## TISSUE CULTURE OF AVOCADO

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Progress Report

The main objectives of tissue culturing of avocados are to eliminate the viroid-like organism causing sunblotch disease from rootstocks and clonal material and to propagate scarce vegetative material rapidly.

Tissues from nearly all parts of the avocado plant can be cultured as callus masses *in vitro* (Schroeder, 1968, 1971, 1973, 1975). Differentiation of the callus masses into definite, recognizable structures has been observed in many experiments. However, the reproduction of an entire and normal plant from callus tissue derived from a relatively pure specific tissue of avocado has not yet been achieved. The investigations reported here by the authors are presented as a progress report on *in vitro* studies with avocado tissues.

Single node cuttings were cultured in sample tubes containing medium for proliferation after surface sterilization with 1% NaOCI for 15 min and rinsing in sterile water. Tubes were incubated in a growth cabinet at 23°C and a photoperiod of 18h. In some cases shoots, 5—6 cm long, developed after 6 weeks. These were cut into single-bud explants which were transferred to fresh medium. By repeating the process, a considerable number of explants can be obtained. Thus far, this technique has not been entirely successful for the tissue culturing of avocado cultivars. However, the wild species *Persea indica* reacted better and it was possible to obtain rooted plantlets after subculturing *ca* 2 cm cuttings on the basic proliferation medium minus benzyl-aminopurine but plus 1 mg/ $\ell$  indolylbutyric acid (IBA) (Fig. 1).

A more suitable technique for clonal propagation of avocado proved to be the *in vitro* treatment of shoot tips with a cytokinine (6-benzylaminopurine) to decrease apical dominance. Through this technique, bunches of shoots were obtained (Fig. 2, 3 and 4). From one axillary bud, 60 explants can be obtained within eight weeks by transferring individual shoots to fresh proliferation medium with cytokinine. Rooting of these shoots on media containing 0—5 mg/ $\ell$  IBA has thus far been unsuccessful.



FIG. 1: Persea indica – development of a single bud cutting (left) and rooted plantlet (right)

Composition of proliferation medium:

Macro-elements-half strength Murashige and Skoog (1962) Micro-elements-full strength Murashige and Skoog (1962) NaFe EDTA-25 mg/ $\ell$ ; NaH<sub>2</sub>PO<sub>4</sub>-2H<sub>2</sub>O-170 mg/ $\ell$ ; adenine sulphate — 80 mg/ $\ell$ ; ascorbic acid — 25 mg/ $\ell$ ; glycine-2 mg/ $\ell$ ; nicotinic acid — 1 mg/ $\ell$ ; pyridoxine HCl — 1 mg/ $\ell$ ; thiamine HCl — 1 mg/ $\ell$ ; Capantothenate — 1 mg/ $\ell$ ; gibberellic acid — 0,5 mg/ $\ell$ ; 6-benzylaminopurine — 2 mg/ $\ell$ ; inositol — 100 mg/ $\ell$ ; sucrose — 30 g/ $\ell$ .

Medium for embryo culturing of avocado:

 $\begin{array}{ll} Macro-elements & - \ half \ strength \ Murashige \ and \ Skoog \ (1962)\\ Micro-elements & - \ full \ strength \ Murashige \ and \ Skoog \ (1962)\\ NaFe \ EDTA & - \ 25 \ mg/\ell; \ sucrose & - \ 40 \ g/\ell; \ inositol \ - \ 100 \ mg/\ell; \ gibberellic \ acid \ - \ 0,5 \ mg/\ell, \ thiamine \ - \ 1 \ mg/\ell. \end{array}$ 



FIG. 2: Tip cutting 14 days after inoculation



FIGS. 3-4: Development of axillary buds





FIGS. 5-7: Production of an avocado plantlet from an em bryo shoot explant







FIG. 8: Germinating embryo



FIG. 9: Shoot apex graft

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