J. Aquat. Plant Manage. 39: 112-117

Simazine Toxicity and Uptake by Parrotfeather

PATRICK C. WILSON[1], TED WHITWELL[2] AND STEPHEN J. KLAINE[3]

ABSTRACT

The potential use of *Myriophyllum aquaticum* L. (parrotfeather) for removing simazine residues from contaminated water was examined by establishing toxicity thresholds and characterizing the uptake and distribution. Toxicity thresholds were determined by growing plants for 7 d in nutrient medium amended with 0, 0.01, 0.03, 0.1, 0.3, 1.0, or 3.0 mg simazine/L. Measured endpoints included: fresh biomass production, stem elongation, and branch production. Pesticide uptake and distribution within plants was determined by growing plants in nutrient medium amended with [^{14}C] ring-labeled simazine at concentrations of approximately 0.242 mg/L simazine. Plants were randomly harvested after exposure, dissected into individual parts, combusted, and analyzed by liquid scintillation counting. In comparison to unexposed plants, fresh weight gains were reduced 47, 98, and 105% at 0.3, 1.0 and 3.0 mg/L simazine, respectively, after 7 d exposure. Elongation was reduced 60 and 49% at 1 and 3 mg/L, respectively, and branch production was reduced 55, 95, and 95% at the 0.3, 1, and 3 mg/L, respectively. Simazine activity in plant-containing solution was reduced 9, 18, 31, and 49%, after 1, 3, 5, and 7 days, respectively. Activity levels did not decline significantly in reference solutions lacking plants. By day 5, [^{14}C] activity was detected predominantly in leaves. Simazine uptake was correlated with water uptake throughout the 7 days. Activity in non-plant-containing solution remained relatively constant throughout the experimental period. These results suggest that parrotfeather may be a good candidate for incorporation into a phytoremediation scheme for simazine.

Key words: Myriophyllum aquaticum, triazine, ^{14}C, herbicide.

INTRODUCTION

The turfgrass and nursery industries have become popular specializations of production agriculture. This research was undertaken to help these industries reduce risks to nontarget organisms associated with the common practice of washing pesticide application equipment following use.

Simazine (2-chloro-4,6-bis(ethylamine)-s-triazine) is the herbicidal active ingredient of commercial formulations of Princep (Novartis Crop Protection, Greensboro, NC). It is a broad-spectrum, systemic herbicide. Simazine is moderately soluble in water (6.2 mg/L at 22C) and is not volatile (vapor pressure: 1.5×10^{-8} mm Hg at 25C) (Anonymous 1995). Reported half-lives in the field range from 28 d to 149 d (Wauchope et al. 1992). Simazine inhibits photosynthetic electron transport by binding to the plastiquinone B (Q_B) protein binding site on the D1 protein of the Photo-System II (PSII) complex in chloroplast thylakoid membranes (Ahrens 1994). The bound simazine prevents transfer of electrons from plastiquinone A (Q_A) to Q_B, thus breaching photosynthetic electron transport, CO_2 fixation, and the production of ATP and $NADPH_2$. However, plant death most commonly results from subsequent lipid peroxidation caused by triplet state chlorophyll and singlet oxygen, common products resulting from the inability to reoxidize Q_A (Ahrens 1994).

Commercial formulations of simazine are labeled for use in the agricultural, turfgrass, and nursery industries. Expected simazine concentrations in 568 L of rinse water may range from 9.2-92.5 mg/L, assuming a moderate application rate, 378 L original mix volume, and a residual rinse volume ranging from 3.8 to 37.9 L between rinses. These industries may generate significant amounts of simazine-contaminated water by the common practice of rinsing sprayers after use. Most manufacturers recommend that the rinsates be applied to areas that are normally treated. Improper disposal of these contaminated rinsates may result in significantly altered nontarget ecosystems due to simazine's effectiveness as a photosynthetic inhibitor.

