

ETIOLATION AND AVOCADO BUD ELONGATION IN VITRO

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Several lines of investigation on the behavior of specific tissue pieces of avocado plant axis, flower, and fruit indicate that the species can be maintained for long periods of time as a continuously growing callus mass from which well developed roots can be induced occasionally (4). Likewise the generation of callus tissue from pericarp cells, while not a unique characteristic, nevertheless can be accomplished without difficulty. Investigations on the response of stem pieces have provoked several questions related to the problem of tissue proliferation and cellular differentiation in the callus tissues which are induced in such studies. The effect of exogenous materials in the nutrient media on the growth pattern and subsequent behavior of avocado explants suggests that many rather simple exogenous substances such as indolebutyric-acid, indole-3-acetic-acid and other hormonal molecules can increase cell number and affect subsequent fresh weight of callus masses derived from woody stem tissue.

The general response observed when stem pieces together with an included bud are placed on nutrient agar media, is the massive proliferation of the stem piece at the cut surface but the limited growth or inhibition of length growth of the included bud system. The utilization of stem sections which include either apical or tip buds with lateral buds taken directly from the field or from plants grown under rather regular light conditions in a glass house shows this general response.

The general conditions of the tissue culture procedures involved the use of 6 dram screw top vials each with 10 ml of appropriate nutrient agar medium based on the formulation of Nitsch and modified by Schroeder (4) or that of modified Murashige and Skoog as available from Flow Laboratories. All preparation and transplant operations were conducted under sterile conditions. The vials each with a single explant were maintained in a culture room at 25°C under a continuous light period at an intensity of 100-400 fc using Grolux lamps.

Avocado stem pieces about 2 cm long, with or without "lateral bud", were selected from seedlings growing in the glass house or from comparable seedlings maintained in a dark room. The latter were transferred to the dark room upon the initiation of germination. Some seedlings about 120 cm tall which had been growing in the glass house were cut back to 30 cm, then transferred to the dark. The tissues which subsequently developed in the dark either directly from the seed or from the lateral buds of the decapitated seedlings resulted in thin, long, white stems on which lateral buds appeared subtended by very small vestigial leaves. The development of the white-stemmed etiolated tissue provided lateral stems sometimes 100 cm or more in length. Comparisons were made on identical media of stem pieces from light-grown and etiolated stem sections of approximately the same size. Some stem pieces contained

lateral buds while other pieces were without buds.

The major response of stem pieces of light-grown and etiolated sources was the development of massive callus masses at the cut ends. This massive proliferation occurred primarily at the exposed or upper end of the vertically oriented stem section when maintained on Nitsch medium, A rather distinctive more friable proliferation at the basal end within the agar medium was evident in the Flow Lab medium.

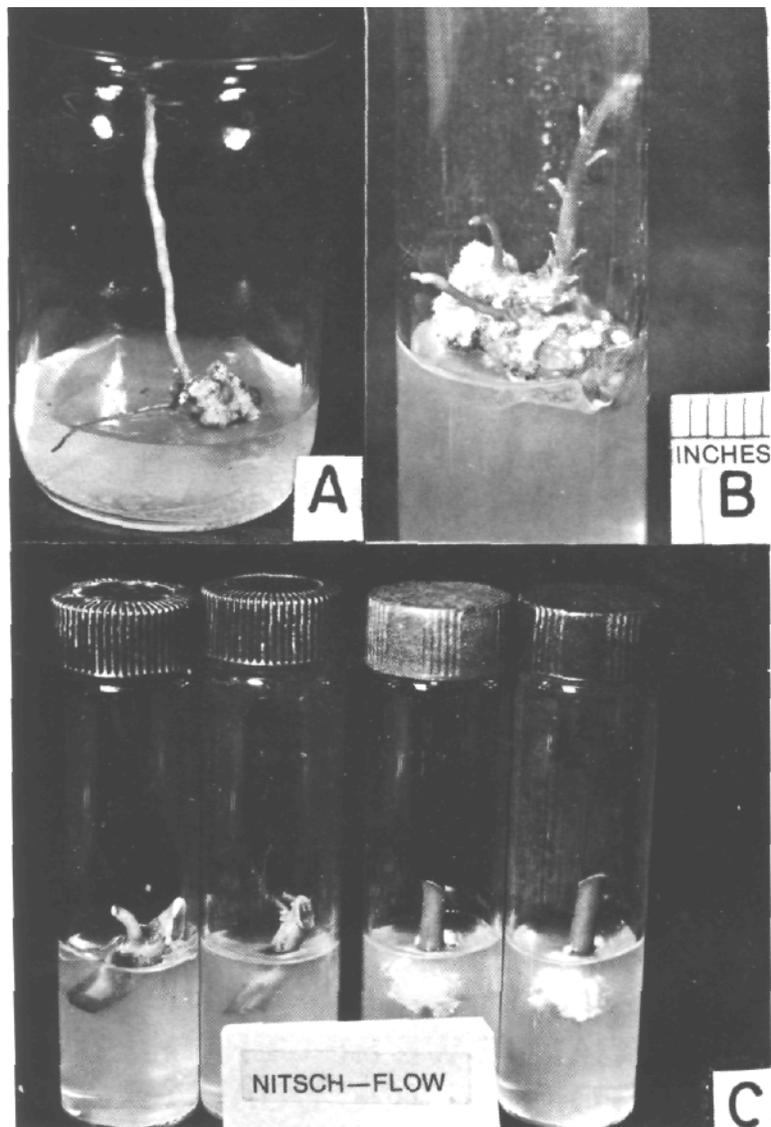


Figure 1. Avocado seedling stem pieces or buds from etiolated tissues grown in vitro. A. Lateral bud which has elongated and produced root from basal callus. B. Lateral bud showing some stem elongation, massive callus formation and roots from the callus. C. Stem segments grown on two media. Note apical callus on Nitsch medium and massive basal callus on Flow.

Massive callus formation generally results from stem sections of avocado seedlings

grown in the light or in the dark. If lateral buds are present, most of the sections taken from light-grown plants will show little or only occasional elongation of the bud. Lateral buds from etiolated stems, however, show marked increase in number of buds which elongated, and under comparable time periods, indicated considerable length growth compared to light buds.

These general observations, under the conditions described, suggest an inhibition of lateral bud length growth in tissues devised from light-grown plants. The factors associated with the development of tissues in total darkness appear to release the lateral bud and induce stem elongation when placed under conditions of in-vitro culture.

The effects of dark treatments on the development of tissues in absence of light causes many responses of such tissue when returned to normal light conditions. Etiolation treatment to induce rooting in otherwise difficult to root woody tissues has long been known and well demonstrated in avocado (1), apple (2), and other species (3), Pretreatment of tissues by exposure in darkness many prove valuable in the development and improvement of techniques in tissue culture procedures related to propagation of clonal materials.

Literature Cited

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