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GROWTH OF AVOCADO FRUIT TISSUE ON ARTIFICIAL MEDIA

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Investigations on the development and physiology of the avocado fruit have indicated that it is unique compared with such temperate zone fruits as apples, peaches, or pears. The latter fruits have been studied extensively and a knowledge has been acquired concerning their chemical composition, anatomical development, and physiological behavior on the tree and under post-harvest conditions. Studies on the physiology of the avocado fruit by Biale (1) have shown that the avocado is characterized by an extremely high rate of evolution of carbon dioxide when measured against a given weight of fruit for a given time. The respiration rate of 150 to 180 mg. of CO_2 per kilogram of fruit per hour is among the highest recorded for important fruits. Another aspect of fruit development markedly exhibited by the avocado is the sudden increase in respiration activity for a short period and then a rapid decline, a phenomenon known as the climacteric. This climacteric point in the avocado has been demonstrated by Biale to be coincidental with the softening of the fruit.

Morphological and anatomical studies of the avocado fruit (3) have suggested that this fruit differs substantially in its type of development compared with deciduous fruits. The latter fruits, as a group, are characterized essentially by the following mode of development. As a consequence of pollination and fecundation of the ovules, the ovary is stimulated to undergo a period of cell multiplication, during which time cell divisions occur and cell numbers increase in rapid succession. This period of cell division continues for about three weeks, then ceases. Then follows a period during which expansion and enlargement of the previously formed cells occurs. Cell enlargement continues until the fruit is mature or harvested.

The avocado appears to be unique in respect to its developmental histology in the following manner. Pollination and subsequent fecundation of the ovule stimulates the avocado ovary to undergo cell division. Such divisions do not cease after the first month, but instead continue as long as the fruit remains on the tree. Evidence that cell divisions occur in nearly mature or mature fruits may readily be observed in properly prepared materials *(*Plate 1).While the avocado fruit develops from an ovary about 1/16 inch in diameter to a mature size of four to five inches in diameter, the individual cells reach a maximum size of approximately 0.0024 inch in diameter in the Fuerte variety, at which time they undergo division to form two new cells. Because the maximum size of cells appears to be approximately constant, the difference in size between fruits of a given variety is determined by differences in number of cells. Hence, the larger the fruit the greater the number of cells in that fruit. A calculation, based on a single sample, indicated that a Fuerte fruit of 7 oz. contains approximately 1,350,000,000 cells.

Because the avocado fruit is characterized by this ability to undergo cell division at all stages, even as it approaches maturity, an attempt was made to maintain this state of cell multiplication in pieces of tissue separated from the fruit. Slices of tissue 8 mm. in diameter and 2 mm. thick were taken under sterile conditions from nearly mature fruit and placed in glass vials on an agar media *(Figure 1).* The culture media contained the basic ingredient; required for the nutrition of plants in general, such as nitrate, sulphate, magnesium, calcium, etc., and in addition there were added such materials as vitamin B6, growth promoting substances such as indoleacetic acid, amino acids of various types, and coconut milk.

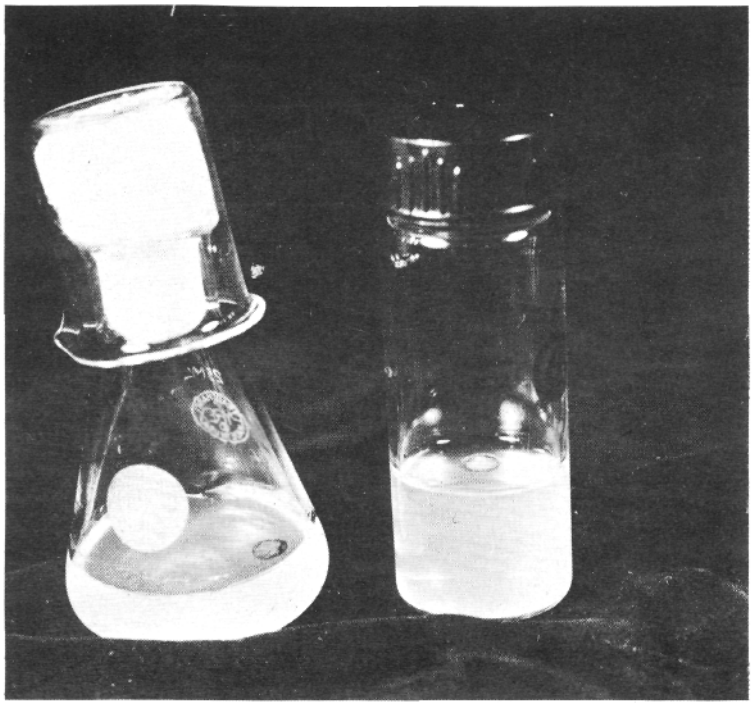

Figure 1. Avocado fruit tissue growing in vitro on agar nutrient media.