Phytoremediation offers one possible method for removing pesticides from contaminated water. Phytoremediation is the use of plants, plant growth, and metabolism as a technique for detoxifying environmental sites contaminated with organic and inorganic pollutants. This technology exploits the ability of plants to extract and/or mineralize xenobiotics in the surrounding environment, as well as the tolerance of these plants to the contaminants. According to Schnoor et al. (1995), this developing technology is best suited for terrestrial sites with shallow contamination (<5 m depth); moderately hydrophobic pollutants (log K_{ow} = 0.5-3); short-chain aliphatic chemicals; and excess nutrients. The log K_{ow} for simazine is 2.08 (Ahrens 1994). It is generally recognized that plants can remediate organic pollutants by: 1) direct root uptake of contaminants and subsequent accumulation of non-phytotoxic metabolites in plant tissue, 2) direct foliar uptake of volatile contaminants from the surrounding air by foliage, and 3) release of exudates and enzymes that enhance biochemical transformations and/or mineralization due to mycorrhizal fungi and microbial activity in the rhizosphere (Schnoor et al. 1995). Anderson et al. (1993) and Anderson and Coats (1994) have reviewed bioremediation in the rhizosphere by microbial and biochemical processes.

Research reported in this paper was part of a larger project concerned with the development of a porous-root-zone based constructed wetland for remediation of pesticide-

[1]Indian River Research and Education Center, University of Florida/IFAS, Fort Pierce, FL.

[2]Department of Horticulture, Clemson University, Clemson, SC.

[3]Department of Environmental Toxicology, Clemson University, Pendleton, SC. Received for publication October 26, 1999 and in revised form September 14, 2001.

contaminated rinsates. In the development of such a system, it was imperative that resident plant health not be compromised by excessive pesticide concentrations. Likewise, knowledge of uptake capacities and distribution within the plant was essential. Simazine was chosen as a model compound because of its common use by both industries. Most work concerning the uptake and distribution of simazine reported in the literature has centered on ascertaining or enhancing herbicidal modes of action. In their literature review on triazine uptake, translocation, and degradation in plants, Esser et al. (1975) reported that root uptake readily occurred with all plants studied regardless of whether they were resistant or susceptible to the herbicides. They also reported that increasing concentration and time-of-exposure resulted in increased uptake. Uptake rates were also accelerated by high temperatures and low relative humidity (Esser et al. 1975). As a class, the triazines were seen to be evenly distributed by way of the xylem into all aerial parts of plants following root uptake. Specific plant species that have been shown to readily transport triazines acropetally from roots to leaves include: corn, cotton, and cucumber (Davis et al. 1959), Norway spruce (Lund-Hoie 1969), black walnut and yellow poplar (Wichman and Byrnes 1975), poplar clones (Akinyemiju et al. 1983), radish seedlings (Shone and Wood 1976), and barley (Shone et al. 1974, Shone and Wood 1972, 1974). Very little work on simazine uptake has been reported in the context of phytoremediation, with the exception of Burauel and Fuhr (1988) who reported enhanced mineralization of simazine in soil following uptake by maize and subsequent breakdown of the plant.

Specific objectives of this research included: 1) determining the toxicity threshold of parrotfeather for simazine, 2) quantifying [^{14}C]simazine uptake into the plant from a spiked solution, and 3) determining the distribution of [^{14}C]simazine throughout the plant.

MATERIALS AND METHODS

Toxicity tests were conducted using technical grade simazine (reported purity: 99.6%). The uptake and distribution studies were conducted using uniformly ring-labeled [^{14}C]simazine. This material had reported chemical and radio-purities of 99.3% and 98.7%, respectively. Both of these compounds were obtained from Novartis Crop Protection.

Parrotfeather (*Myriophyllum aquaticum* L., syn. *M. brasiliense* Camb.) was selected because of its aesthetic characteristics and hardiness in the southeastern United States. This is a perennial aquatic plant with elongated stems and whorled leaves (Correll and Correll 1975). Plants may be rooted in mud or free-floating, with the feathery foliage floating above the water surface. Plants are typically pallid or pale-green in color. Plants reproduce primarily by vegetative means, readily forming adventitous roots at nodes. Sexual reproduction also occurs by means of axillary, unisexual and perfect flowers present on the same plant. Ducks, other wildfowl, and muskrats are known to eat the fruits and occasionally herbage of this species (Correll and Correll 1975). Parrotfeather colonies provide shelter and breeding grounds for fish and insects, which in turn attract waterfowl (Correll and Correll 1975). This species is considered a noxious weed in some areas due to its aggressive growth and reproduction. However, it is also sold by commercial nurseries as an ornamental plant for water gardens.

