr \ NP \ N \ P \ C$</td>
</tr>
<tr>
<td>Diatoms</td>
<td>$&lt;0.001$</td>
<td>1.000</td>
<td>$N \ Atr \ NP \ C \ P$</td>
</tr>
<tr>
<td>Prasinophytes</td>
<td>0.072</td>
<td>0.611</td>
<td>$P \ C \ Atr \ NP \ N$</td>
</tr>
</tbody>
</table>

4. Discussion

Many estuarine and coastal waters with agricultural watersheds are exposed to increasing rates of herbicide loading. Subsequently, phytoplankton in these habitats may be impacted by the magnitude and frequency of herbicide loading events. In addition, these loading events are likely to co-occur with inputs of growth-stimulating nutrients. However, experimental data for mixed species assemblages, especially for estuarine
phytoplankton communities, are limited. Much of the published data on atrazine effects on algal species are based on the determination of the EC$_{50}$ (the effective concentration resulting in a 50% growth inhibition) using laboratory monocultures. These studies have shown that microalgae exposed to atrazine exhibit reduced photosynthetic capability, chlorophyll synthesis, cell growth, and nitrogen fixation (Hoagland et al., 1996). However, short-term (hours) exposure to as little as 1 µg l$^{-1}$ of atrazine can reduce photosynthesis and growth of some algal species (Hersh and Crumpton, 1989; Hoagland et al., 1996). Typical EC$_{50}$ values for marine phytoplankton range from 20 to 600 µg atrazine l$^{-1}$ in bioassays from 1 h to 10 days in duration (Solomon et al., 1996). The atrazine concentration used in the present study (25 µg l$^{-1}$) is at the lower end of reported EC$_{50}$ values, but represents a reasonable and realistic exposure level for phytoplankton in Galveston Bay.

Scaling these laboratory toxicity studies to natural systems is difficult because the synergistic effects of other stressors (e.g., competition, nutrient status, light, etc.) could result in a higher sensitivity of some species to atrazine exposure. The sublethal effects of atrazine may be manifested as shifts in species composition promoted by changes in competitive hierarchies (Drenner et al., 1998; Proulx and Mazumder, 1998; Siegel, 1998). Other studies provide considerable direct and indirect evidence to justify our initial hypothesis that low concentrations of atrazine would have significant sublethal effects on phytoplankton communities (Solomon et al., 1996).

The bioassay approach applied in this study represents a realistic appraisal of the potential ecological impacts of sublethal atrazine concentrations in estuarine ecosystems (Couture et al., 1989; Clements and Kiffney, 1994; Bonilla et al., 1998). For the experiments, we chose an atrazine concentration (25 µg l$^{-1}$) which represents an upper limit of potential exposure for phytoplankton communities in Galveston Bay. Because there are no direct measurements of atrazine concentrations within Galveston Bay, we estimated the relative concentrations of atrazine based on salinity. Using an atrazine concentration of 4 µg l$^{-1}$ at the mouth of the Trinity River and assuming conservative mixing, the lower reaches of the bay may have exposures on the order of 1 µg l$^{-1}$ of atrazine. Therefore the exposure levels used in the bioassays were far in excess (by a factor of 25) of concentrations likely experienced by phytoplankton in Galveston Bay. Therefore our experiments should reflect a worst-case scenario. The date of the bioassays (March) was selected because this represents the time of the year when inputs of atrazine are expected to be at a maximum (Frank et al., 1979; Goolsby et al., 1991; Goolsby, 1994; Clark et al., 1999).

Atrazine is a pre-emergent herbicide that is applied to fields before planting. Early spring rains may transport large quantities of atrazine into the estuary and therefore the resident phytoplankton community during this season will likely receive the highest exposure. Atrazine inputs into the estuary are likely to co-occur with nutrient loading events associated with agricultural runoff and riverine discharge into Galveston Bay. Sediment resuspension and release of atrazine bound to sediment particles may be an additional mechanism for episodic exposure in estuarine habitats (Bergamaschi et al., 2001). Therefore our experimental design focused on the potential synergistic impacts of atrazine exposure under conditions of high nutrient (N and P) concentrations.

Short-term bioassays were used to measure initial phytoplankton responses to atrazine exposure in this study. Changes in the biomass of specific algal groups (i.e., diatoms, cyanobacteria, dinoflagellates, cryptomonads, etc.) in natural Galveston Bay phytoplankton assemblages were measured to identify algal groups that may be affected by an acute (24-48 h) exposure to sublethal concentrations of atrazine. Although these results cannot be used to predict changes in natural assemblages, they provide a realistic assessment of which groups are affected by atrazine treatments under ambient (light, nutrient, temperature, etc.) conditions (Drake et al., 1996). Incubations of 1–2 days were sufficient to measure group-specific responses.

In both bioassays, the addition of nitrate stimulated significant increases in phytoplankton biomass relative to the controls and additions of phosphate alone. Nitrate and ammonium concentrations were reduced to <1.00 µM during the incubations, while phosphate and silicate concentrations reflected the patterns of N utilization and phytoplankton biomass. However, neither P nor Si appeared to be limiting in either of the bioassays. These results suggest that the phytoplankton community in Galveston Bay (at the time of the bioassays) was N-limited and the rapid biomass increase following N additions reflects a highly-responsive phytoplankton community, even in the presence of atrazine.

Atrazine additions (25 µg l$^{-1}$) did not have any detectable effect on phytoplankton community composition or biomass in either of the bioassays performed in this study. In fact, the addition of atrazine in combination with nitrate resulted in increases in the biomass of all phytoplankton groups (except prasinophytes) that were not significantly different from additions of nitrate alone. Therefore, the general conclusion for these experiments is that there was no significant negative effect of atrazine on phytoplankton under the specified environmental conditions for the bioassays. The absence of a significant negative response to atrazine could reflect a phytoplankton community that has become acclimated to atrazine exposure. However, our best estimate of potential exposure levels for Galveston Bay phytoplankton (i.e., <5 µg atrazine l$^{-1}$) suggests that a “pre-
acclimated” community is not likely for this estuary, especially for the concentrations used in the incubations (i.e., 25 μg atrazine L⁻¹).

These results should not be extrapolated to make general statements about the environmental effects of atrazine in estuarine habitats. The limited scope of this study does not reflect the inherent variability in phytoplankton community composition or the wide range of environmental conditions in estuarine and coastal waters. The timing of atrazine loading could be very important for determining the impacts on phytoplankton.

There is sufficient evidence to suggest that the sublethal effects of low concentrations of these compounds may be partly responsible for the observed increase in the incidence of harmful algal blooms in estuarine and coastal waters (ECOHAB, 1995). Nuisance species with a high tolerance for atrazine may be able to outcompete less tolerant species, especially over longer durations of exposure. More incipient problems may include alterations in the trophic structure of these systems, leading to the disappearance of important fisheries, an increase in the incidence of pathogens, and impairment of ecosystem function. Although the results of this study suggest that low concentrations of atrazine will have minimal impacts on phytoplankton communities, the identification of the sources, mechanisms, routes, frequencies, and magnitudes of herbicide loadings need to be further characterized before generalizations can be applied to estuarine and coastal ecosystems.

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