1990 Summary of Avocado Research, pages 3-4 Avocado Research Advisory Committee University of California, Riverside

Molecular Genetics of Avocado

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<u>PROGRESS REPORT</u>: The primary objective of this project is to develop a large number of molecular markers to address a number of practical problems in avocado improvement. Our current and preliminary work firmly establishes the feasibility of molecular methods as applied to avocado genetics.

We have used cloned DNA probes for a cellulase gene, for the ribosomal RNA genes, and for chloroplast genes to study the genetic relationships among avocado varieties. Our previous work, which is now in press (Furnier et.al., Journal of Heredity, 1990), suggests that <u>Persea americana</u> var. <u>guatemalensis</u> has genetic markers from both P. <u>styermarkii</u> and P. <u>nubigena</u> which may indicate a hybrid origin for var. <u>guatemalensis</u>. Similarly, the rootstock cultivar G755 appears to be a hybrid of P. <u>americana</u> and P. <u>schiedeana</u> based on chloroplast DNA and ribosomal gene markers.

DNA has been prepared from 46 cultivars representing all three major varieties of avocado. Twenty of these samples have previously been screened using the cellulase probe. We found that some, but not all cultivars within varieties were identifiable based on RFLP patterns. We are currently using anonymous single-copy clones (described below) for further evaluation of cultivars.

A large number of DNA clones are required to provide genetic markers for parental identification and for genome mapping. To begin work towards these two objectives, we have cloned random avocado DNA fragments into a plasmid vector and are characterizing these for their usefulness as genetic markers. To date, 15 clones have been characterized. Eleven of these clones represent single-copy sequences that are polymorphic when tested against cultivars representing three major avocado varieties and are thus good RFLP markers. Three of the remaining probes are multiple-copy sequences that appear to be highly polymorphic between cultivars within varieties. These "hypervariable" probes may be especially useful in cultivar identification. We are now characterizing 17 more clones and expect to increase the number of single-copy probes to 20-25. In addition, there are approximately 50 more clones yet to be screened and characterized.

We have now begun using our probes to screen a panel of cultivars representing several commercial and experimental selections in order to address certain questions of parentage and cultivar identification which might aid the breeding program and the industry. DNA has also been prepared from approximately 85 Hass seedlings that can be regarded as F2 selfed progeny with respect to loci heterozygous in the parent. This material is now being used to test for genetic linkage among single copy probes.

<u>PRACTICAL APPLICATIONS AND SUMMARY</u>: Practical applications of this project include the use of DNA markers to (1) identify the genetic relationships among the major varieties of avocado; (2) to provide a precise genetic identification of various avocado cultivars within varieties; (3) to identify the parentage of specific cultivars; and (4) to begin a program to use restriction fragment length polymorphisms (RFLPs) to map the avocado genome.

Genetic markers will have substantial utility in avocado improvement because (a) they will permit the retrospective identification of lineages that have contributed to successful cultivars; (b) they will facilitate the genetic dissection of complex traits such as drought or salt tolerance, disease resistance and fruit quality; and (c) once the genetic basis of such traits is understood, markers can be used to monitor their genetic transfer in sexual crosses.