Original plant stocks were obtained from Carolina Biological Supply (Burlington, NC). Plants were propagated hydroponically by rooting stem cuttings in 10% Hoagland's nutrient medium. Plants were watered and fertilized as needed. Approximately 2 to 3 weeks before tests were initiated, plants were transferred from ambient conditions in the greenhouse to hydroponic culture in the lab. The hydroponic system consisted of several 0.5-1 L vacuum flasks filled with 10% Hoagland's nutrient solution. Plants were grown (lower stems submersed) in these containers during the acclimation periods. The nutrient solution was changed weekly. Liquid lost due to evapotranspiration was replaced with distilled, deionized water. Plants were grown under test conditions during the acclimation period.

Toxicity Assessment

Toxicity tests with simazine were conducted at 25 ± 2C under metal halide lamps with a photon flux density of 375 ± 25 µmol/m^2/sec and a 16 h light: 8 h dark photoperiod. Treatment solutions were made by dissolving simazine overnight in 10% Hoagland's nutrient medium at the following concentrations: 0, 0.01, 0.03, 0.1, 0.3, 1.0, and 3.0 mg/L. Simazine concentrations were confirmed using SDI RaPID Immuno Assays. Individual plants were exposed in glass jars to 250 mL of each pesticide concentration for 7 d. All exposure jars were autoclaved before treatment. Each jar was also covered with aluminum foil to exclude extraneous light from the root zone and exposure medium. Each plant was held in place with lower stems and roots submerged in medium using the lower half of a 236 mL foam cup with a hole cut into the bottom. Plant roots were rinsed with distilled water after the 7-d exposure period. Plants were then placed in simazine-free nutrient medium and allowed to grow for an additional 7 d in order to observe any latent effects or recovery.

Fresh weights and plant lengths were recorded before exposures, after 7 d exposure, and after the 7-d post-exposure period. These measurements were used to calculate fresh weight and lengthwise growth gains during the exposure and post-exposure periods. The number of branches produced per plant was also measured after the post-exposure period.

A completely randomized statistical design with 4 replications for each exposure concentration was used. All data were ranked and analyzed by ANOVA (P = 0.05). Results were further analyzed using calculated Least Significant Differences (LSD). ANOVA and LSD analyses were conducted using SAS statistical software. Regression analyses were conducted using M.S. Excel.

Uptake and Distribution

The uptake and distribution of simazine by parrotfeather was evaluated using [^{14}C]simazine labeled at the 2, 4, and 6 positions in the triazine ring. The specific activity of the [^{14}C]simazine was 30.3 µCi/mg. Approximately 28 µCi of [^{14}C]simazine were dissolved overnight in 3.82 L 10% Hoagland's nutrient medium. This was equivalent to approximate-

ly 0.242 mg simazine/L, or 4,509,574.01 disintegrations per minute (dpm)/275 mL nutrient solution. Fourteen 250 mL side-arm vacuum flasks were each filled with 275 mL of the spiked nutrient medium. Plants in non-spiked nutrient medium served as untreated controls and were subject to the same conditions as spiked samples. Side-arms of the exposure flasks were equipped with one-way valves that allowed movement of gases from outside the flask to the inside. Individual plants were held in place (with roots submerged in the spiked or non-spiked nutrient medium and shoots above the flasks) by #6 silicon stoppers with holes bored through the tops, and a slit along the side. Stoppers were wrapped around individual plant stems and sealed using Qubitac (Qubit, Kingston, ON, Canada) nontoxic, nonreactive putty. A smaller hole in the stopper was fitted with a teflon tube (1.6 mm I.D., 3.1 mm O.D.) that was connected to an in-line $[^{14}C]CO_2$ and volatile organic carbon (VO$[^{14}C]$) trap. The traps contained 15 mL of 0.5 M NaOH and 2 g of 20-60 mesh activated charcoal (Sigma, St. Louis, MO), respectively. Two to four flasks containing the spiked nutrient medium, but without plants were also included as references. These reference flasks were equipped and handled in the same manner as the others.

