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Growth Responses of *Typha latifolia* and *Scirpus acutus* to Atrazine Contamination

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Nonpoint source (NPS) pollution is the major cause of impairment of U.S. surface waters (Baker 1992). Pesticides, often an important component of NPS, can have a significant and complex impact on the structure and function of the trophic chain (Coman and Dordea 1990). Herbicides can percolate into subsurface water flow and be carried long distances in overland flow (Wu *et al.* 1983). Wetlands receive runoff and may be recharged through groundwater, both of which have been found to be contaminated by herbicides (Pionke *et al.* 1988, Exner and Spalding 1990, Baker 1992). Wetlands exposed to these pollutants behave similarly to terrestrial and other aquatic systems, e.g. species abundance and diversity can change, habitat quality can deteriorate, and energy transmission through food networks can be altered (Catallo 1993).

Atrazine (2-chloro-4-ethylamino-6-isopropylamino-s-triazine), used primarily for pre- and post-emergence control of germinating weeds in corn (Hartley and Kidd 1983), is the most commonly applied herbicide in the U.S. It acts as a powerful photosynthetic inhibitor, interrupting the light-driven flow of electrons (Esser *et al.* 1988). In studies done in the northeastern U.S., Wu *et al.* (1983) found that even in areas where alachlor was applied in greater quantities, atrazine was still detected in runoff waters more frequently and in greater concentration. Atrazine concentrations of 13.9 $\mu g \cdot L^{-1}$ can negatively affect stream drift populations of both phytoplankton and zooplankton (Lakshminarayana *et al.* 1992). Atrazine (20 $\mu g \cdot L^{-1}$) also affects nonpredatory aquatic insects in artificial pond systems, primarily through indirect effects, i.e. by reduction in food for nonpredators and reduction in habitat because of decreases in periphyton and macrophytes (Dewey 1986). It has been shown to inhibit photosynthetic rates and development of aquatic macrophytes (Forney and Davis 1981, Jones and Winchell 1984, Jones *et al.* 1986, Christopher and Bird 1992). Aquatic communities with submerged macrophytes may also experience changes in competitive interactions in response to atrazine contamination (Cunningham et *al.* 1984). The highest inputs of agrichemicals to wetland habitats are at the time of or immediately following chemical application (Clark *et al.* 1993). In stream water samples collected during post-planting storm events, Langan *et al.* (1993) found atrazine in concentrations as high as 691 $\mu g \cdot L^{-1}$, along with 635 $\mu g \cdot L^{-1}$ alachlor, 117 $\mu g \cdot L^{-1}$ cyanazine, and six other herbicides, where background scans of the stream water were negative except for 2,4-D

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at 0.7 μg·L^{-1}. However, sediment scans showed atrazine at 0.03 μg·L^{-1} and alachlor at 0.02 μg·L^{-1}.

Wetlands serve important functions in water quality enhancement (Richardson *et al.* 1978, Tiner 1991), such as transforming, filtering, and storing various nutrients and pesticides (Landers and Knuth 1991, Hook 1993). Wetlands have been shown to be effective in decreasing nutrient loads in wastewater (Barten 1983). Microbes attached to substrates within the wetland modify these loads (Surrency 1993). Highly efficient leaf display and extensive carbohydrate stores allow high productivity in emergent vegetation (Boston *et al.* 1989), increasing their ability to decrease nutrient levels in water. Emergents also reduce the flow of sediment in runoff, exchange nutrients with associated ecosystems (Catallo 1993), and keep sediment from resuspending (Dieter 1990).

It has been proposed that discharging agricultural runoff into wetlands parallel to stream channels may be one method for reducing NPS herbicides and nutrients entering streams via runoff from tile drains (Osborne and Kovacic 1993). The effects of pesticides on restored or constructed wetlands need to be studied before effective guidelines for using these wetlands for the reduction of NPS pollutants can be developed (van der Valk 1993). The purpose of this study was to assess the ability of two major wetland macrophytes, *Scirpus acutus* Muhl. and *Typha latifolia* L., to tolerate atrazine contamination. Experimental microcosm bioassays were conducted to determine the direct effect of atrazine on these wetland species, by assessing its impact on plant height.

MATERIALS AND METHODS

Typha latifolia (broad-leaved cattail) and *S. acutus* (hardstem bulrush) were used as representative wetland species. Rhizomes were obtained from Wildlife Nurseries Inc. (Oshkosh, WI). One rhizome and approximately 257 g of dry soil were placed in each 9x8-cm pot, and four pots were put into one plastic 12-L tub (Tucker, Arlington, TX). Distilled water contaminated with the appropriate level of atrazine was added to the tubs to keep the water level approximately 1 cm above the soil surface. Water levels were maintained throughout the experiment by adding distilled water. The combined measurements of four plants in one tub were considered one experimental unit and there were six replicate tubs of each treatment concentration.

