

Injection of Nutrients Into Sand Rooting Media for Culture of Dioecious Hydrilla¹

DAVID L. SUTTON²

ABSTRACT

Growth studies were conducted outdoors with dioecious hydrilla (*Hydrilla verticillata* (L.f.) Royle) cultured in concrete tanks filled with flowing pond water. Nutrients were supplied to the root zone of hydrilla plants either by injecting Hoagland's nutrient solution from the surface of the water through tubing connected to a silica glass air diffuser located in a sand rooting media, or by placing a layer of fertilizer in the sand. The fertilizer layer consisted of either Vigoro or a combination of Osmocote, Esmigran and dolomite. Each culture container was surrounded with a large-mesh plastic netting and window screening to form a water column 80 cm in height by 380 cm in surface area which enclosed growing hydrilla plants. Growth of hydrilla was similar for plants cultured for 8 wk with Vigoro or Osmocote plus Esmigran and dolomite. High amounts of nitrogen in Hoagland's nutrient solution, 187.50 mg per injection, severely reduced growth of hydrilla but growth improved with reduced amounts of nitrogen. This study shows the potential of an

injection system to evaluate various nutrients or other chemicals placed in the root zone on growth of hydrilla.

Key words: aquatic plants, fertilizer, hydrosol, sediments, propagules, tubers.

INTRODUCTION

Hydrilla causes serious submersed weed problems in many tropical and subtropical areas (Cook and Lüönd 1982, Pieterse 1981). Its ability to conduct photosynthesis under low light (Van *et al.* 1976), produce new plants from each node (Langeland and Sutton 1980) in addition to those that can form from turions and tubers (Haller 1967), and form a canopy just below the surface of the water that can shade other submersed plants (Haller and Sutton 1975) are some of the characteristics that allow hydrilla to colonize and grow as a monoculture by replacing indigenous plants in a variety of aquatic habitats. However, the influence of macronutrients and micronutrients in water and substrate on growth of hydrilla is not clearly understood.

Sutton (1986) found dry weight of hydrilla cultured in sand plus fertilizers to be dependent on the concentration of fertilizer in the root zone and was from 6 to 14 times that of plants cultured in sand alone. Of nine nutrients measured in plant tissue from hydrilla cultured in sand amended with fertilizers (Sutton 1986), only phosphorus in both shoots and roots was dependent on the level of fertilizer in the root zone.

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²Professor, University of Florida, IFAS, Fort Lauderdale Research and Education Center, 3205 College Ave., Fort Lauderdale, FL 33314.

MATERIALS AND METHODS

Water surrounding hydrilla shoots was found to be the primary source for potassium (Barko 1982), but sediments were the primary source for two other macronutrients, nitrogen and phosphorus (Barko 1982, Barko *et al.* 1991, Steward 1984). Studies by Barko (1982) and Steward (1984) stressed the need for research to evaluate the influence of sediment composition and sampling procedures on the growth of submersed plants in general.

A method for injecting nutrients in the root zone of hydrilla plants was developed to provide additional information on the influence of various nutrients on growth of hydrilla. The method employed an air diffuser placed in the root zone that supplied Hoagland's nutrient solution to the sand rooting media by means of a syringe and tubing. Dry weight of plants cultured by this method was compared with hydrilla plants grown with commercially available fertilizers known to promote growth of hydrilla.

Hydrilla was cultured outdoors at the Fort Lauderdale Research and Education Center (FLREC), which is located 26°05'N and 80°14'W, in concrete tanks (6.2 m in length by 3.1 m in width) filled with pond water. Pond water, from the same source as described by Steward (1984), flowed into the tanks at the surface of one end and out from bottom drains at the other at a rate which allowed for a complete exchange of water every 24 hr. Nutrient treatments were arranged in rows perpendicular to the flow of water. Treatments were assigned at random within a row, and four rows were used for each culture period.