Once plants were secured in the exposure vessels, they were transferred to a Conviron CMP3244 (Controlled Environments, Winnipeg, MB, Canada) environmentally controlled growth chamber (light intensity, 375 ± 25 µmol/m²/s generated by fluorescent and incandescent lamps; photoperiod: 16 h light: 8 h dark; relative humidity: 60%; and temperatures: 25C light: 22C dark). Headspace within each flask was purged through the traps daily with 2 to 3 volumes of air using a 60 mL syringe attached to the side-arm one-way valve assembly. Water transpired through the plant was replenished with distilled, deionized water using the same syringe. Water use was recorded daily.

On days 1, 3, 5, and 7, three exposed and two control plants were randomly harvested. Plant roots and lower stems were rinsed in running tap water for 45 s and blotted dry. Plants were dissected into individual leaves, stems, and roots. Stems were arbitrarily divided into two categories—upper and lower. Upper stems were stem sections above the flask stopper (aerial), while lower stems were sections below the flask stopper. Fresh weights for each plant part were recorded. Plant tissues were wrapped in aluminum foil, flash frozen using liquid nitrogen, and stored at -80C until the tissues could be analyzed. At the time of analysis, tissues were freeze dried, weighed, and combusted in a R. J. Harvey Biological Oxidizer (Hillsdale, NJ) at 900C for 3 min. $[^{14}C]CO_2$ generated by combustion was captured using R. J. Harvey $[^{14}C]CO_2$ trapping cocktail (Hillsdale, NJ). The captured $[^{14}C]CO_2$ content was analyzed using a Beckman LS 6500 Liquid Scintillation Counter (Fullerton, CA). Each sample was counted for 8 min in the DPM (Disintegrations Per Minute) mode.

Exposure Solution Analysis

In addition to harvesting the plant tissue, samples of exposure solutions and the NaOH CO_2 scrubbers were analyzed for total $[^{14}C]$ content at the same time intervals. Total $[^{14}C]$ content was assessed by adding 0.2 mL of the exposure or scrubber solution to 6 mL Scinti Verse BD scintillation cocktail, and analyzed by liquid scintillation spectroscopy as mentioned earlier. The VOC scrubbers were not analyzed since simazine is not likely to volatilize due to its low vapor pressure.

Disposition of simazine in the exposure solution was determined by high pressure liquid chromatographic (HPLC) analysis. Samples (3 mL) were filtered through 0.2 micron Polytetrafluoroethylene (PTFE) Acrodiscs. The filtered samples were analyzed using a Waters 600E HPLC system controller equipped with a Waters Nova-Pak C_{18} Radial Compression Analytical column (4 µm particle size, 60 A pore size, 8 × 100 mm), β-RAM yttrium silicate radio-chemical detector (IN/US Systems, Fairfield, NJ) and a Waters 484 tunable UV absorbance detector (λ: 254 nm). The mobile phase consisted of a gradient starting at 10% MeOH:90% H_2O and changing to 70% MeOH:30% H_2O over a 30 min period. The injection volume was 0.700 mL and the flow rate was adjusted to 1.5 mL/min.

RESULTS AND DISCUSSION

Toxicity

Fresh weight gains for parrotfeather at the 0.3, 1, and 3 mg/L treatment levels after 7 d exposure, respectively, were 53, 2, and -5% of the controls (Figure 1). Fresh weight gains during the post-exposure period were similar to the controls for the 0.01, 0.03, and 0.1 mg/L treatment levels, but were only 20, -13, and -15% of the controls at the 0.3, 1, and 3 mg/L treatment levels, respectively. Leaf chlorosis and desiccation was noticeable after 5 d exposure.

Increases in primary stem elongation were only 40 and 51% of the controls for plants exposed to 1 and 3 mg/L simazine for 7 d, respectively (Figure 2). Following the post-exposure period, it was observed that many branches had grown from axillary buds along the stems. It was likely that the plants were putting their energy into branch production instead of primary stem elongation. For this reason, measurements of total increases in length (including branches) were used post-exposure. Stem elongation was 81, 40, and 51% of the controls at the 0.3, 1, and 3 mg/L treatment levels, respec-

