Rooting and transplant growth of tissue-cultured Magnolia grandiflora L. cv. Bracken's Brown Beauty

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Introduction

Magnolia grandiflora cv. Bracken's Brown Beauty explants were induced to proliferate *in vitro* from embryonic shoot tips dissected from mature terminal and axillary buds. The explant growth was optimal on WPM (Woody Plant Media) with BA (6-benzylamino-purine, a synthetic cytokinin that stimulates cell division) concentrations of 1.0 mg/l to 5.0 mg/l. A more detailed explanation of methods and procedure was given in an earlier communication, see Tobe, 1990.

Root Induction—*In Vitro*

For root induction microcuttings consisting of axillary buds of explants were removed from the existing shoot tips in tissue culture and placed on WPM with IBA (indolebutyric acid, a synthetic auxin that stimulates cell swelling, division and adventitious root formation) at a concentration of 5.0 µM for seven days, then transferred to hormone free WPM. Generally from two to three axillary buds were used for each shoot tip. Half of the microcuttings were not exposed to the IBA and were simply placed on hormone free WPM. After approximately eight weeks, as of the end of November 1990, the explants exposed to IBA had begun to swell at their base. At ten weeks the explants exposed to the IBA had begun to grow long, thick white roots within the agarous gel, generally one root/plant. After approximately one week some of these had elongated over 4.0 cm in length and initiated secondary rootlets (Photo 1). After twelve weeks and numerous subculturings all of the explants (even those without exposure to IBA) developed roots, but in general the exposure to an auxin was necessary for earlier root induction.

Establishment in the soil—*In Vivo*

Four weeks after initial root growth the plantlets were robust enough to be transferred to the greenhouse. A sterilized soil mixture of 1/3 peat, 1/3 perlite and 1/3 vermiculite was used. The top of the magenta box (most any glass or clear plastic container that fits around the plantlet could be used) was carefully placed over the plantlets such that a seal between soil and box was created (Photo 2). After two weeks in the greenhouse the plantlets began to show new leaf expansion but growth was slow and became chlorotic prompting the application of a half strength water soluble fertilizer (20-20-20) to the foliage and soil. The explanation for this slowing of growth and chlorosis might be related to (1) the transition of plantlets from continuous light to the light/dark cycle of early spring,

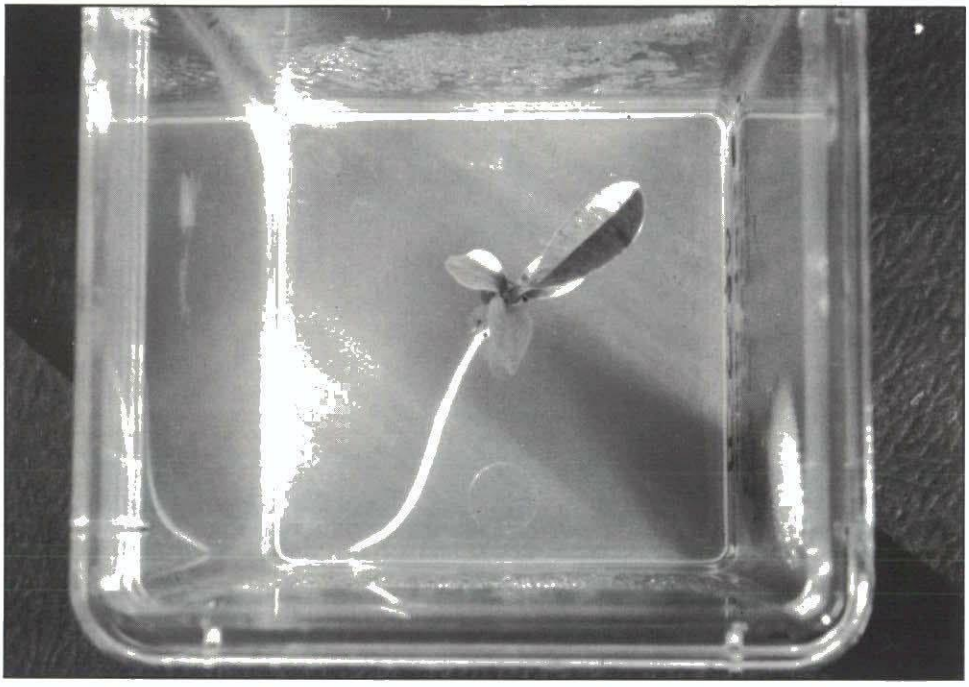

Photo 1. Magnolia grandiflora *'Bracken's Brown Beauty' explant after sixteen months growth on WPM media. Explant root growth is indicated at the arrow. X2*

Photo 2. Explant after seventeen months growth on WPM media. The explant is covered with a magenta box turned upside down. X1/2

February, 1991, (2) a reduced root surface area over which to absorb nutrients and (3) the lowering of humidity outside the in vitro environment of the magenta box. In mid-April, after the danger of frost, the plantlets were transferred from the greenhouse to a shadehouse under "field conditions" for acclimatization to the outside temperature and humidity. By July 1, 1991, the plantlets had produced three new leaves on average.

Results and Discussion

The results described above demonstrate that the micropropagation of *Magnolia grandiflora* is feasible. Shoot multiplication or the growth of axillary buds is stimulated by the rejuvenating effects of repeated subculturing in a cytokinin (Pierik, 1987) over time. Continued removal of explants from the axillary buds could be used to perpetuate new explant material over a relatively long time period *in vitro*, but this may not be advantageous for clonal maintenance as somaclonal, or genetic variation from mutations, are more likely to occur over time (Scowcroft, 1985). This change in genotype might be advantageous if the novel genetic combinations of a mutant were wanted for phenotypic change, *e. g.* changes in leaf and flower morphology. Although successful, methods used in this experiment will not be a financially viable alternative to the current practice of rooting cuttings until a less labor intensive protocol is discovered. ❧

References

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