

IN VITRO SHOOT AND LEAF PROLIFERATION OF *ENCYCLIA TAMPENSIS* (LINDL.) ORCHIDS

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ABSTRACT

Encyclia is a genus of orchids with four members native to Florida that are Endangered or commercially exploited. If a commercial orchid tissue culture media was found that could be used to propagate *Encyclia*, the resulting plants could be introduced into protected habitats to increase the wild populations or could be grown commercially to lessen the demand for collected plants. Six month *Encyclia tampensis* (Lindl.) seedlings were grown in Orchid Maintenance/Replate Medium w/ Banana and Charcoal with 6g/L agar (C), ½ strength Orchid Multiplication Medium with 6g/L agar and 10g/L charcoal (OM), Knudson C Orchid Medium – Morel Modification with 6g/L agar and 10g/L charcoal (K), or Vacin & Went Modified Orchid Medium w/ Agar and Sucrose with 10g/L charcoal (VW) for seven weeks. Plants grown in OM and VW media both produced more shoots and roots (7.56 leaves, 1.92 shoots and 6.55 leaves, 1.73 shoots respectively) than the control medium (5.68 leaves, 1.64 shoots) with OM producing the highest averages. Plants grown in the K medium averaged significantly less shoots and leaves than the other three media (4.48 leaves, 1.32 shoots).

INTRODUCTION

In Florida there are four native species of *Encyclia* orchids (*E. tampensis*, *E. boothiana*, *E. cochleata*, and *E. pygmaea*). All of these species are either endangered or commercially exploited from being collected for personal use or sale and the destruction of their habitats by developers.

In vitro micropropagation is already used to provide an efficient way to multiply some orchids. If this technology applied to *Encyclias*, it can alleviate the pressure on wild populations by providing a more marketable supply of plants and increase the wild populations by reintroducing *in vitro* grown plants to protected areas. Thus far, the only published micropropagation efforts with *Encyclia* orchids are on seed germination and not plant multiplication.

MATERIALS AND METHODS

Plant Materials

The plants for the experiment were six month old *Encyclia tampensis* (Lindl.) plants that were germinated on a Vacin & Went Modified Basal Salt medium and transferred to an Orchid Maintenance/Replate medium with Banana and Charcoal at four months. Healthy seedlings with one shoot and one to two roots were chosen for the experiment.

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Conditions. For the experiment’s seven week duration, the plants were kept in the plant biotechnology lab’s clean room in 100mL baby food jars. They received a 16 hours light/8 hours dark light cycle with full spectrum grow lights.

Data Collection. The numbers of leaves and shoots visible on the plants were counted and recorded to measure growth and multiplication rates. Data was collected weekly. Since the *E. tampensis* seedlings did not have readily visible stalks, shoots were defined as separate rosettes of leaves for this experiment. Most of the shoots had a pseudobulb at the base of the rosette.

Media Variations. There were one hundred plants divided into four groups of twenty-five. Each plant received 10mL of an orchid media for the duration of the experiment. A maintenance medium was chosen for the control and three commercial orchid multiplication and/or mericlone media were chosen for the experiment. Group 1 (Control) received Orchid Maintenance/Replate Medium w/ Banana and Charcoal with 6g/L agar added. Group 2 (OM) received ½ strength Orchid Multiplication Medium with 6g/L agar and 10g/L charcoal added. Group 3 (K) received Knudson C Orchid Medium – Morel Modification with 6g/L agar and 10g/L charcoal added. Group 4 (VW) received Vacin & Went Modified Orchid Medium w/ Agar and Sucrose with 10g/L charcoal added.

RESULTS AND DISCUSSION

Shoots. The VW plants showed signs of additional shoots within one week, the other three groups showed new shoots within two weeks with the Control growing new shoots the fastest. By week five the OM plants had caught up and by week seven the VW plants had caught up with the control. The Control group had an average of 1.64 shoots per plant and had six plants with three or more shoots. The OM group had an average of 1.92 shoots per plant and had five plants with three shoots. The K group had an average of 1.32 shoots per plant and had one plant with three shoots. The VW group had an average of 1.73 shoots per plant and had four plants with three shoots. (See Figure 1 and Table 1)

