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Research Note

Testig Growth of *Elodea nuttallii* (Palnch.) H. St. John with Different Culture Media

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Abstract: The Growth rate of *Elodea nuttallii* (Planch.) H.St.John was tested in different culture media. Effects of snail was also determined on the growth rate of the plant. *E. nuttallii* grew well in the canal sediment medium. It also grew well in Steinberg Solution No 1, but not in Steinberg Solution No 2 and JIC:sand media.

Overall the snail treatment did not significantly affect the growth rate of *E. nuttallii* in all tested growth media.

Key Words: *Elodea*, snail, nutrient

Farklı Kültür Sıvılarında *Elodea nuttallii*'nin Büyültülme Testi

Özet: *Elodea nuttallii* (Planch.) H.St.John'nin farklı kültür sıvılarındaki büyüme oranı test edilmiştir. Ayrıca salyangozun bitki büyümesi üzerindeki etkisi de belirlenmiştir. *E. nuttallii* kanal çamuru içeren sıvıda iyi büyümüş, ancak 2 Nolu Steinberg Solusyonu ve JIC:sand sıvısında iyi büyüyememiştir.

Test edilen kültür sıvılarının tümünde de salyangozun büyüme üzerine belirgin bir etkisi gözlenmemiştir.

Anahtar Sözcükler: *Elodea*, salyangoz, besin

Introduction

Submersed macrophytes grow between the shoreline and deep open water and they can intercept or modify material flows from land to the pelagic. Most are rooted and constitute a living link between sediment and overlying water (Özimek et al., 1993). They can play a central role in nutrient cycling, especially in small shallow lakes. They often accumulate large quantities of inorganic elements (Boyd, 1971; Hucthinson, 1975) and can thus have major effects on phosphorus and nitrogen cycling (Carpenter & Lodge, 1986; Reddy et al., 1987).

Although many submersed macrophytes have well developed root systems, they can, in some cases, absorb nutrients directly from the water through their large surface area of foliage (Agami & Waisel, 1986). The degree to which the water nutrient source is used depends on the nature of the nutrient (Barko, 1987) and its concentration in the water (Karignan & Kallf, 1982).

Macrophytes provide a large surface area for colonisation by algae and bacteria and a significant proportion of the vegetative biomass in the littoral zone can be contained in the epiphytic cover (Bronmark, 1989). Thus a mixed algal, bacterial and protozoan community develops on the leaves of *Elodea nuttallii* (Planch.) H.St.John, increasing in density as the leaves age (Paterson & Wright, 1986) and the growth of *E. nuttallii* can be significantly reduced by the algal population (Sand-Jensen, 1977).

Macrophytes offer a rich food source to a number of invertebrate grazers, especially snails which are generally considered to be herbivores. The main feeding method is by scraping the algae/detritus/bacteria complex from the macrophyte surface with radular teeth (Bronmark, 1989). Grazing snails can potentially benefit the macrophytes by reducing epiphytic cover and decaying tissue, thereby decreasing shading effects and nutrient competition (Thomas, 1982).

The aim of this study is to test the effect of both the growth media and snails on the growth rate of *E. nuttallii*.

Materials and Methods

E. nuttallii was collected from stock cultures, sorted from debris and stored at 15°C and in a constant temperature room for four days. Two weighed shoots which were about 8 to 10cm long and without roots were used for each culture jar in this experiment. Plants were set up in 36, 3 l capacity, 25x13x10cm glass jars. Four culture media were used to provide a range of nutrient conditions (Table 1). These were i) canal sediment with dechlorinated tap water, ii) 1:6 John Innes Compost (JIC):sand with dechlorinated tap water, iii) 1:4 Steinberg Solution 1 (-N and -P) : deionised water, iv) 1:4 Steinberg Solution 2 (+N and +P) : deionised water. Details of Steinberg solutions are in appendix 1. With each culture medium was included (+) and (-) snail treatment groups and a control (culture medium without plant). Four replicates and one control was used for each treatments. The experimental conditions were 15°C, 12:12 hours light: dark cycle, in a constant temperature room. The light intensity was 53±2.2 mmol PAR $m^{-1}s^{-2}$, measured with a Macam Quantum Radiometer/ Photometer Q101 (Macam Photometrics Ltd. Livingston, Scotland) underwater probe. The experiment was continued for 28 days. Conductivity and pH were measured twice a week, to check that their ranges did not become extreme for plant growth, using a pHOX 52E conductivity probe (pHOX System Ltd. Ivel Road, Shefford) and a Camlab pH Boy-P2 pH probe (Camlab Ltd. Nuffield Road, Cambridge) respectively. Snails (*Lymnaea peregra* (Mull.)), one for each jar, were put into the jars in the second week, when algal growth started to become visible. Snails were not added at the start in case, in the absence of significant algal growth at that stage, they fed upon macrophyte tissue, thereby damaging the plants. Continuous gentle bubbling with compressed air was used to ensure each jar had efficient O2 / CO2 exchange.