For all culture periods, nutrients were supplied to the root zone of hydrilla plants by either placing a layer of fertilizer 7.6 cm below the surface of the sand or injecting nutrient solution (Figure 1). The top of the air diffuser was placed the

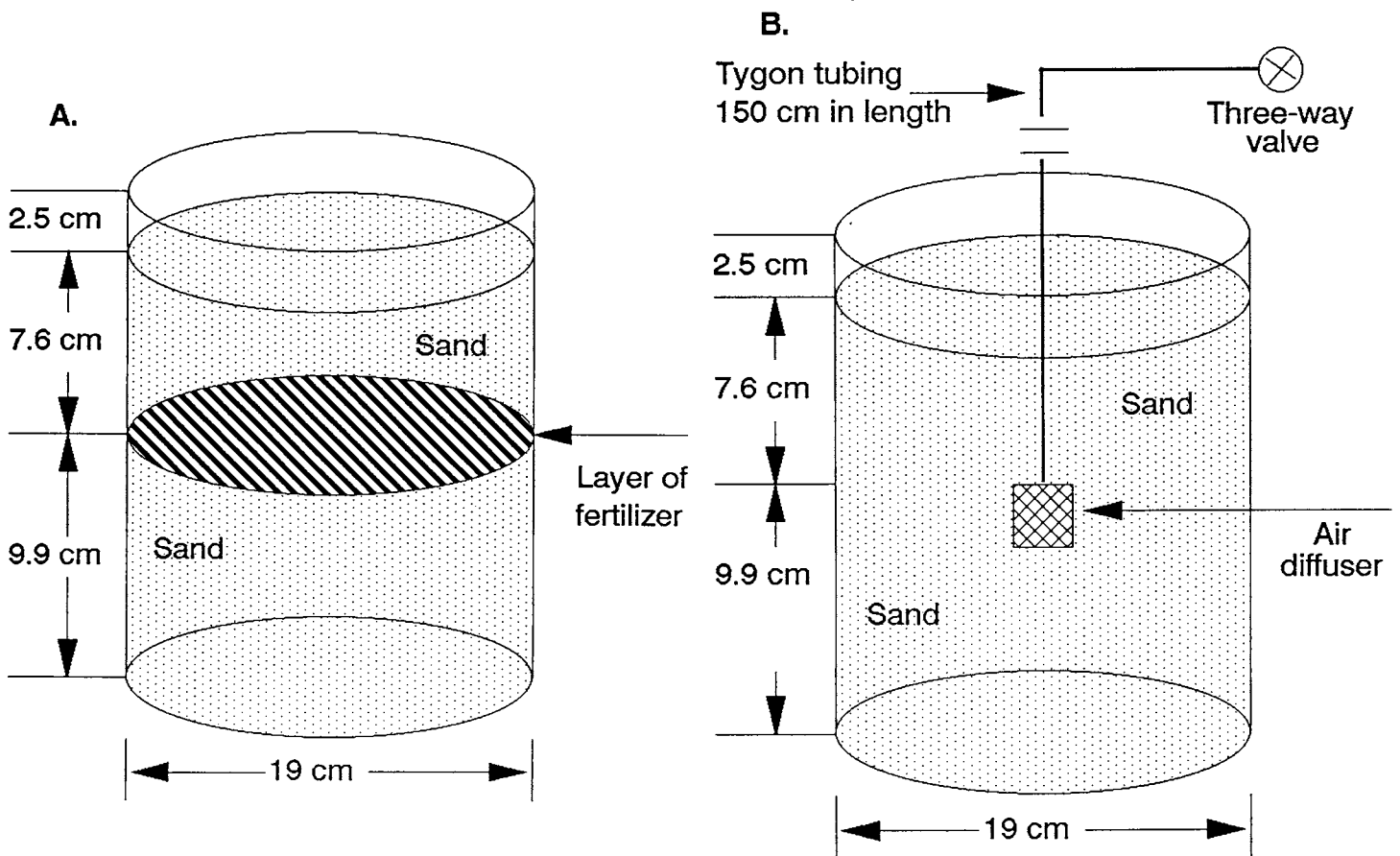


Figure 1. Diagrammatic representation of culture containers showing placement of nutrients in sand rooting media used for culture of hydrilla in outdoor tanks filled with flowing pond water. A. (Left) Location for layer of Osmocote or Vigoro fertilizers. B. (Right) Location of air diffuser for injection of Hoagland's nutrient solution.

same distance from the surface of the sand as the layer of fertilizer. The air diffuser was constructed of silica glass with dimensions of 1.5 cm in length by 1.5 cm in width by 1.5 cm in height. A three-way valve at the top of the tubing allowed for a solution of nutrients to be injected without air being pushed in the root zone.