Commercially available atrazine (Aatrex®, Ciba-Geigy Corp., Greensboro, NC) was used to establish a concentration gradient. To reduce variability, the water for each treatment concentration was combined and contaminated before filling the experimental microcosms. Arandomized complete block design was used because of an east to west temperature and light gradient in the greenhouse. Thus, the experiment consisted of six blocks, two species, and seven treatments: a control with no atrazine added, a gradient of nominal atrazine concentrations including 10, 50, 100, 500, and 1500 μg·L^{-1}, and a decontaminated treatment in which the water from a previously contaminated 500 μg·L^{-1} treatment was replaced with distilled water after 2 wk to simulate a wetland dilution event. At 16 wk, the atrazine levels in both 50 and 500 µg·L^{-1} treatments were found to be 1.27 and 1.55 µg·L^{-1}, respectively.

Metalarc lamps (400 W) were used to minimize light intensity differences among the blocks and to maximize the amount of incident irradiance. Light, measured with a quantum irradiance meter and sensor (Li-Cor, Model LI-185A), ranged between 75 and 100 µmol·$m^{-2}s^{-1}$ without sunlight. Lamps were kept on a 12:12 h light:dark cycle and the temperature was maintained at 70°C.

The height of *S. acutus* and the combined length of all leaves of *T. latifolia* were measured at biweekly intervals during a 16 wk period. Means comparisons were made on the contaminated treatments and the control using the Least Significant Difference procedure. Growth curves were compared using an ANOVA with repeated measures. Comparisons between the control, the 500 µg·L^{-1} treatment, and the decontaminated 500 µg·L^{-1} treatment were made using a repeated measures ANOVA with orthogonal contrasts on the same measurements. All results reported as significant have a p-value of less than 0.05 ($p<0.05$).

RESULTS AND DISCUSSION

Scirpus acutus

Plant height was negatively affected by atrazine at the 500 and 1500 µg·L^{-1} levels (Fig. 1), despite the low parent compound concentration of 1.55 µg·L^{-1} found in the 500 µg·L^{-1} treatment water at the end of 16 wk. The duration of the study and the experimental conditions used presumably allowed microbial (Hamilton *et al.* 1993) or chemical degradation, which may be more important in the environment (Solomon *et al.* 1996).

Scirpus acutus was able to recover after short-term exposure to the herbicide, as its linear growth response was not significantly different between the control and the decontaminated treatments by the end of the experiment, even though the means had differed significantly earlier in the experiment (Pig. 2). Growth in both the contaminated and decontaminated treatments was inhibited at 2 wk. Plant growth began to increase in the decontaminated treatment, becoming significantly greater than the contaminated treatment at 10 wk. Growth of the plants was significantly inhibited for the contaminated treatments. Differences in growth between the control and the decontaminatedtreatments were no longer evident by 16 wk (Fig. 2).

Typha latifolia

Typha latifolia was also affected by high levels of atrazine (Fig. 3; Table 2). No significant differences in plant growth were apparent during the first 4 wk of the experiment. However, the 1500 µg·L^{-1} treatment had a significant negative effect on growth from 6 wk to 16 wk (Fig. 3; Table 2). Plant growth in the contaminated treatment was significantly inhibited at 8 and 10 wk, but no other effects on total height were found (Fig. 4).

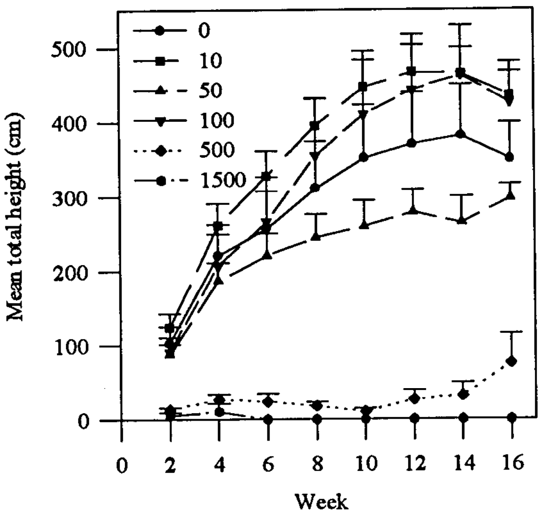

Figure 1. Mean total height (cm) of *S. acutus* along an atrazine gradient of 0 to 1500 µg/L. ($\overline{x}$ ± S.E.)

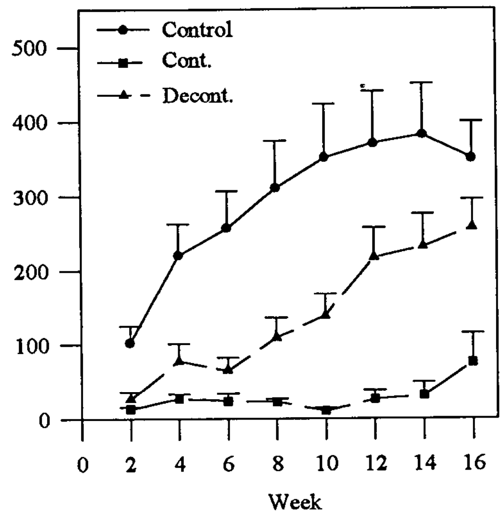

Figure 2. Mean total height (cm) of *S. acutus* for control, contaminated,an decontaminated treatments. ($\overline{x}$ ± S.E.)