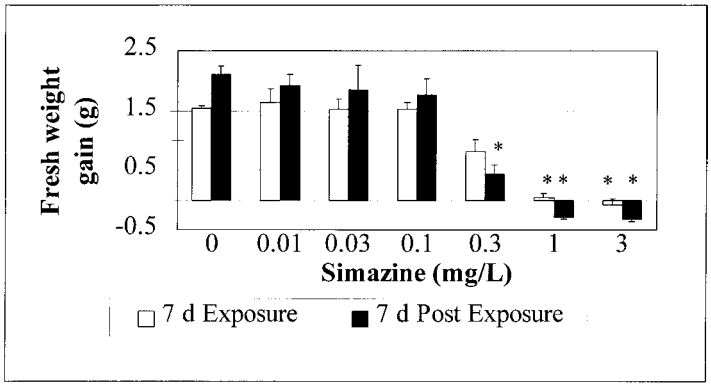

Figure 1. Effects of simazine on fresh weight gains of parrotfeather (g/plant) after a 7-d exposure and post-exposure period. Bars represent standard errors of the mean. Statistical comparisons are only valid within exposure categories. Asterisk (*) indicates mean is significantly different from controls ($P = 0.05$).

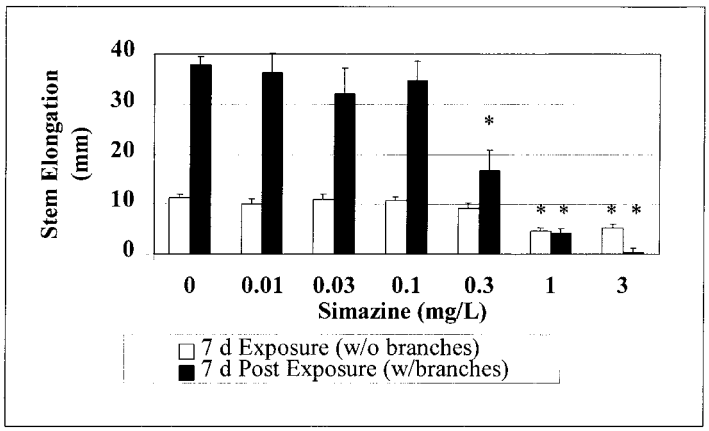

Figure 2. Effects of simazine on stem elongation of parrotfeather after a 7-d exposure and post-exposure period. Bars represent standard errors of the mean. Statistical comparisons are only valid within exposure categories. Asterisk (*) indicates mean is significantly different from controls (P = 0.05).

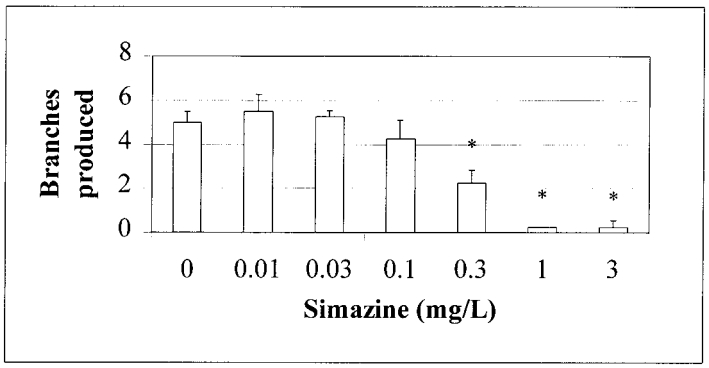

Figure 3. Effects of simazine on branch production of parrotfeather after a 7-d exposure and 7-d post-exposure period. Bars represent standard errors of the mean. Asterisk (*) indicates mean is significantly different from controls (P = 0.05).

tively. Likewise, the total number of branches produced per plant during the 14 d test period were 45, 5, and 5% of controls for the 0.3, 1, and 3 mg/L treatments, respectively (Figure 3).