At the end of the experiment the Relative Growth Rates of the plants, based on wet weights, were calculated for each growth medium as below (Hunt, 1990);

RGR= ($\log_e$ final dry wt - $\log_e$ initial dry wt) / duration of experiment.

The chlorophyll content of leaves of *E. nuttallii* was determined by the method of Arnon (1949) after harvesting. This analysis was done on plants from the canal sediment and Steinberg Solution 1 cultures only, because only plants in these two media grew well.

Results

E. nuttallii grew very well in the canal sediment medium (i). It also grew well in Steinberg Solution 1 (iii), but not in Steinberg Solution 2 (iv) and JIC:sand (ii) media (Table 2).

With the canal sediment, *E. nuttallii* plants became very green, produced three or more new lateral shoots on each 'parent' shoot and each new shoot grew as long as the original shoot. They also produced thick and reddish roots. The new leaves were dark green and stem internodes were short. Total chlorophyll content was about 5.645 ± 1.04 µg per whorl.

Plants also grew well in Steinberg Solution 1, but were pale green, produced short new shoots, had long internodes and there were some black spots on leaves. Roots were very long and thin. Leaf chlorophyll content was only about 0.541 ± 0.003 µg per whorl, some ten times less than for plants grown with canal mud.

Table 1. Nutrient concentrations in the growth media i-iv. Description of media i-iv are in text.

nutrient - medium	i	ii	iii	iv
SRP (µg P. l^{-1})	56.00	484.00	<0.1	22790.00
NO_3-N (mg. l^{-1})	0.56	3.70	<0.1	370.00
NH_4-N (µg. l^{-1})	56.00	55.00	<0.1	3.20

SRP : Soluble Reactive Phosphorus NO_3-N : Nitrate NH_4-N : Ammonium

Table 2. The increases in *E.nuttallii* weight calculated for each group over 28 days. Initial dry weight are calculated from initial wet weights using the final wet weight/dry weight ratio. Values shown are means (n=4) and RGR with standard errors in parentheses.

group	initial wet wt (g)	initial dry wt (g)	final wet wt (g)	final dry wt (g)	RGR (g. $g^{-1}.d^{-1}$) wt (g)
i					
(-) snail	0.1585	0.0230	2.6109	0.3871	0.0985 (0.011)
(+) snail	0.1615	0.0225	3.6893	0.5795	0.1092 (0.010)
ii					
(-) snail	0.1706	0.0238	0.5145	0.0713	0.0380 (0.005)
(+) snail	0.1900	0.0247	0.6490	0.0847	0.0570 (0.010)
iii					
(-) snail	0.1865	0.0250	0.9431	0.1287	0.0567 (0.004)
(+) snail	0.1719	0.0266	1.9275	0.2141	0.0702 (0.010)
iv					
(-) snail	0.2461	0.0270	0.3339	0.0370	0.0127 (0.014)
(+) snail	0.1715	0.0246	0.1846	0.0279	0.0095 (0.010)

Overall the snail treatment did not significantly affect the growth rate of *E. nuttallii* in all tested growth media ($p=0.792$, $p=0.937$, $p=0.270$ and $p= 0.676$ respectively from ANOVA).

