PROPAGATION OF *PLATANUS* × *ACERIFOLIA* FROM MERISTEM TIPS[1]

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Abstract. Explants of *Platanus* × *acerifolia* aseptically excised and placed on the MS medium supplemented with 0.1 mg/l vitamin C, 0.1 mg/1 BAP and 0.01 NAA will develop into complete plantlets. A month of darkness following seven weeks exposure to continuous light (1300-1500 lux) is necessary to induce rooting.

Meristem tip culture was used in the early 50's by Morel and Martin (1952) to produce virus-free clones from infected plants. They obtained virus-free explants from infected dahlia cultivars but were unable to induce rooting. They did obtain healthy plants by grafting these explants on virus-free seedlings. Meristem culture was then extended to other crops and was responsible for the development of cultured carnation, chrysanthemum and geranium programs. It has also been extended to woody species (Bonga, 1977). Although success in meristem culture with the tree crops has been limited, virus-free clones of hybrid poplars (Berbee *et al.* 1972) and cassava (Berbee *et al.* 1973) have been obtained from meristem tips. We have studied the propagation of sycamore, *Platanus* × *acerifolia* Willd. by means of meristem tip culture.

The terminal 5 to 6 cm of one-year-old shoots of *P. acerifolia* were removed from trees growing on the University of Warsaw campus on March 28, 1978, and again on May 9. Excision and transfers of apical meristems were performed in a small (1.5 × 2.0 × 2.5 m) closed transfer room equipped with ultraviolet lights. Prior to use, the walls, floors and ceiling of the transfer room were washed with 95% ethanol and then exposed to at least 3 hours of irradiation. Preliminary studies with *Platanus* and other woody species indicated that the Murashige-Skoog (MS) medium (1962) supplemented with benzyladenine (BAP), naphthaleneacetic acid (NAA) and 0.1 mg/l vitamin C supported growth of the explants and demonstrated a superiority of solid medium over a liquid medium with filter paper bridges. Conse-

Fig. 1. Explant (X4) of *Platanus acerifolia* after six weeks exposure to continuous light (1330 to 1500 lux). Note good callus growth and shoot development.

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quently, the supplemented MS medium solidified with 1% agar was used in our studies.

In preparation of the March collection, the outer bud scales were first removed from the terminal bud. The meristem covered with 2 or 3 remaining scales was then dipped in 79% ethanol, immersed in 3% monochloraminum solution for 30 min and rinsed in five changes of sterile distilled water. The remaining bud scales were removed and the meristem containing 2 or 3 leaf primordia excised and placed on agar in test tubes (20 × 150 mm) which were then plugged with cotton. In the May collection, the emerging shoots were sterilized immediately, then, the meristems excised and treated as earlier.

Fig. 2. Explant (X4) of *Platanus acerifolia* after seven weeks exposure to continuous light (1300 to 1500 lux) followed by one month of darkness. Note development of roots and shoot.

The March bud collections consisted of 200 meristems in test tubes of which 100 were exposed to continuous light of 1300 to 1500 lux at 25-30 to 50C under 80% humidity. The other 100 were kept in the dark under the same conditions of temperature and humidity. On May 20, the surviving explants in each of the two treatments were divided into four groups and then transferred to four different MS media supplemented with vitamin C plus 0.01 or 0.1 mg/l BAP and 0.01 or 0.1 mg/l NAA. No shoots developed from any of the explants but the rooting and survival percentage were superior on the explants exposed to the dark conditions.

The May collection consisted of 160 terminal buds which were breaking dormancy. These were divided so that 40 explants were placed on each of the different BAP:NAA combinations and exposed to continuous light for seven weeks when half were transferred to continuous dark conditions and the other half kept under continuous light (Table 1). After one month only a few of the explants exposed to continuous light had roots and only one developed into a complete plant. Of the plants exposed to the dark conditions 30 to 60% had roots and 45% of these developed into complete plantlets. The greatest percentage of rooting occurred on those explants growing on the agars containing 0.1 mg/l BAP but that with the 0.01 mg/l NAA had the greater percentage of fully differentiated plantlets. These plantlets had pale, etiolated leaves which rapidly turned green upon exposure to light.

These observations indicate that plantlets of *P. acerifolia* will differentiate from meristem tips. Presently, the usefulness of meristem tip culture of *P. acerifolia* appears to be restricted to obtaining healthy plants from infected trees but if the plantets can be induced to multiply *in vitro* as in strawberry (Boxus *et al.* 1977) it will provide a rapid method for vegetative propagation of healthy clones of sycamore.

The MS medium supplemented with BAP, NAA and 0.1 mg/l vitamin C supports good growth of the explants under the proper environmental conditions and growth regulator balance. Precise temperature control isn't critical but seven weeks

Table 1. Rooting and production of plantlets from explants of *Platanus* × *acerifolia*

Concentration of growth regulators (mg/l)		No. of explants		
BAP	NAA	Surviving initial[1] growth period	Surviving six weeks later	
			rooted	plantlets
Continuous light (1300 to 1500 lux)				
0.1	0.1	12	1	0
0.01	0.1	12	1	1
0.1	0.01	9	2	0
0.01	0.01	7	0	0
Dark				
0.1	0.1	10	6	1
0.01	0.1	11	4	2
0.1	0.01	8	5	3
0.01	0.01	7	3	2

[1]Initially, 40 explants were started on each of the four different BAP-NAA combinations and held for 7 weeks under continuous light after which ca. 50% of the survivors were exposed to continuous light and the other half to dark conditions.

of initial exposure to continuous light followed by a month of darkness provides favorable conditions for organogenesis. Concentrations of 0.1 mg/l BAP and 0.01 mg/l NAA appear to be an effective regulator balance.

Our study also demonstrates that meristem tip culture does not require elaborate equipment, confirming the observations of Stoltz (1980). Only those conditions which minimize contamination and provide reasonable temperature control appear to be essential.

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