Uptake and Distribution

[^{14}C]simazine activity in the plant-containing exposure solutions decreased with time (Table 1). These reductions were 9, 18, 31, and 49%, respectively, after 1, 3, 5, and 7 d of exposure. Regression analysis indicated the amount of activity remaining in solution was inversely proportional to the cumulative water-use by each plant [$P = 1.42 \times 10^{-10}$; % remaining = -0.00203*(cumulative water used) + 0.97163, $R^2 = 0.99$]. No significant amount of [^{14}C] was detected in the NaOH traps, indicating little loss due to mineralization in the rhizosphere. Though mineralization of [^{14}C]simazine by the plant shoots was not measured, Shone and Wood (1972) found no [^{14}C]CO_2 in air passed over barley plants that were grown in medium dosed with 0.200 mg [^{14}C]simazine/L for 48 h. Losses due to volatilization were probably not significant due to simazine's low vapor pressure of 1.5×10^{-8} mm Hg and Henry's Law constant of 9.5×10^{-10} atm/(mole/m^3) (Ahrens 1994). The non-simazine radio-impurity and/or metabolite detected by HPLC increased slightly relative to unplanted solutions. However, this slight increase may have been due to the presence of the plants, to day-to-day variation in measurement techniques, or to some microbial activity. The increases never exceeded 3% more than those in unplanted controls. This unknown was not identified, but appeared to be more polar than simazine as evidenced by its shorter retention time of 12 min as compared to 20 min for simazine. Nevertheless, since impurity/metabolite levels in

TABLE 1. PERCENT[1] DISTRIBUTION OF TOTAL [^{14}C] ACTIVITY (DPM; DISINTEGRATIONS PER MINUTE) FROM [^{14}C]SIMAZINE INITIALLY PRESENT IN EXPOSURE SOLUTIONS WITH AND WITHOUT *MYRIOPHYLLUM AQUATICUM*. INITIAL CONCENTRATION OF [^{14}C]SIMAZINE WAS 0.242 MG/L.

	Day 1		Day 3		Day 5		Day 7	
Fraction	n = 3	C.V.[2]	n = 3	C.V.	n = 3	C.V.	n = 3	C.V.
Experimental vessels with plants								
Solution[3]	0.849	0.01	0.731	0.02	0.599	0.06	0.431	0.07
Impurity[4]	0.064	0.01	0.090	0.02	0.089	0.06	0.076	0.07
Roots	0.012	0.049	0.023	0.359	0.019	0.139	0.024	0.291
Lower Stems	0.016	0.216	0.019	0.211	0.030	0.088	0.031	0.082
Upper Stems	0.023	0.151	0.065	0.327	0.058	0.327	0.079	0.170
Leaves	0.042	0.303	0.076	0.464	0.173	0.099	0.291	0.107
Unaccounted[5]	+0.006	—	+0.004	—	0.032	—	0.068	—
Reference vessels with no plants								
Solution[3]	0.92	0.009	0.92	0.006	0.93	0.007	0.93	0.011
Impurity[4]	0.07	0.01	0.08	0.01	0.08	0.01	0.07	0.01

[1]Fractional form.
[2]C.V. = Coefficient of variation.
[3]Solution = metalaxyl in exposure solution.
[4]Impurity = unidentified impurity and/or metabolite in exposure solution.
[5]"+" = activity in excess of what was originally added.

these flasks remained relatively constant, the assumption that the activity detected in plants was due primarily to uptake of parent simazine was valid.

Nearly all of the [^{14}C]simazine removed from the exposure solution was detected in the plants. After 1, 3, 5, and 7 d exposure, 9, 19, 28, and 43% of the total activity added, respectively, was detected in the whole plants. Activity detected in roots and lower stem sections may have also originated from [^{14}C] adsorbed to the exterior of these plant parts. The mass balance for [^{14}C] was generally good throughout the test, with unmeasured label never exceeding 6.8% (Table 1). Some of the activity not accounted for throughout the experimental period may have been lost when plant roots were rinsed under the tap. Rinse water was not analyzed for activity. Regression analyses revealed the amount of activity accumulated in plants was directly proportional to cumulative water-use by the plants [$P = 7.55 \times 10^{-7}$; % accumulated = 0.001914*(cumulative water used) + 0.02895, $R^2 = 0.92$].

Analysis of the dissected plant tissues revealed accumulation of [^{14}C] primarily in the stems and leaves after 3 d exposure (Table 1). No accumulation was seen in the roots, indicating they serve primarily as a pathway for acropetal transport. This lack of accumulation in roots may have also been due to the fact that roots rarely exceeded 15% of total plant biomass, except after 7 d exposure. The leaves were the major sink for [^{14}C] from [^{14}C]simazine after 5 d.