Nutrient solutions were injected with a syringe three times each week on Monday, Wednesday, and Friday. This resulted in a total of 24 injections during an 8-wk culture period. Each nutrient solution injection consisted of 50 ml of distilled water which contained either 15.63 mg, 31.25 mg, 62.50 mg, or 125.00 mg of nitrogen; 27.25 mg of phosphorus; 0.6 mg of iron (Fe) supplied by Sequestrene 330Fe³; and the amounts of other nutrients normally contained in 50 ml of full-strength Hoagland's nutrient solution (Hoagland and Arnon 1950). Injections of 15.63 mg, 31.25 mg, 62.50 mg, or 125.00 mg of nitrogen resulted in total amounts of 375 mg, 750 mg, 1,500 mg, or 3,000 mg of nitrogen, respectively, added to the sand in the culture containers during the hydrilla growth period. After the nutrient solution was injected, 50 ml of distilled water was used to flush the tubing and air diffuser. Four containers (Figure 1) were injected with each different nutrient treatment of nitrogen for each culture period. Commercially available fertilizers were supplied by adding either 75 g of Vigoro⁴, an all-purpose 6-10-4 granular formulation with micronutrients, or a combination of 25 g of Osmocote⁵ (18-6-12), 0.7 g of Esmigran (micronutrients in sustained-release form) and 4.2 g of dolomite (mined material containing 55% calcium as CaCO₃ and 30% magnesium as MgCO₃) (hereinafter referred to as Osmocote) to each of four containers for each culture period.

In a preliminary study, when hydrilla plants were injected three times a week for 8 wk with each injection consisting of 187.50 mg of nitrogen and 27.25 mg of phosphorus, sprouted tubers of hydrilla did not grow well. Thus, the starting concentration for Experiment 1 was reduced to 125 mg of nitrogen per injection.

Tubers were collected from stock hydrilla plants grown at the FLREC. Stock plants were originally collected from Lake Okeechobee. Tubers were sprouted in pond water until

shoots reached approximately 12 cm in length and root initiation had begun. For each culture period, four sprouted tubers were placed with three to four nodes below the surface of the sand of each container.

After planting sprouted tubers, each container was surrounded with large mesh plastic netting and window screening to form a water column 80 cm in height by 380 cm² in surface area to enclose growing hydrilla plants. Screening enclosed each container with no excess space between the container and screen.

The study consisted of two separate culture periods: (Experiment 1) 11 September to 6 November 1990 and (Experiment 2) 26 November 1990 to 21 January 1991. At the end of each culture period, hydrilla plants were removed from the tanks and hydrilla shoots were cut at the sand surface and washed with pond water to remove algae, sand, and other debris, then dried to a constant weight in a forced-air drying oven at 60C. Below-ground biomass including roots, rhizomes, stem fragments (hereinafter referred to as roots) and tubers in each culture container was washed with pond water to remove sand, fertilizer, and any other adhering debris, and dried at 60C. Tubers were counted when present.

Water temperatures were recorded during each culture period. A maximum/minimum thermometer was placed 30 cm below the surface of the water and temperature was recorded 5 days a week, generally at 4:00 p.m. Water temperature for each culture period was calculated as a daily mean of maximum and minimum values obtained.

Once hydrilla plants reached the surface of the water, an emulsifiable concentrate of malathion (0,0-dimethyl dithiophosphate of diethyl mercaptosuccinate) was added to achieve a concentration of 1.0 ppm in the tank as necessary to control feeding activity of the herbivorous moth *Parapoynix diminutalis* Snellen.

