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Transplanting and Survival of the Seagrass *Halodule wrightii* Under Controlled Conditions

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TRANSPLANTING AND SURVIVAL OF THE SEAGRASS *Halodule wrightii* UNDER CONTROLLED CONDITIONS[1]

The importance of seagrasses as primary producers, as a habitat for a variety of marine organisms and for controlling erosion and the deposition of sediments in estuarine systems is well documented (Burkholder, *et al.,* 1959; Pomeroy, 1960; Odum, 1963, and others). Also, the importance of attempting to reestablish seagrasses in dredged or otherwise disturbed coastal areas has been recognized (Phillips, 1960; Strawn, 1961; Sykes, 1967).

Studies attempting to transplant different seagrasses either in the field or in the laboratory have met with varying degrees of success (Fuss and Kelly, 1969; Kelly *et al.,* 1971; Thorhaug, 1974; van Breedveld, 1975; Phillips, 1976).

An area of controversy in transplanting seagrasses is the use of root stimulants. Usually some concentration of naphthalene acetic acid (NAPH) is applied to the plant's root-rhizome system before transplanting. Kelly *et al.* (1971) in their work with *Thalassia testudinum* found that soaking short shoots for one hour in a 10% solution of NAPH resulted in 100% survival when construction rods were used as anchors. They felt that the use of NAPH was one of the main factors contributing to transplant success. However, van Breedveld (1975) found that the use of 5% NAPH solution with *T. testudinum* resulted in 90-100% mortality. He also found 100% mortality of *T. testudinum* apices dipped in 10% NAPH in laboratory studies and consequently did not recommend the use of NAPH at 5-10% concentrations.

The type of substrate used in transplanting seagrasses is also an important factor to be considered. Fuss and Kelly (1969) were able to grow *T. testudinum*

with some success and *Halodule wrightii* with minor success under artificial conditions. Their sprigs were planted in aquaria and tanks containing washed builders sand over a layer of fine gravel. Thorhaug (1974) had moderate success growing *T. testudinum* from seedlings under artificial conditions using sand and gravel as the substrate, but was quite successful in an area which had previously supported extensive *T. testudinum* beds, thus stressing the need for compatible sediment types. Van Breedveld (1975) and Kenworthy and Fonseca (1977) also stressed the need for sediment compatability when transplanting *T. testudinum* and *Zostera marina,* respectively. McRoy and McMillan (1977) and Patriquin (1972) have also emphasized the role of sediments in acting as sources of nutrients to the plants via their root-rhizome system.

MATERIALS AND METHODS

For this study we used low concentrations of NAPH as a root-rhizome stimulant and sediment collected from *H. wrightii* beds from the Indian River as the transplant substrate. This sediment is comprised of 95-98% sand through the upper 15 cm. A thorough description of these sediments is found elsewhere (Zimmermann, 1980).

The Indian River is a narrow estuarine-lagoon system which extends approximately 200 km along the east coast of Florida. A more complete description of the Indian River system is reported by Gilmore (1977). Of the seagrasses reported from this area, *Halodule wrightii* Ascherson and *Syringodium filiforme* Kützing comprise more than 90% of the distribution. *Thalassia testudinum* Banks ex König and Sims and *Ruppia maritima* L. each comprise 3%. The presence of *Halophila engelmanni* Ascherson and the recently described species *Halophila*

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johnsonii Eiseman (Eiseman and McMillan, 1980), in the Indian River, although not found in abundance, has been documented and comprises the remainder of the seagrass population (Thompson, 1978).

Halodule wrightii was collected on 28 November 1978 using a post hole digger which took a 15 x 15 cm plug of grass and sediment. The sediment was then removed by immediately sieving through a 1 mm plastic mesh. Roots and rhizomes were gently washed free of sediment using sea water. Most of the grass collected consisted of a single plant complex with extensive root-rhizome systems. These plants were placed in a cooler containing seawater until they were returned to the laboratory a few hours later. Extra sediment was collected from an area which had previously supported a *H. wrightii* community.

The concentrations of NAPH selected for this study were based on the results of van Breedveld (1975). He found that the use of Root Dip®, a commercial brand root hormone which contains only 0.05% NAPH, produced higher survival rates among transplanted *T. testudinum* than the 5 and 10% solutions recommended by Kelly *et al.* (1971). We simply extended the concentrations, using 0.05, 0.1, 0.5 and 1.0% NAPH (J.T. Baker) prepared the day before the collection of seagrass samples. These NAPH solutions were prepared in artificial seawater (30.0 g NaC*l*, 10.0 g MgS0$_4$•7H$_2$0, and 0.04 g NaHC0$_3$/liter deionized water).

The *H. wrightii* transplants were divided into five approximately equal groups. The roots and rhizomes from each transplant of each group were soaked for two hours in the various NAPH solutions. The control group was soaked in seawater.

A transplant consisted of plant complexes with both single and multiple erect leaf bearing shoots and a rhizomal axes with at least one root bearing internode.

Multiple shoots refers to axes with two to five erect shoots.