Phytotoxicity results indicated that highest No Observable Effects Concentration (NOEC) and Lowest Observable Effects Concentration (LOEC) for parrotfeather exposed to simazine was 0.1 and 0.3 mg/L, respectively, for fresh weight gains and branch production. NOECs and LOECs for stem elongation were 0.3 and 1 mg/L, respectively, suggesting this endpoint was less sensitive than the other parameters tested. These ranges are included in the EC_{50} ranges reported for several aquatic photosynthetic organisms. Photosynthesis was inhibited 50% in filamentous algae at 0.222-0.949 mg/L, and at 0.949 mg/L simazine in nonfilamentous algae (O'Neal and Lembi 1983). Likewise, *Myriophyllum verticillatum* L., *Potamogeton pectinatus* L., and *Vaucheria dichotomya* Ag. (after Walz) senesced at simazine concentrations of 1 mg/L, while *Rhizoclonium heiroglyphicum* Kutz. and *Cladophora glomerata* (L.) Kutz. senesced at 0.5 mg/L (Fowler 1977). However, toxicity thresholds for our study were much higher than those reported for *Myriophyllum spicatum* L. in a more sensitive physiological assay (Salah et al. 1989). These researchers developed a bioassay for photosynthetic inhibitors in water and aqueous soil extracts using *M. spicatum* and reported that oxygen generation was reduced 50% at 20 µg/L simazine in water.

Based on these results, some caution is warranted when using parrotfeather for the phytoremediation of simazine. However, these tests represented a worst case scenario where other substrates capable of reducing bioavailability were not present. Under natural conditions in the field, significant portions of each pesticide may be sorbed to substrates or degraded by micro-organisms and sunlight. In the case of a gravel-based phytoremediation system designed for removing simazine from water, caution is still warranted due to the expected pesticide concentrations. The expected simazine concentration in 568 L of rinse water was calculated to range from 9.2-92.5 mg/L, respectively, assuming a moderate application rate, 378 L original mix volume, and a residual rinse volume ranging from 3.8 to 37.9 L between rinses. These concentrations far exceed the tolerance threshold for simazine. Since the toxicity of simazine can not be changed, a possible strategy for reducing toxicity risks may be through dilution with uncontaminated water.

One question that this research did not address is the metabolic fate of simazine once it is in the plants. This was not possible because of the destructive nature of combusting the samples. However, it is likely that some metabolism occurred. Burauel and Fuhr (1988) suggested that simazine was more readily degradable by microorganisms following plant metabolism. Likely metabolic products include hydroxy-simazine, mono- and di-dealkylated simazine, and various conjugates (Castelfranco et al. 1961, Funderburk and Davis 1963, Beynon et al. 1972, Esser et al. 1975, Akinymiju et al. 1983, Burnet et al. 1993).

Future work will evaluate the actual phytoremedial ability of parrotfeather in constructed wetlands at The Walker Course, Clemson, South Carolina. Additional ornamental plant species will also be evaluated for their phytoremedial abilities.

ACKNOWLEDGMENTS

The authors wish to thank the South Carolina Agriculture and Forestry Research System for funding and Novartis Crop Protection for their generous donations of materials. We also thank Vance Baird, Nihal Rajapakse, Sonja Maki, John Wells, Tom Fernandez, and Larry Addis for the use of their facilities and expertise. This publication is South Carolina Agricultural and Forestry Experiment Station contribution #4508 and Clemson Institute of Environmental Toxicology contribution #9905.

LITERATURE CITED

Ahrens, W. 1994. Herbicide handbook of the weed science society of America. Weed Science Society of America, Champaign, IL. pp. 270-274.

Akinyemiju, O., D. Dickmann, and R. Leavitt. 1983. Distribution and metabolism of simazine in simazine-tolerant and -intolerant poplar (*Populus* sp.) clones. Weed Sci. 31: 775-778.

Anderson, T. and J. Coats (eds.). 1994. Bioremediation through Rhizosphere Technology. ACS Symposium Series 563, American Chemical Society, Washington, DC. pp. 1-249.

Anderson, T., E. Guthrie, and B. Walton. 1993. Bioremediation in the rhizosphere. Environ. Sci. Technol. 27: 2630-2636.