Such tissue slices, placed in vials and kept at room temperature, may begin to show indications of growth in three weeks. The first evidence that the sections are alive is the appearance of cellular proliferation on the upper surface of the tissue disk. This surface tuft results from divisions of cells at the surface and the uplifting of these algal-like strings of cells (Figures 2 & 3). In some tissue disks no external evidence of growth can be detected, but instead internal cell division may be observed upon sectioning and examination under the microscope. This activity does not result in immediate expansion or growth of the tissue. Cambial-like layers are occasionally found, the activity of which results in a large number of small brick-shaped cells which are lifted en masse, such that they cause the disk to increase in thickness and diameter.

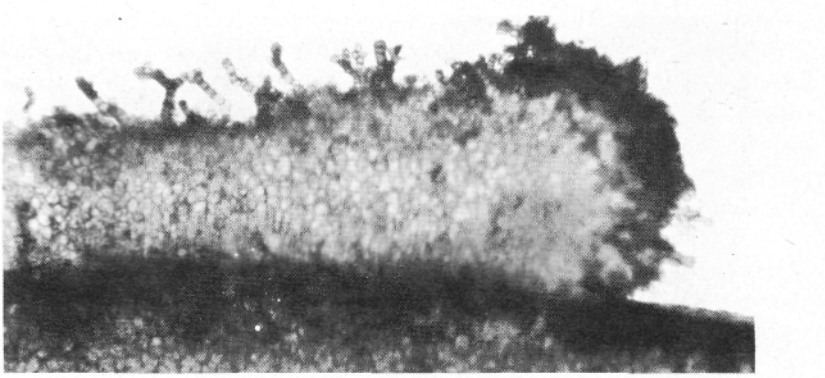

Figure 2. Section through surface layers of avocado tissue grown in vitro showing proliferation of new tissue and of algal-like cells on upper surface.

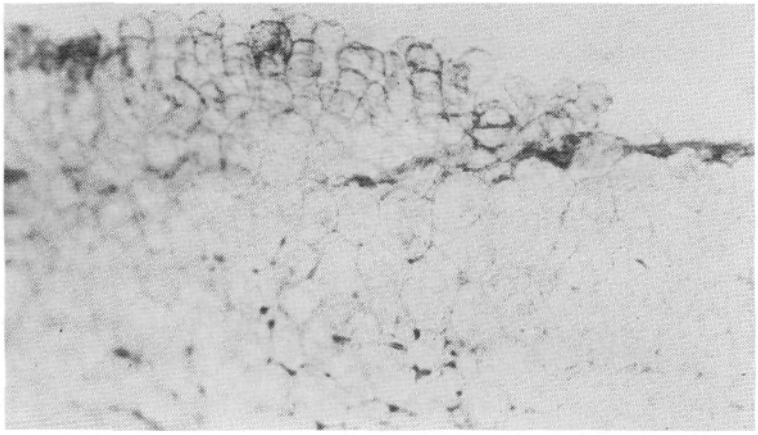

Figure 3. Proliferation of surface cells of avocado tissue grown in vitro. A—newly formed cells. B—original tissue.

The investigations now under way are directed toward the objective of learning the basic requirements of such excised tissue disks. These requirements can only be determined by extensive trials utilizing various media of different composition and subjecting the materials to various conditions of light, temperature, aeration, etc. The goal of all the studies is to maintain the tissues on a completely artificial media for an indefinite time so that effects of specific factors, such as beat or light, or a given mineral element can be determined. A knowledge of these basic requirements will be helpful in understanding and more intelligently pursuing the problems of fruit development encountered in the field and in handling processes.

These attempts to "tissue culture" explants from mature fruit tissues appear to be the first to result in success, as judged by reports in the literature (3). Previous attempts (4) to culture apple and other deciduous fruit tissues have given negative results, probably because of the fact that such-tissues lack the potentiality to undergo cell division as the fruit approaches maturity, or perhaps success might be attained with other techniques.

In the present experiments, tissue sections of avocado fruit have been maintained successfully in living conditions and showing evidence of cell division for periods greater than 5 months. These studies will be continued to determine the ultimate life length to learn of the basic requirements for growth of the tissues.

LITERATURE CITED

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