Twenty transplants from each treatment were then carefully planted into separate 24-liter aquaria containing 5 cm of sediment. Rhizomal axes with 10 single erect leaf-bearing shoots constituted one row, while 10 multiple erect leaf-bearing shoots constituted the second row. Replicate tanks were used for each treatment including the control for a total of 10 tanks each containing 20 transplants.

For better thermal stability, the aquaria were then partially submerged in a 0.5 x 2.3 x 0.8 m concrete tank supplied with running seawater. Each aquarium was supplied with a surface stream of water at a rate of 1.5 l/minute (Figure 1). Tanks and grasses were cleaned weekly by hand in an effort to keep epiphytic growth under control. No chemicals were used.

Salinity and temperature data were recorded throughout the study using a refractometer and bucket thermometer. Mean monthly salinities (morning readings) ranged from 24.0‰ in January to 33.0‰ in April while the mean monthly temperature (morning readings) ranged from 17.4° C in January to 30.5° C in July.

In January, 1980, the transplants (leaves, roots, and rhizomes) were carefully removed from the tanks. Leaf number, length and width as well as dry weight measurements of leaf and root-rhizome systems were recorded.

RESULTS

The first few months after transplanting only cursory monitoring was performed. This period was set aside as a time for the transplants to stabilize. During this time (December 1978 - March 1979), plants in the control aquaria (untreated) generally showed more leaf growth than the treated plants.

The 0.05, 0.1 and 0.5% NAPH treated *H. wrightii* transplants showed noticeable

leaf growth during April 1979. In late May and early June, the transplanted *H. wrightii* treated with the higher concentrations of NAPH (0.5 and 1.0%) demonstrated increased leaf growth and new leaf-bearing shoots. Also during this time (May-June) the control transplants declined.

Survival rates were highest in transplants treated with 0.5 and 1.0% NAPH. After seven months 87% of the plants had survived compared with 38-39% survival of the lower NAPH concentrations and control groups. One month later (July 1979), the survival rate of the 0.5 and 1.0% NAPH treated plants had dropped to 74%, but was still much higher than the other groups. The test for equality of percentages (Sokal and Rohlf, 1969) indicated (see Table 1) that the plants treated with 0.5 and 1.0% NAPH exhibited significantly greater survival rates than those treated with lower concentrations or the untreated group ($P < 0.05$).

Epiphytic growth on the plants was somewhat of a problem during the length of the study. Examination of leaf scrapings indicated five dominant diatoms. They were *Melosira moniliformis*, *Grammatophora marina*, *Nitzschia paradosa*, *Navicula* sp. and *Cymbella* sp. The macroepiphyte *Microcoleus lyngbyaceus* (blue-green) was also prevalent throughout the study. It occurred in large numbers on the grass and the aquaria. Observations on *H. wrightii* in the natural environment did not indicate the large concentrations of *Microcoleus* found in the closed system. This difference may be due to the release of nutrients caused by increased water circulation in the aquaria and/or lack of macroepiphytic grazers in the aquaria.

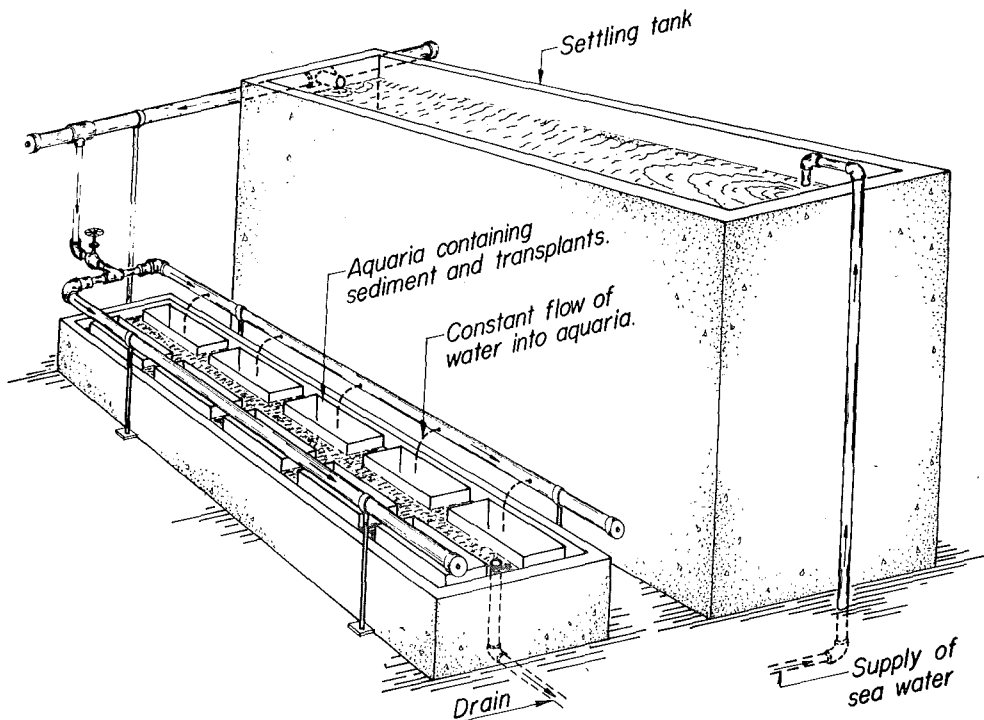Field observations on the condition of

Figure 1. Diagrammatic illustration of aquaria containing NAPH treated *Halodule wrightii* and control transplants.