Anonymous. 1995. Ciba Turf and Ornamental Sample Label and MSDS Guide. Ciba-Geigy, Greensboro, NC. pp. 145-148.

Beynon, K., G. Stoydin, and A. Wright. 1972. A comparison of the breakdown of the triazine herbicides cyanazine, atrazine and simazine in soils and in maize. Pestic. Biochem. Physiol. 2: 153-161.

Burauel, B. and F. Führ. 1988. The enhanced mineralization of simazine and bentazon in soil after plant uptake. Z. Pflanzenernaehr Bodenk-D 151: 311-314.

Burnet, M. W., B. R. Loveys, J. A. Holtum, and S. B. Powles. 1993. Increased detoxification is a mechanism of simazine resistance in *Lolium rigidum*. Pestic. Biochem. Physiol. 46: 207-218.

Castelfranco, P., C. L. Foy, and D. B. Deutsch. 1961. Nonenzymatic detoxification of 2-chloro-4,6-bis(ethylamino)-s-triazine (simazine) by extracts of *Zea mays*. Weeds 9: 580-591.

Correl, D. S. and H. B. Correl. 1975. Aquatic and Wetland Plants of Southwestern United States. 2 vols. Stanford University Press, Stanford, CA. pp. 1202-1208.

Davis, D., H. Funderburk, and N. Sansing. 1959. The adsorption and translocation of C14-labeled simazine by corn, cotton, and cucumber. Weeds 7: 300-309.

Esser, H., G. Dupuis, E. Ebert, C. Vogel, and G. Marco. 1975. "S-Triazines". *In:* P. Kearney and D. Kaufman, eds. Herbicides: Chemistry, Degradation, and Mode of Action. Marcel Dekkar, New York, NY. pp. 129-185.

Fowler, M. 1977. Laboratory trials of a new triazine herbicide (DPX 3674) on various aquatic species of macrophytes and algae. Weed Res. 17: 191-195.

Funderburk, H. and D. Davis. 1963. The metabolism of C14 chain- and ring-labeled simazine by corn and the effect of atrazine on plant respiratory systems. Weeds 11: 101-104.

Lund-Hoie, K. 1969. Uptake, translocation and metabolism of simazine in Norway spruce (*Picea abies*). Weed Res. 9: 142-147.

O'Neal, S. and C. Lembi. 1983. Effect of simazine on photosynthesis and growth of filamentous algae. Weed Sci. 31: 899-903.

Salah, A. S., S. W. O'Neal, M. A. Ross, and C. A. Lembi. 1989. Bioassay of photosynthetic inhibitors in water and aqueous soil extracts with Eurasian watermilfoil (*Myriophyllum spicatum*). Weed Sci. 37: 810-814.

Schnoor, J., L. Licht, S. McCutcheon, N. Wolfe, and L. Carreira. 1995. Phytoremediation of organic and nutrient contaminants. Environ. Sci. Technol. 29: 318A-323A.

Shone, M. and A. Wood. 1972. Factors affecting absorption and translocation of simazine by barley. J. Exp. Bot. 23: 141-151.

Shone, M. and A. Wood. 1974. A comparison of the uptake and translocation of some organic herbicides and a systemic fungicide by barley: I. Absorption in relation to physico-chemical properties. J. Exp. Bot. 25: 390-400.

Shone, M. and A. Wood. 1976. Uptake and translocation of some pesticides by hypocotyls of radish seedlings. Weed Res. 16: 229-238.

Shone, M., B. Bartlett, and A. Wood. 1974. A comparison of the uptake and translocation of some organic herbicides and a systemic fungicide by barley: II. Relationship between uptake by roots and translocation to shoots. J. Exp. Bot. 25: 401-409.

Wauchope, R. D., T. M. Buttler, A. G. Hornsby, P. W. M. Augustijn-Beckers, and J. P. Burt. 1992. The SCS/ARS/CES Pesticide Properties Database for Environmental Decision-Making. Rev. Environ. Contam. Toxicol. 123: 1-164.

Wichman, J. and W. Byrnes. 1975. Uptake, distribution, and degradation of simazine by black walnut and yellow poplar seedlings. Weed Sci. 23: 448-453.