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CALLUS TISSUE GROWTH ON AVOCADO STEM SEGMENTS CULTURED ON ARTIFICIAL MEDIA

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In recent years the cultivation of plant tissues on artificial media has been extensively employed as a research tool in many fields of plant research. Black (1) and Morel (5), as well as many others, have described its utilization in plant pathological research. The writer's interest in the culture of avocado tissue on artificial media was engendered by the possibility that it might serve as an aid in research on the avocado sun-blotch virus disease.

Published reports include records of successful cultivation of avocado cotyledon tissue (3) and fruit pericarp tissue (6, 7). The present paper is a preliminary report on the growth of callus tissue on stem segments cultured in vitro.

MATERIALS, METHODS AND RESULTS

Both solid and liquid media were used in the initial experiments included in these studies. However, subsequent to the development of a satisfactory means of tissue support in a liquid medium (4), various liquid media were consistently used. The composition of the two types of media was essentially the same except that the solid medium contained 1.5 per cent washed Japanese agar.

Basically the media were composed of the ingredients normally required for plant tissue growth. The mineral component was qualitatively similar to the mineral nutrient solutions described by Gautheret (2) and by White (8). The vitamin and amino acid component included glycine, nicotinic acid, pyridoxine, thiamine, and calcium pantothenate. Sucrose in a concentration of two per cent was used as a carbon source, and in addition such growth promoting substances as alpha naphthalene acetic acid and coconut milk were sometimes used.

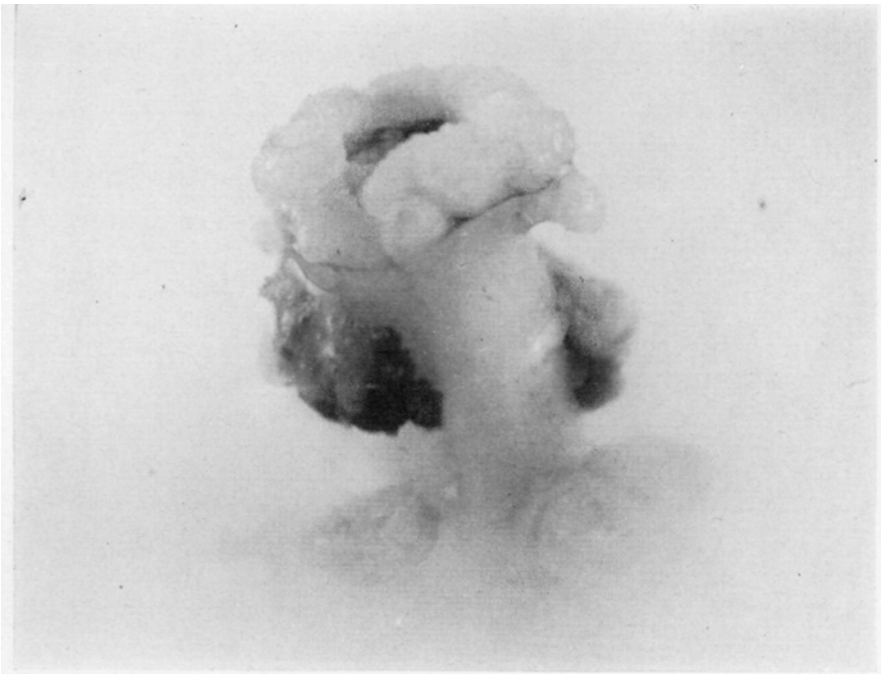
Wide-mouth flasks with a cotton plug were used for culture containers with solid media. The special flask recently described by Lange and Desjardins (4) consisting of a wide-mouth flask with a perforated test tube suspended in it was used for the liquid media cultures.

Early in these studies it was found that temperature was an important factor. Practically no callus tissue developed on stem segments in cultures incubated at room temperature (20°C), whereas fairly good growth usually developed when such cultures were incubated at 28°- 30°C. This was true of stem segments on both solid and liquid media.

Figure 1 illustrates a stem segment on the solid agar nutrient medium with the characteristic callus tissue that developed on the cut end and on areas below the cut end where the bark tissue was ruptured. Similar callus development was also obtained on stem segments in liquid medium cultures. In the early stages of growth the callus tissue had the appearance of a whitish, gelatinous mass of cells. Later when growth had apparently ceased, the masses of tissue became somewhat brownish in color and had the appearance of the hardened callus tissue sometimes found associated with cuts in the bark of whole young trees.

As shown in Figure 1, the callus growth at the cut end of stem segments formed circular masses of tissue which presumably originated from the cambial tissue between the bark and the wood of the stem. The growth of callus on the cut surface of stem segments split longitudinally also indicated that such tissue originated from the cambium.

In conjunction with the work on stem segment culture, some experimental attempts were made to culture avocado roots in a liquid medium. However, successful culture of the roots was not achieved.



An avocado stem segment on agar nutrient medium. Callus tissue has developed on the cut end of the segment and on areas below the cut end where the bark was ruptured. (x7, actual size.)

DISCUSSION AND SUMMARY

The results thus far obtained limit the writer's description to the macroscopic appearance of the callus tissue which developed on the stem segments and to the methods employed. The extent of cellular differentiation in such tissue will have to await microscopic study of stained tissue sections. Whether or not such callus tissue can be

further cultured in an isolated state of nutrient media will also have to be determined by future studies.

In the present experiments the development of callus tissue on avocado stem segments cultured in vitro has been very consistent when the cultures involved contained the proper nutrient medium and were incubated at 28°-30°C. Future investigations will be directed toward the initial objectives of the work; namely, to determine whether the culturing of avocado tissue on artificial media can be effectively utilized as a research tool in the study of the avocado sun-blotch virus disease.

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