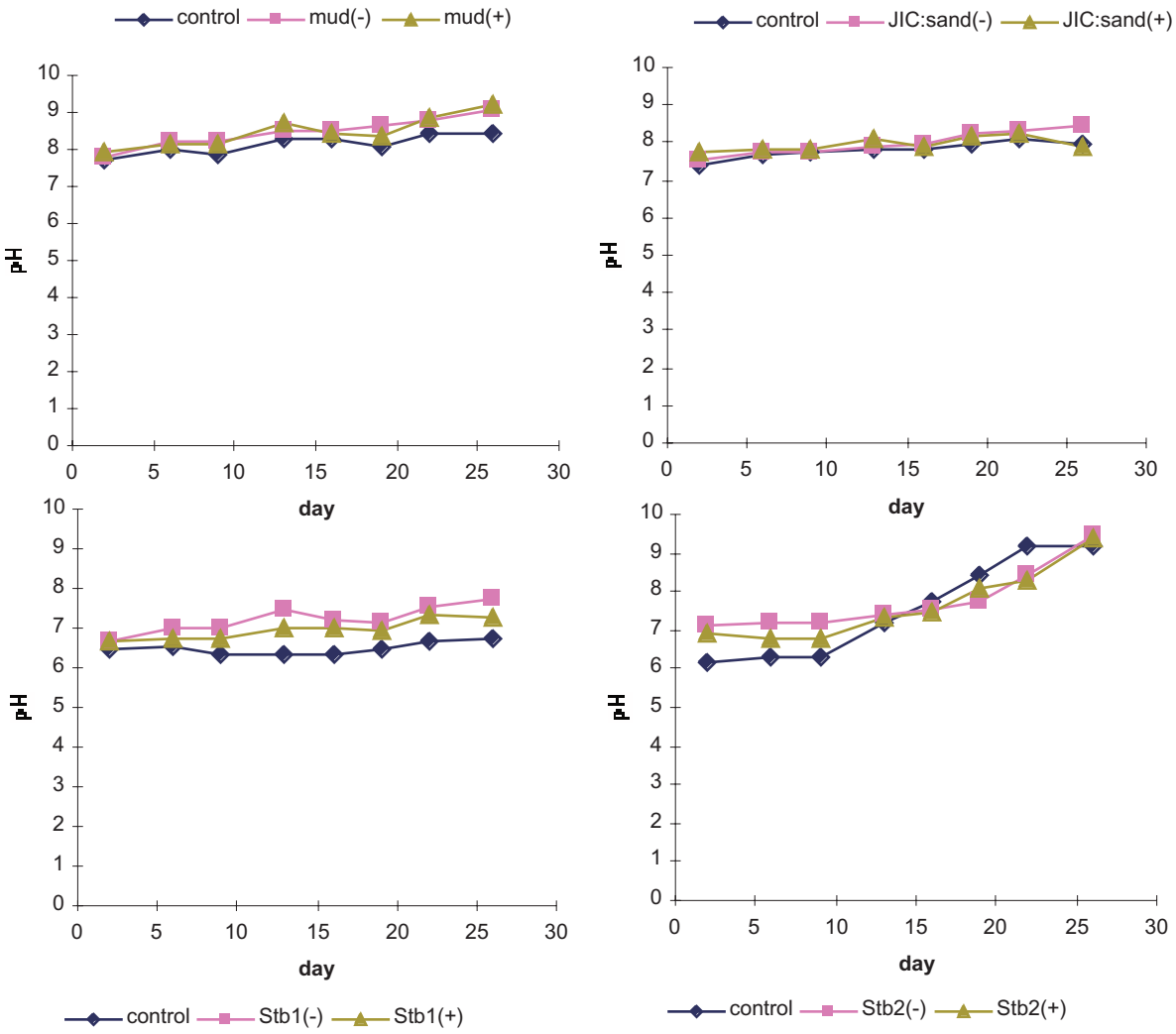
The pH tended to increased during the experiment in all media and treatments (Figure 1). On the other hand conductivity increased in mud and JIC:sand mediums while it decreased in Stb2 and it was almost stable in Stb1 with all treatments (Figure 2).

Discussion

Submersed macrophytes can take up their nutrients from the sediment by the roots as well as from the surrounding water by the shoots. Roots may or may not play an important role in the nutrition or the metabolism of submersed aquatic plants. Waisel and Agami (1983) suggested that when plants of *Najas marina* developed with roots in the sediment, the roots were physiologically highly active, since after root removal, immediate inhibition of shoot growth was observed. In contrast roots developed in the aquatic medium seem to be inactive and removal of these roots did not affect the growth of plants. It is possible that uptake of nutrients by the roots is more efficient and better in soil. Furthermore it is possible that normal activities of roots might be inhibited by light in the aqueous medium. In this condition, removal of roots could lead to a faster shoot growth, but in the soil, removal of roots results in an apparent inhibition of growth because the plant loses essential and functional organs (Agami & Waisel, 1986).