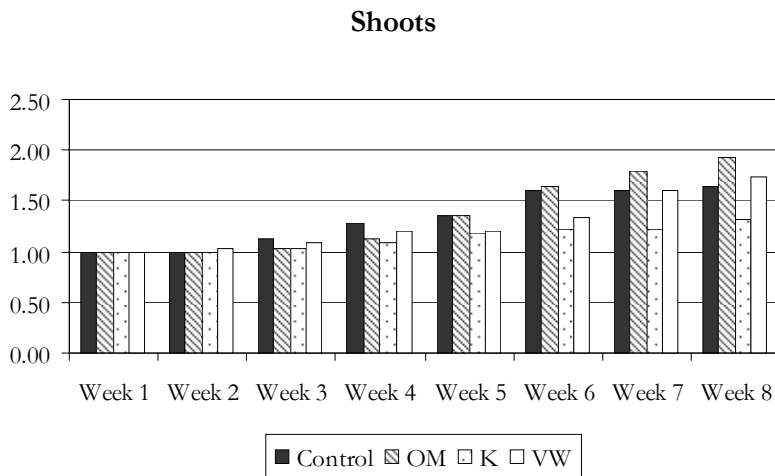


Figure 1. Overall shoot growth

Table 1. Average Numbers of Shoots

Treatment	Number of Shoots							
	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8
Control	1.00	1.00	1.12	1.28	1.36	1.60	1.60	1.64
OM	1.00	1.00	1.04	1.12	1.36	1.64	1.80	1.92
K	1.00	1.00	1.04	1.09	1.17	1.22	1.22	1.32
VW	1.00	1.04	1.08	1.21	1.21	1.33	1.61	1.73

Leaves. VW also had the largest initial spurt of leaf growth although it started with the lowest average number of leaves. VW and OM both passed the control’s rate of leaf production within one week and grew consistently throughout the time frame. The Control group had an average of 5.68 leaves per plant and had four plants with nine or more leaves. The OM group had an average of 7.56 leaves per plant and had eleven plants with nine or more leaves. The K group had an average of 4.48 leaves per plant and had one plant with nine or more leaves. The VW group had an average of 6.55 leaves per plant and had four plants with nine or more leaves. (See Figures 2 and Table 2)

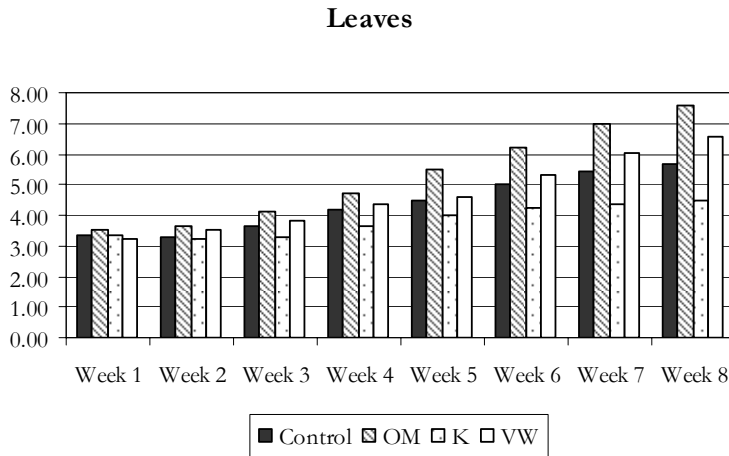


Figure 2. Overall Leaf Growth.

From the results, it seems that OM is the best medium to use for the micropropagation of *Encyclia* orchids. OM provides fairly constant results throughout a diverse population of plants as compared to the other media. VW could possibly surpass OM’s abilities if given enough time but doesn’t have as consistent results. K appears to be inadequate for micropropagating *Encyclias* since its results were consistently below the Control’s.

Table 2. Average Numbers of Leaves

Treatment	Number of Leaves							
	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8
Control	3.32	3.28	3.64	4.16	4.48	5.00	5.44	5.68
OM	3.52	3.64	4.12	4.72	5.48	6.20	6.96	7.56
K	3.32	3.20	3.28	3.65	4.00	4.26	4.35	4.48
VW	3.24	3.52	3.79	4.38	4.58	5.29	6.04	6.55

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8 Month Old Plant



14 Month Old Plant