

NOTES

Influence of Allelopathic Chemicals on Sprouting of Hydrilla Tubers¹

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INTRODUCTION

Allelopathy refers to biochemical interactions that take place among plants, and its effect depends on a chemical compound being added to the environment (8). The chemical may have an inhibitory or a stimulatory influence on plants, the nature and extent of which varies according to species.

The presence of allelopathic chemicals helps explain the growth of certain plants. In recent years, considerable attention has been devoted to evaluating the feasibility of using allelopathic compounds as a way to regulate the growth of certain plant species used for agricultural purposes (5,11). Management of the undesirable growth of some weed species appears feasible through allelopathy (8).

Oborn *et al.* (6) found during a 2-year period under greenhouse conditions that pondweeds (*Potamogeton* spp.) were eliminated from cultures planted with the spikerush (*Eleocharis acicularis* (L.) R. & S.) or with dwarf arrowhead (*Sagittaria subulata* (L.) Buch.). Since the spikerush and dwarf arrowhead produce shoots in the water column of only a few centimeters in height as compared to the pondweeds which may grow 2 to 4 m in height, competition for light does not appear to be a factor resulting in the poor growth of the pondweeds.

Frank and Dechoretz (3) used the leachate from containers planted with cultures of dwarf spikerush (*Eleocharis coloradoensis* Britt. Gilly) to reduce the production of new shoots of American (*Potamogeton nodosus* Poir.) and sago pondweed (*P. pectinatus* L.). Additional evidence of allelopathy with some of the spikerushes was produced by the study of Ashton, *et al.* (1) when a leachate was collected from exenic cultures of dwarf spikerush. This leachate contained a compound, or perhaps several compounds, with a molecular weight between 600 and 1000 that was phytotoxic to excised parts of sago pondweed and hydrilla (*Hydrilla verticillata* Royle).

Phenolic acids, alkaloids, coumarins, and quinones elicit allelopathic responses from some terrestrial plants

(8), but little is known of their influence on aquatic species. A study was initiated therefore with the objective of evaluating several of these compounds for their allelopathic potential on the sprouting and subsequent growth of hydrilla tubers. All of the compounds selected for this study have shown allelopathic influences on other plants (7,9,12).

MATERIALS AND METHODS

Experimental Conditions. Hydrilla tubers were collected from plants that had been cultured for 1 year or more under outdoor conditions. The tubers were washed to remove adhering debris, and then soaked with stirring in a 1.3% sodium hypochlorite solution for 20 minutes. After this they were placed in petri dishes or glass tubes and exposed to various concentrations of analytical-grade allelopathic compounds in distilled water. Both the molar concentrations and equivalent amounts in $\mu\text{g/ml}$ for the allelopathic compounds used in the experiments are listed in Table 1.

Tests conducted in petri dishes consisted of 25 tubers being placed in 30 ml of solution in a petri dish, and four dishes were used for each concentration. For tests in the glass tubes, 50 ml of each concentration used was placed in 20 separate tubes and a single tuber placed in each tube. With the glass tubes, 20 tubers were used for each treatment concentration. The control consisted of tubers placed in distilled water only.

Tubers were placed in the dishes or tubes, containing the desired concentration of allelopathic compound, under conditions of dim, indirect light. The dishes or tubes were wrapped with aluminum foil and placed in an incubator set at 25°C. After 48 hours, the aluminum foil was removed and the tubers exposed daily to 14 hours of light at 28 $\mu\text{E/m}^2 \times \text{s}^{-1}$. The solutions were replaced weekly; however, the treatment containers were not wrapped after the replacement of the solutions but the preparation and change of solutions were carried out in dim light. All experiments were repeated three times for a total of 300 tubers for each chemical concentration in the petri dishes and 60 tubers for each chemical concentration in the glass tubes.

Experiment 1. Hydrilla tubers were placed in petri dishes each containing distilled water (control) or 10^{-2} , 10^{-3} , 10^{-4} , 10^{-5} , 10^{-6} M concentrations of salicylic acid. The number of tubers that sprouted after 3 weeks exposure to these concentrations was determined.

Experiment 2. Hydrilla tubers were placed in petri dishes and exposed for 3 weeks to 10^{-2} and 10^{-3} M concentrations of hydroquinone, salicylic acid, umbelliferone, caffeic acid, p-hydroxybenzoic acid, m-coumaric acid, vanillic acid,

¹Contribution of the University of Florida's Fort Lauderdale Agricultural Research and Education Center. Published as Journal Series Number 7100 of the Florida Agric. Exp. Sta. Agricultural Research Service (ARS), Southern Region, South Atlantic, USDA, Cooperating.

