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Seasonal Regeneration of Avocado Fruit Tissue in Vitro¹

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Several investigations have been conducted to obtain basic information concerning the factors associated with growth and development of the avocado fruit (1, 2, 3, 5). The present report is concerned with studies of tissue regeneration of fruit segments under controlled conditions. The major objective was to determine the seasonal trend in the regenerative capacity of the pericarp tissue as evidenced by growth response in tissue culture. Information concerning factors which affect growth of fruit tissue in any manner can be of value in understanding responses and behavior of the fruit structure.

MATERIALS AND METHODS

Single fruit samples of the Hass variety were collected at approximately 4-week intervals from a single tree in the university orchard, Los Angeles, during the period September, 1960 to October, 1961. Simultaneous collections were made of the newly developing crop during the overlapping period June to October, 1961, when the tree was bearing fruit from both the new and old crops.

The fruits were prepared for tissue culture under sterile conditions by surface flaming and slicing into 2 mm thick pieces from which cylinders 8 mm in diameter were punched with a cork borer (1). These standard sections were planted on modified Nitch's media (4) in screw top vials which were placed in controlled, constant temperature incubators at 20°C, 25°, 30° and 35°. Some vials were kept under continuous light (500 fc fluorescent) or wrapped in an aluminum foil to provide a dark environment. Each observation included 12 to 15 individual tissue segments from a given fruit. The dry weight of a similar sample was determined at the beginning of each experiment. The tissue cultures were incubated 4 weeks at which time the dry weight of the incubated sample was determined.

The regenerative capacity of the pericarp tissue can be estimated by determination of the increase in dry weight of tissue sections which have been grown in vitro utilizing tissue culture techniques. The growth response of the tissue under these conditions is expressed as a growth value which is determined as the ratio of final dry weight divided by original dry weight for any given sample. This allows determination of the comparative growth between any collections on a ratio basis.

RESULTS

The general seasonal trend in increase of dry weight per section of standard disk from the avocado fruit pericarp is shown in Fig.1. The observations were initiated in September, 1960, when the fruit for that season had attained an average length of

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approximately 80 mm and an average dry weight per section of 12 mg. The dry weight per section increased until complete fruit maturity in October, 1961, when the fruit was 100 mm in length with an average dry weight of 25 mg per section. It is apparent that at no time throughout the entire period of fruit development does the pericarp cease to maintain a capacity for cellular regeneration.

While the growth response of the older avocado tissue sections under different constant temperature conditions was essentially satisfactory at all levels, it appears that an optimum temperature for tissue regeneration was 25 °C. The data in Fig. 2 indicate that the higher temperature of 35 °C, while not lethal, did reduce the total growth of excised tissues markedly in all instances. Temperatures of 20 °C and 30°, respectively, allowed for satisfactory tissue growth and were not limiting.

The behavior of tissue explants from younger fruits of the new crop collected in summer, showed a more favorable response and growth value at all higher temperature conditions compared with tissue from the more mature fruit. Such comparatively younger tissues apparently are less sensitive to the higher temperature range under the experimental conditions.

Tissue sections maintained in complete and continuous darkness exhibited greater growth values than sections in light (Fig. 3).

DISCUSSION

It is evident that avocado pericarp tissue is capable of regeneration to a considerable degree at all times during the season. Comparable tissue samples from other fruit types such as the peach have shown very little regenerative response in vitro (6). This difference in behavior may be attributed to the evidence of continued mitotic activity in the pericarp tissue of avocado throughout the period of fruit development from fruit set to maturity (2) Cell division in the peach fruit in contrast is restricted primarily to the period of 3 weeks immediately following fruit set after which time little or no mitotic activity is discernable. Increase in fruit size of the peach later in the season is primarily the result of cell enlargement within cells previously formed.

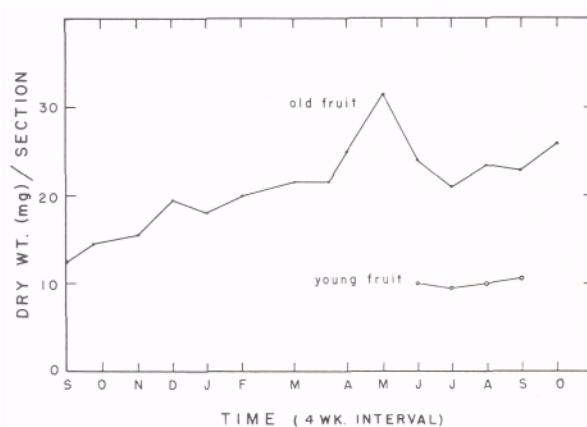


Fig. 1. Dry weight of avocado pericarp segments of standard size, sampled periodically throughout the season.