TABLE 1. Percent survival of *Halodule wrightii* treated with varying concentrations of NAPH and the control group containing no NAPH.

	Control		NAPH							
			0.05%		0.1%		0.5%		1.0%	
Aquarium #	1	2	3	4	5	6	7	8	9	10
Number of *Halodule* short shoots with attached rhizomes 30 Nov. 1978	20	20	20	20	20	20	20	20	20	20
Individuals surviving after seven months June 1979	12	3	3	9	9	10	20	20+	20+	10
% Survival	38		30		48		100**		75**	
Individuals surviving after eight months July 1979	10	2	1	10	8	10	9	20	20	10
% Survival	30		28		45		73**		75**	

** Indicates significantly greater survival ($P < 0.05$).

H. wrightii plants in the Indian River coincided closely with observations of the transplants. In late fall growth decreases. There is usually a reduction in October followed by a slight recovery before declining to a minimum in January-February. During March and April there is noticeable leaf growth and from May to September the greatest biomass occurs (Eisman, unpublished data).

After the study was terminated, *Halodule wrightii* was found to be surviving in all the NAPH treated tanks, but in only one of the two control tanks.

Biomass data is found in Table 2. In most cases rhizome biomass was approximately twice that of leaf biomass. Increased leaf biomass was observed in the one tank containing the surviving untreated *H. wrightii* transplants, in some cases better than those treated with NAPH. This was not expected, but leaf length and width as well as rhizome comparisons of the control plants versus NAPH treated plants yielded results indicating longer and wider leaf lengths and more developed rhizomes of the NAPH treated plants.

Leaf length comparisons showed no significant difference between the control and the 0.05% NAPH treatment (paired t

statistic, $P < 0.05$). However, significant differences in leaf length were evident between the control plants ($\bar{x} = 8.2$ cm) and the other treatments (0.1% NAPH, 11.9 cm; 0.5% NAPH, 12.9 cm; 1.0% NAPH, 11.0 cm).

Leaf width measurements indicated the leaf widths of NAPH treated plants were between 1.5 and 2.0 mm while the majority of the control plants were 1.0 mm or less.

Examination of the surviving control plant's rhizome system indicated poor development. No branching was evident and while several rhizomes were present, the average length was 2.5 cm. The NAPH treated plants were better developed. The majority of the 0.05% NAPH treated rhizomes were from 4-12 cm long, with others as long as 29 cm, while the 0.1% NAPH treated rhizome lengths ranged between 7 and 10 cm, with two longer rhizomes (12.8 and 16 cm) which contained many branches.

Rhizome development in the 0.5% NAPH treated plants was also pronounced. The mean length of these rhizomes was 9.5 cm, some with extensive branching. The 1.0% NAPH treated rhizomes were not as developed as the 0.5% treatment, but branching rhizomes were still very much in evidence.

TABLE 2. Biomass (grams dry weight of leaf and rhizomes) of control and NAPH treated *Halodule wrightii* (January, 1980).

Type	Total (g)	Leaf (g)	Rhizome (g)	% Leaf	% Rhizome
Control	1.023	0.3530	0.6700	34.5	65.5
0.05% NAPH	0.8866	0.2551	0.6315	28.8	71.2
0.1% NAPH	0.2400	0.0919	0.1481	38.3	61.7
0.5% NAPH	1.3663	0.4668	0.8996	34.2	65.8
1.0% NAPH	0.4681	0.1515	0.3266	30.2	69.8

DISCUSSION

The similarity of the growth cycles of transplated *Halodule wrightii* treated with NAPH to undisturbed *H. wrightii* growing in the field indicates that the concentrations of NAPH used in this study did not affect the plants normal growth cycle. The elongate rhizomal axes noted in the NAPH treated plants indicate new meristematic growth in the rhizome system, while the original transplants had rhizomal axes not more than 2-3 cm long. Untreated rhizomes displayed little development. Transplants treated with 0.5% NAPH exhibited much· better leaf growth and rhizome development than the untreated plants. Although the other transplants treated with varying concentrations of NAPH demonstrated increased leaf and rhizome development, their final number of surviving plants was less than the 0.5% NAPH treated plants which consequently influenced the biomass data.

The use of low concentrations of NAPH did not result in plant morality, but in many cases enhanced growth. While the role of NAPH in transplanting seagrasses under field conditions needs further evaluation, this study indicates that the hormone, coupled with sediment compatibility, can be used·with moderate success in transplanting *H. wrightii* to aquaria systems.

ACKNOWLEDGMENTS

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