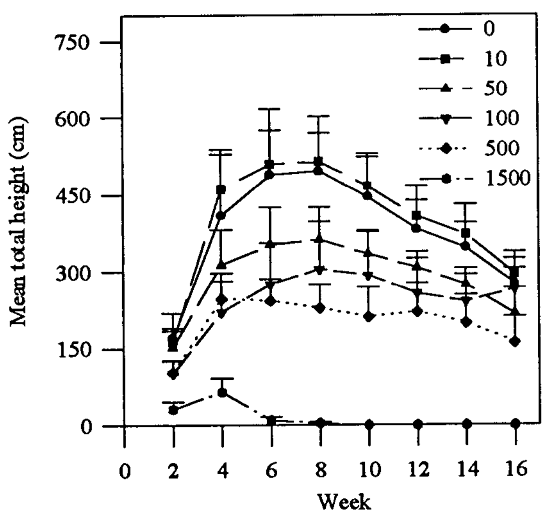

Figure 3. Mean total height of *T. latifolia* along an atrazine gradient of 0 to 1500 µg/L. ($\overline{x}$ ± S.E.)

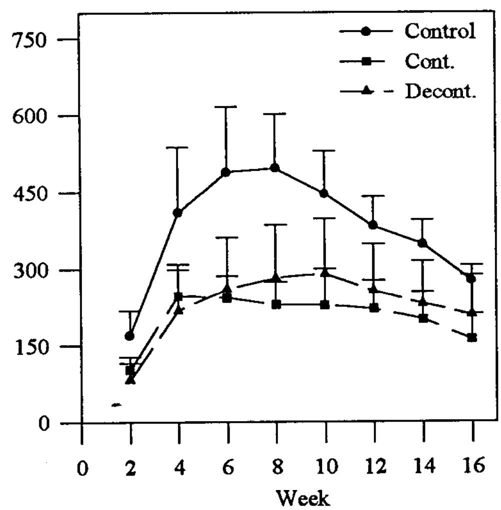

Figure 4. Mean total height (cm) of *T. latifolia* for control, contaminated,and decontaminated treatments. ($\overline{x}$ ± SE.)

Table 1. Least significant differences for mean total height of *Scirpus acutus*. Means are listed in descending order, means for underlined treatments do not differ significantly. Weeks 2-6 and 8-16 did not differ and are grouped. Atrazine concentrations in $\mu g \cdot L^{-1}$ ($p < 0.05$).

Species	Week	Least Significant Differences
S. acutus	2-6	10 100 0 50 500 1500
	8-16	10 100 0 50 500 1500

Table 2. Least significant differences for mean total height of *Typha latifolia*. Means are listed in descending order, means for underlined treatments do not differ significantly. Weeks 6-12 did not differ and are grouped. Atrazine concentrations in $\mu g \cdot L^{-1}$ ($p < 0.05$).

Species	Week	Least Significant Differences
T. latifolia	2	0 10 50 500 100 1500
	4	10 0 50 500 100 1500
	6-12	10 0 50 100 500 1500
	14	10 0 50 100 500 1500
	16	10 0 100 50 500 1500

Scirpus acutus and *T. latifolia* grew at low levels of atrazine exposure. *S. acutus* reached its greatest height at 12 wk, *while T. latifolia* reached full height at 8 wk. Plant growth eventually declined at all concentrations, including the control. Growth was inhibited at some atrazine concentrations, but *S. acutus* was not completely prevented from growing except at 500 and 1500 $\mu g \cdot L^{-1}$, and *T. latifolia* at 1500 $\mu g \cdot L^{-1}$. Chlorosis was observed in plants that were negatively affected, but not at the lowest concentration. Though not statistically sign&cant, a trend toward slightly enhanced growth at the 10 $\mu g \cdot L^{-1}$ concentration was seen for both species. This may be a result of plant hormone metabolism being influenced by the triazine herbicides (Esser et al. 1988). It should be noted that phytotoxic effects may be greater in greenhouse studies than in the field studies because controlled conditions may make growth more rapid; with moisture conditions closer to optimum and all of the roots in treated soil, the risk from herbicide residues may be exaggerated (Riley and Eagle 1990).

Large watersheds with heterogeneous land use have much lower herbicide levels in runoff than those reported for ditches or crop field plots (Wu *et al.* 1983). Bulk edge of field atrazine concentrations have been found as high as 4700 $\mu g \cdot L^{-1}$ (Wauchope 1978). If wetlands near agrichemical application sites were receiving runoff with atrazine concentrations at these levels, it is clear that the negative effects on these macrophytes would be direct. Even short-term growth depression could affect the ability of these wetlands to improve water quality, consequently runoff water containing atrazine at high levels would require prior dilution. As this study demonstrates, wetland macrophytes may not tolerate herbicide contamination equally, thus species composition is an important consideration in wetland design for treatment of highly contaminated runoff. In this case, *T. latifolia* is more tolerant of atrazine, and could be used in wetlands constructed for these purposes. The effects of short- or long-term agrichemical contamination of natural wetlands are unknown.

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