The Statistical Analyses System (SAS)⁶ software designed for use on personal computers was used to analyze plant dry weight following Analysis of Variance (ANOVA) procedures. For analysis purposes, hydrilla dry weights were converted to natural logs and tuber count data were transformed by using the square root of the count plus one (Steel and Torrie 1960), but the nontransformed values are presented. The Waller-Duncan Bayesian LSD procedure was used for mean separation.

RESULTS AND DISCUSSION

Daily water temperature values averaged 28.7C with a maximum of 32.0C and a minimum of 21.0C for the 11 September to 6 November 1991 culture period. For the 26 November

⁶SAS Institute Inc., Cary, NC 27512-8000.

³Sequestrene 330Fe is manufactured by CIBA/GEIGY Corp., Greensboro, NC 27419. Mention of a trademark or a proprietary product does not constitute a guarantee or warranty of the product by the University of Florida or the USDA and does not imply its approval to the exclusion of other products that also may be suitable.

⁴Vigoro® All-Purpose (6-10-4) is manufactured by Vigoro Industries, Inc., Fairview Heights, IL 62208.

⁵Osmocote with an 8- to 9-month release time and Esmigran are manufactured by Grace Sierra Horticulture Products Company, Milpitas, CA 95035; and Dolomite (Soil Doctor) by Soil Doctor, Inc., Crystal River, FL 32629.

1990 to 21 January 1991 culture period, daily water temperature values averaged 24.2C with a maximum of 28.0C and a minimum of 18.5C. Mean water temperatures for both culture periods are in the range for good growth of hydrilla as suggested in the study by Van *et al.* (1978).

Dry weight of hydrilla was similar for plants cultured with Osmocote or Vigoro fertilizers, but the magnitude of dry weight varied with culture period (Tables 1 to 2). Total dry weight of pooled values for Osmocote and Vigoro for Experiment 1 was 64% higher than for Experiment 2. This ranking is in the same order as that for mean water temperatures for the two culture periods.

In one of the first studies using Osmocote, a commercially available controlled release fertilizer, for culture of hydrilla in sand rooting media, Sutton (1986) reported that growth of hydrilla under south Florida conditions with this fertilizer depended on temperature and concentration of the fertilizer. Based on findings of Harbaugh and Wilfret 1981, Sutton (1986) concluded that temperature, as related to rate of release of nitrogen and phosphorus in the Osmocote prills, was probably a major factor in the observed differences in hydrilla growth. However in the experiments with Osmocote and Vigoro, a granular material not formulated for nutrient release related to temperature, growth of hydrilla was similar. These results indicate that water temperature differences are influencing metabolic processes related to hydrilla growth rather than influencing the rate of nutrient release from Osmocote.

Photoperiod was conducive for tuber formation for both culture periods (Van *et al.* 1978 and Sutton *et al.* 1992). Interestingly, no tubers were produced by plants cultured with Vigoro fertilizer. It is not known why plants cultured with Vigoro fertilizer did not produce tubers during these periods in contrast to tubers produced by plants cultured with Osmocote.

Although concentrations of nitrogen for the Vigoro and Osmocote fertilizers were the same, the amounts of phosphorus, potassium, and micronutrients were not present in the same amounts due to differences in formulations of these fertilizers.

In general, dry weight of hydrilla cultured with Osmocote or Vigoro was higher than plants injected with Hoagland's nutrient solution with various amounts of nitrogen (Tables 1 to 2). Since only nitrogen, supplied by KNO^3 and $Ca(NO_3)_2 \cdot 4H_2O$, varied in the Hoagland's nutrient solution treatments, it appears that the amounts of nitrogen added to the root zone was a major factor influencing growth of hydrilla under these conditions. Although other nutrients in the Hoagland's nutrient solution were not varied, their interaction with the various amounts of nitrogen may have influenced growth of hydrilla. Additional studies will be needed to evaluate the influence of these other nutrients on growth of hydrilla.

The preliminary study showed that when hydrilla plants were injected with Hoagland's nutrient solution at a rate

TABLE 1. DRY WEIGHT OF HYDRILLA AFTER GROWTH OUTDOORS IN SAND AMENDED WITH FERTILIZER OR INJECTED WITH HOAGLAND'S NUTRIENT SOLUTION. SPROUTED TUBERS WERE PLANTED 11 SEPTEMBER 1990, AND PLANTS WERE ALLOWED TO GROW UNTIL 6 NOVEMBER 1990.