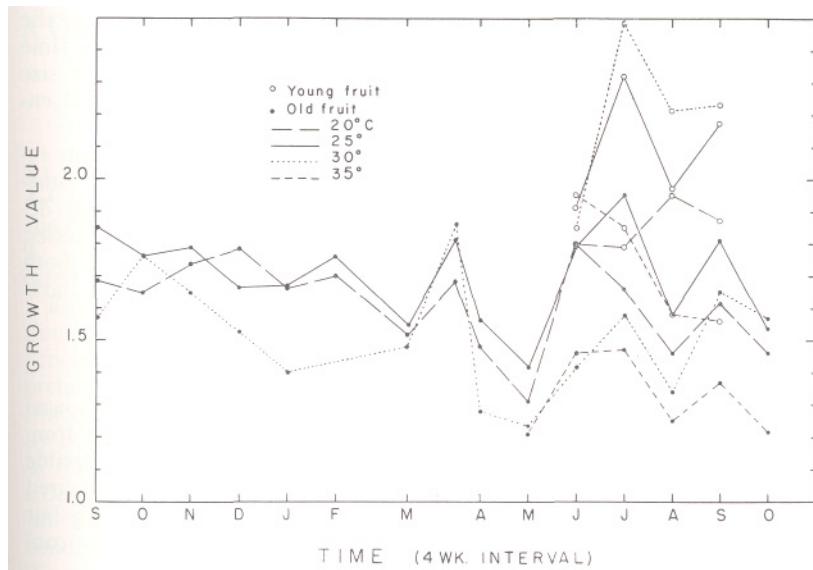


Fig. 2. Growth of avocado pericarp segments taken throughout the season and cultured at various temperatures in continuous darkness. Measurements are ratios of original dry weights to dry weights after culture.

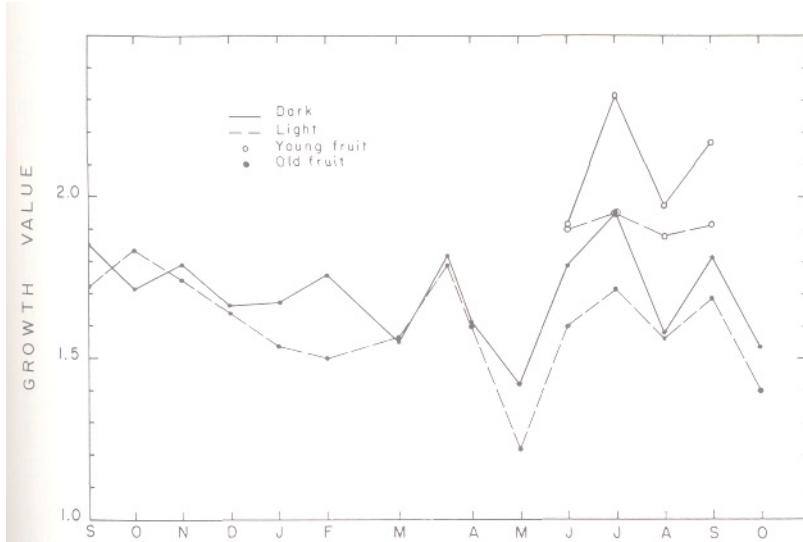


Fig. 3. Growth of avocado pericarp segments taken throughout the season and cultured in continuous light or darkness at 25°C.

While no distinctive seasonal pattern of tissue regeneration response was found in the avocado, tissue growth *in vitro* was somewhat more in sections taken from younger fruit. A gradual decrease in growth potential can be discerned as the season progresses, regardless of the temperature level. Explanted tissues from both young and old fruits develop more favorably in complete darkness *in vitro* irrespective of the

temperature.

SUMMARY

Avocado pericarp tissue grown in modified Nitsch's media showed sections from younger fruits proliferated more than sections from older fruits, though horticulturally mature fruits were still capable of regeneration. At 4 levels of temperatures tested, 25 °C appeared to be optimum, 20 °C and 30° satisfactory, and 35 °C limiting but not lethal. At a given temperature, sections grew better in continuous darkness than in continuous light.

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