On the other hand *E. nuttallii* did not grow well in a JIC:sand mixture. JIC is a very rich rooting medium. It would have released more nutrients into the water than the other media and it seems to have negatively affected the growth of *E. nuttallii* when compared with canal sediment. Steinberg 2 solution, which has a high N and P content, also proved to be a poor growth medium. It seems that *E. nuttallii* cannot grow in media high in N and P. Anova and Tukey's tests showed that plants grew best in canal sediment, which releases markedly smaller amounts of nutrients into the water ($p<0.001$ +snail and $p=0.001$ -snail from ANOVA).

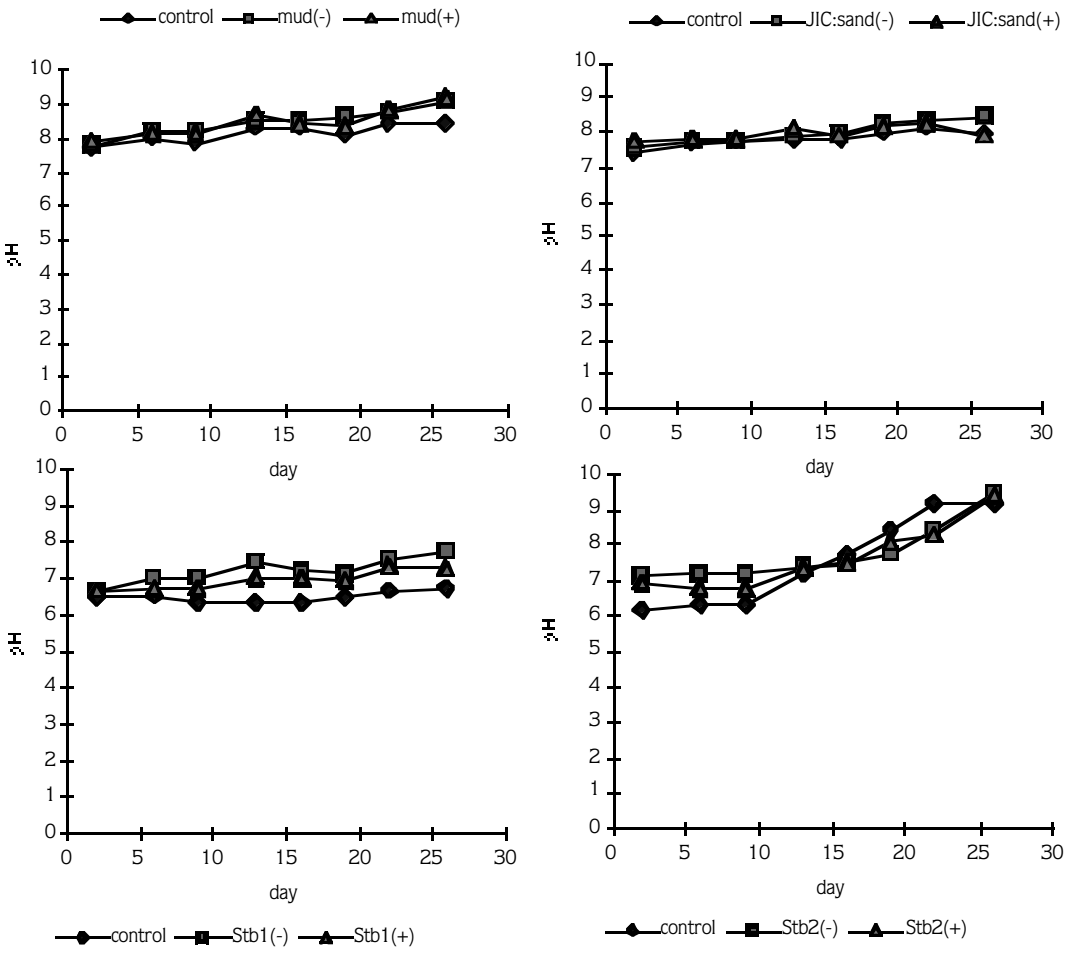
Algal growth was observed on the sediment, surface and walls of the jars in JIC:sand and canal sediment treatments, while it occurred on the bottom, surface and walls of the jars in Stb2 treatment. However there was no visible accumulation of algae in the Stb1 + snail treatment. Stb1 has a low nutrient concentration. The algae are likely to have low growth rates in waters which



mud(+) : mud growth condition with snail
mud(-) : mud growth condition without snail
JIC:sand(+): JIC:sand growth contion with snail
JIC:sand(-) : JIC:sand growth condition without snail
stb1(+) : Steinberg growth condition -N, -P with snail
stb1(-) : Steinberg growth condition -N, -P without snail
stb2(+) : Steingerg growth condition +N, +P with snail
stb2(-) : Steinberg growth condition +N, +P without snail