Total amount of nutrient (mg) ^a		Plant dry weight (g) ^b			Number of tubers
Nitrogen	Phosphorus	Shoots	Roots	Total ^c	
Osmocote					
4,500	654	12.26 a	1.35 a	13.61 a	0 a
Vigoro					
4,500	3,273	12.45 a	1.18 a	13.63 a	0 a
Hoagland's nutrient solution					
3,000	654	5.36 c	0.68 b	6.05 c	1 a
1,500	654	8.10 b	1.10 a	9.26 bc	1 a
750	654	9.78 ab	1.16 a	11.05 ab	3 a

^a Total amounts of nitrogen and phosphorus in Osmocote or Vigoro applied as a layer in sand rooting media prior to planting. For Hoagland's nutrient solution, one twenty-fourth of the total amounts of nitrogen and phosphorus were injected three times each week during the culture period.

^b Values for plant dry weight in a column followed by the same letter are not significantly different at the 5% level according to Waller-Duncan Bayesian LSD Procedure. Each value is the mean of plants from four culture containers.

^c Includes weight of shoots, roots, and tubers.

TABLE 2. DRY WEIGHT OF HYDRILLA AFTER GROWTH OUTDOORS IN SAND AMENDED WITH FERTILIZER OR INJECTED WITH HOAGLAND'S NUTRIENT SOLUTION. SPROUTED TUBERS WERE PLANTED 26 NOVEMBER 1990, AND PLANTS WERE ALLOWED TO GROW UNTIL 21 JANUARY 1991.

Total amount of nutrient (mg) ^a		Plant dry weight (g) ^b			Number of tubers
Nitrogen	Phosphorus	Shoots	Roots	Total ^c	
Osmocote					
4,500	654	7.47 a	1.00 a	8.71 a	8 a
Vigoro					
4,500	3,273	7.30 ab	0.56 bc	7.86 ab	0 c
Hoagland's nutrient solution					
3,000	654	2.09 d	0.42 c	2.52 d	1 c
1,500	654	3.72 c	0.68 ab	4.47 b	3 b
750	654	5.43 b	0.88 a	6.53 ab	7 a
375	654	5.35 b	0.75 ab	6.22 b	3 b

^a Total amounts of nitrogen and phosphorus in Osmocote or Vigoro applied as a layer in sand rooting media prior to planting. For Hoagland's nutrient solution, one twenty-fourth of the total amounts of nitrogen and phosphorus were injected three times each week during the culture period.

^b Values for plant dry weight in a column followed by the same letter are not significantly different at the 5% level according to Waller-Duncan Bayesian LSD Procedure. Each value is the mean of plants from four culture containers.

^c Includes weight of shoots, roots, and tubers.

which resulted in a total of 4500 mg of nitrogen being added to the root zone, the same amount as that contained in the Osmocote or Vigoro fertilizers, sprouted tubers grew very little, and roots at time of harvest were heavily encrusted with a salt-appearing material. When hydrilla was harvested after 8 wk of growth, plants treated with 24 injections of 187.50 mg of nitrogen for a total of 4500 mg of nitrogen averaged 86% less in dry weight than plants cultured with Osmocote or Vigoro.

A decrease in amounts of nitrogen from 3000 to 750 mg injected in the sand root media resulted in an increase in hydrilla dry weight (Table 1). Also, dry weight of plants injected with 750 mg of nitrogen was similar to plants cultured with Vigoro or Osmocote. In Experiment 2, dry weight of plants cultured with a total of 375 mg of nitrogen was similar to hydrilla plants for the 750-mg rate of nitrogen (Table 2).

These data show a method to inject nutrients in the root zone of hydrilla. Additional studies are needed to inject nutrients in the root zone of hydrilla based on measurements of nutrients in lake sediments. In this way it may be possible to predict potential hydrilla growth based on nutrient content of various sediments.

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