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Culture Studies of *Halodule wrightii* Aschers. Edaphic Requirements

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Abstract

Growth rates of *Halodule wrightii* Aschers. were compared during laboratory culture on artificial sediments containing varying amounts of soluble sulfide and iron, but similar amounts of nitrogen, phosphorus, and manganese. These experiments documented that moderate levels of H_2S (ca 0.5 mM) and very low levels of soluble Fe ($0.5-2.0 \mu g ml^{-1}$) in the sediment water correlated with maximal *Halodule* growth rates. Interstitial water-soluble Fe above $20 \mu g ml^{-1}$ appeared to inhibit *Halodule* growth, possibly by interfering with sulfur metabolism or limiting the availability of phosphorus to the plants. Excess phosphate, but not Mn, added to the sediments containing low H_2S levels counteracted some degree of Fe inhibition. Sediment H_2S concentrations greater than 1 mM were also correlated with decreased growth rates, verifying that a poised level of H_2S between 0.2–1.0 mM favored *Halodule* growth. The methodology described herein is proposed for laboratory culture studies of other seagrasses.

Introduction

Measurements of field conditions (Pulich 1982) revealed that sediment steady state pools of free H_2S , acid-volatile sulfide (FeS), and water-soluble Fe and Mn correlated with growth of shoalgrass (*Halodule wrightii* Aschers.) into unvegetated sediments. A positive correlation with shoalgrass production was found for sulfide (H_2S or organic-SH) and Mn^{2+} , while Fe appeared as a negative edaphic factor when interstitial water concentrations exceeded $1 \mu g Fe$ per ml. A buildup of low levels of sediment sulfide during *Halodule* colonization was postulated as necessary to condition the sediments and control soluble Fe (and other heavy metals) levels by complexing with Fe^{2+} , thereby converting it to insoluble FeS.

Excess sediment, soluble Fe^{2+} may be potentially toxic to *Halodule* by similar mechanisms described for rice (Ponnamperuma 1955, Tanaka *et al.* 1968, Howeler 1973), sugar cane (Clements *et al.* 1974), and various legumes (Ambler and Brown 1969, Brown and Jones 1977, Foy *et al.* 1978). These studies have documented that excess Fe interferes with Mn, P, and K uptake and translocation, as well as with metabolism of other trace metals. Another mechanism for Fe toxicity, involving interference with sulfur metabolism, appears possible, if high, tissue organic-S is required in some special process by plants like *Halodule*.

In order to test this hypothesis about *Halodule* edaphic requirements, a series of growth experiments were performed under laboratory conditions. Clean, organic-free sand was used to grow *Halodule* under conditions of controlled sulfate-reduction with known amounts of sediment, soluble Fe. This paper describes the technique developed for controlling the sediment chemical environment and its application to laboratory culture of *Halodule*.

Methods and Materials

Laboratory Culture System

An underground sprinkler system allowed manipulation of sediment conditions without disturbing the overlying water column (Fig. 1). The system consisted of a vertical 15 mm diam. PVC standpipe section, attached with a "T" joint to two sections of underground PVC sprinkler pipe. Pairs of 2 mm holes were drilled every 6 mm in the sprinkler pipe. Rubber stoppers were placed in the ends of the sprinkler pipe. Sprinklers and standpipes were cut to fit in either 4 or 10 liter glass jars (25 or 30 cm diam. respectively).

The sprinkler pipe was buried in a 5 cm layer of tap-water-washed aquarium gravel ("Blasting Sand"), and the gravel overlaid with 5 cm of fine-grained dune sand (obtained on Padre Island, Texas). This artificial rooting substrate was very low in endogenous nutrients, trace metals, and organic carbon content (ca. 0.01%). Pure