Figure 1. pH measured twice a week in each growth medium with both (-) and (+) snail treatments over 28 days.

have low nutrient concentrations (Eminson & Moss, 1980). But algal growth was seen on the plants with Stb1 - snail treatment. Submersed macrophytes secrete organic compounds during photosynthesis or leak such compounds during senescence (Bronmark, 1989) and epiphytes can use these compounds as a nutrient source (Carignan & Kallf, 1982; Allen, 1971; McRoy & Goering, 1974), but epiphytic algae would be removed from *E. nuttallii* tissues by the grazing snail in the Stb1 + snail treatment and therefore *E. nuttallii* does, as expected, grow quite well in Stb1 + snail treatment.



mud(+) : mud growth condition with snail
mud(-) : mud growth condition without snail
JIC:sand(+): JIC:sand growth contion with snail
JIC:sand(-) : JIC:sand growth condition without snail
stb1(+) : Steinberg growth condition -N, -P with snail
stb1(-) : Steinberg growth condition -N, -P without snail
stb2(+) : Steingerg growth condition +N, +P with snail
stb2(-) : Steinberg growth condition +N, +P without snail

Figure 2. Conductivity measured twice a week to each growth medium with both (-) and (+) snail treatments over 28 days.

On the other hand algal growth on the walls of the containers will have reduced lateral light penetration into the water in Stb2, JIC:sand and mud treatments, both with and without snails. However mud had sufficient nutrients for root uptake and growth of *E. nuttallii* while nutrient concentrations in the medium were low. For this reason *E. nuttallii* grew rapidly and reached the water surface before algal growth could interfere. Thus algal growth on the walls of container could not affect the growth of *E. nuttallii* in the mud, plenty of light being still available on the surface, with or without snails present . In contrast, nutrients were released from JIC:sand to the water at quite a high level and Stb2 has high N and P concentrations initially in solution, so algal growth would be expected to become visible earlier in JIC:sand and Stb2 treatments than with mud, as was indeed the case.

In Stb1 medium, pH increased in all treatments, but the increase was very similar with the JIC:sand and both

were smaller than in Stb2 and mud. Stb1 medium had low nutrient concentrations and algal growth was restricted accordingly. Plants grew in this medium, but their leaf chlorophyll content was low. Therefore both algal growth and photosynthesis (algae and plant) would be expected to be reduced in Stb1 with all treatments.

In the Stb2 treatment increase of pH was very obvious and was greater than in the other media. Stb2 had a high nutrient level and algae grew well in this medium, while the plant grew only poorly. The large pH increase was presumably due to algal photosynthesis.

Conductivity increased until day 5 in mud and JIC:sand media and was almost constant with all treatments of JIC:sand and + and - snail treatments of mud, but increase of conductivity continued in the controls. Such increase could be due to ion release processes from sediment and possible plant tissues to the medium, but conductivity tended to be stable after that, perhaps because release processes were complete or uptake by the increasing plant biomass was absorbing any continuing release from the sediment.

On the other hand conductivity decreased in Stb2 and was constant in Stb1 with all treatments. Stb2 media had initially high nutrient levels which favoured algal growth and the observed decrease of conductivity may have been cause by uptake of ions during algal growth in this medium. However Stb1 did not include N and P, algal growth was consequently restricted and therefore conductivity was constant and lower than all other media.

Appendix 1.

The full Steinberg Solution contains nutrient	mg. l^{-1}
KH_2PO_4	100
KNO_3	350
$MgSO_4 7H_2O$	100
$Ca(NO_3)2H_2O$	295
$ZnSO_4H_2O$	0.18
$MnCl_2 4H_2O$	0.18
H_3BO_3	0.12
$(NH_4)6MoO_{24}H_2O$	0.037

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