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TABLE 1. INFLUENCE OF 11 ALLELOPATHIC CHEMICALS ON SPROUTING OF HYDRILLA TUBERS.^a

Allelopathic chemical	Concentration		Number of tubers sprouted (%)
	Molar	($\mu\text{g/ml}$)	
Control	0	0	90 g
Caffeic Acid	10^{-3}	180	85 fg
Caffeic Acid	10^{-2}	1,801	69 efg
Catechol	10^{-3}	110	89 g
Catechol	10^{-2}	1,101	0 a
m-Coumaric Acid	10^{-3}	164	90 g
m-Coumaric Acid	10^{-2}	1,641	16 d
Fumaric Acid	10^{-3}	116	86 fg
Fumaric Acid	10^{-2}	1,160	65 efg
Gallic Acid	10^{-3}	170	60 ef
Gallic Acid	10^{-2}	1,701	2 ab
Hydrocinnamic Acid	10^{-3}	142	88 g
Hydrocinnamic Acid	10^{-2}	1,418	0 g
Hydroquinone	10^{-3}	110	81 fg
Hydroquinone	10^{-2}	1,100	51 e
p-Hydroxybenzoic acid	10^{-3}	138	80 fg
p-Hydroxybenzoic acid	10^{-2}	1,381	1 a
Salicylic Acid	10^{-6}	0.138	81 fg
Salicylic Acid	10^{-5}	1.380	68 efg
Salicylic Acid	10^{-4}	13.80	53 e
Salicylic Acid	10^{-3}	138.0	6 bcd
Salicylic Acid	10^{-2}	1,380	0 a
Umbelliferone	10^{-3}	162	73 efg
Umbelliferone	10^{-2}	1,162	7 cd
Vanillic Acid	10^{-3}	168	85 fg
Vanillic Acid	10^{-2}	1,681	3 abc

^aValues followed by the same letter are not significantly different at the 5% level according to Duncan's Multiple Range Test. Each value is the mean of at least 300 tubers.

fumaric acid, gallic acid, hydrocinnamic acid, and catechol.³ Control tubers were placed in distilled water.

Experiment 3. Hydrilla tubers were placed in glass tubes and exposed to 10^{-2} and 10^{-3} M concentrations of gallic acid or hydroquinone. The number of sprouted tubers and shoot length were determined at the end of the 4-week culture period.

Statistical Analyses of Results. The Statistical Analyses System (SAS) software located at the North East Regional Data Center (NERDC) in Gainesville was used to analyze the data. Values for the percent germination of the tubers were converted to square roots, a standard transformation for percentages (10). However, the un-transformed means for the percent germination of the tubers are presented. Percent germination and shoot length data were statistically analyzed using randomized block design procedures with each time the experiment was conducted considered a block. Values for similar treatment concentrations for Experiments 1 and 2 were pooled. Duncan's Multiple Range Test was used to check for treatment differences (2).

RESULTS AND DISCUSSION

Culture conditions in this study were favorable as 90% of the hydrilla tubers in the distilled water (controls) sprouted (Table 1). This agrees with the data of Miller *et al.* (4) where 93% of the hydrilla tubers sprouted when placed under culture conditions of $12 \text{ microE/m}^2 \times \text{s}^{-1}$ of continuous light and 28°C .

All of the allelopathic compounds except caffeic acid and fumaric acid at a concentration of 10^{-2} M inhibited

sprouting of the hydrilla tubers as compared to the control (Table 1). Furthermore, at this concentration no tubers sprouted in the presence of catechol, hydrocinnamic acid, or salicylic acid. With p-hydroxybenzoic acid, gallic acid, vanillic acid, umbelliferone, and m-coumaric acid at the 10^{-2} M concentration, 16% or less of the tubers sprouted. At this concentration of 10^{-2} M, 51 to 69% sprouting of hydrilla tubers occurred for the propagules exposed to hydroquinone, fumaric acid, and caffeic acid.

Only gallic acid and salicylic acid at the 10^{-3} M concentration reduced sprouting as compared to the control. At this concentration, 60% sprouting occurred in the gallic acid treatment and 6% for the salicylic acid. Furthermore, the number of tubers which sprouted at the 10^{-4} concentration of salicylic acid was almost half that of the controls. No significant difference in sprouting was observed for tubers in the 10^{-5} and 10^{-6} concentration of salicylic acid as compared to the control.

In Experiment 3, shoot length of the control tubers averaged 3.2 cm after 4 weeks. Shoot length of tubers in the 10^{-3} M treatment of hydroquinone was 3.3 cm and not significantly different from the control, but a 46% reduction in shoot length was observed for tubers in the 10^{-2} M concentration of this chemical. With gallic acid, shoot length was shorter than the control for both treatments with measurements of 1.7 cm for the 10^{-3} M and 0.3 cm for the 10^{-2} M.

These data show the potential of allelopathic compounds to inhibit or reduce sprouting of hydrilla tubers and subsequent growth of the shoots. With the exception of salicylic acid, the usefulness of these compounds in the management of hydrilla appears rather limited because of the large amount, greater than 1,000 ppm, required to reduce sprouting. Additional studies are needed to further evaluate the potential of salicylic acid and other allelopathic compounds for use in the management of problem aquatic weed species.

ACKNOWLEDGMENTS

The author wishes to thank Ms. Maria Bravo and Ms. Joanne Korvick for their technical assistance in this study. This material is based upon work supported by the University of Florida, Institute of Food and Agricultural Sciences, and the U.S. Department of Agriculture, ARS, under Cooperative Agreement No. 58-7B30-3-570.

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