POST-PHLOEM TRANSPORT AND METABOLISM OF SUCROSE IN AVOCADO

Ryan F. Cripps

Submitted in partial fulfillment of the requirements for the degree of DOCTOR OF PHILOSOPHY IN AGRICULTURE in the School of Agricultural Science and Agribusiness, Faculty of Science and Agriculture, University of Natal, Pietermaritzburg

September 2001

ABSTRACT

In South Africa, and in several other sub/tropical countries, the avocado represents a commercially important crop. Very little is currently understood about the metabolism of sugars in this fruit. The variety 'Hass' is a popular cultivar that is grown extensively in South Africa. However this cultivar has a tendency to produce two distinct fruit phenotypes: a normal sized variant and a small, undersized variant. Current literature suggests that the small fruit phenotype is characterised by an elevated abscisic acid (ABA) to cytokinin ratio and altered isoprenoid metabolism. The results presented in the current investigation represent the findings from a detailed study into the metabolism and transport of sugars in 'Hass' fruit in an attempt to characterise solute allocation in developing avocado fruit. Furthermore, the activities of sugar metabolising enzymes, routes of solute movement and polyphenolic contents of normal, small and ABA-treated fruit were compared and contrasted to evaluate the potential role of ABA in the induction and expression of the small fruit phenotype.

The enzymes invertase, sucrose synthase (SSy) and sucrose phosphate synthase are involved in the metabolism of sucrose (Suc) and, hence, phloem unloading, post-sieve element transport and fruit growth. Although not the major sugar present, Sue was found in avocado phloem sap, and the enzymology for its metabolism was shown to exist in avocado fruit. It appears that sink strength is established during early fruit growth by high acid invertase activity, especially during the period of rapid cell division. As fruit growth progresses the activity of SSy and an enzyme responsible for the oxidation of perseitol (tentatively termed perseitol dehydrogenase) increases, suggesting that these enzymes play an important role in the supply of carbon during the linear phase of fruit growth. All Suc metabolising enzyme activity diminishes as the fruit approaches maturity.

With the exception of SSy (in the cleavage direction), all enzymes assayed showed a general increase in relative rates of activity in small and ABA-treated fruit. Similarly, ABA-treatment of seed coat discs *in vitro* resulted in the elevation of insoluble and soluble acid invertase, SSy (in the synthesis direction), and sucrose phosphate synthase activity. Furthermore, both small and ABA-treated fruit were characterised by elevated total soluble sugars, glucose and fructose levels. These observations suggest that altered sugar metabolism, as a consequence of changes in endogenous ABA

levels, may contribute to the occurrence of the small fruit.

The seed coat represents an import link between the seed, the mesocarp and the parental plant tissues. Loss of seed coat and endosperm integrity accompanied fruit maturation and a reduction in the movement of solutes into the seed. An increase in polyphenolics in the seed coat tissue seemed critical in this reduced movement. Both the small and ABA-treated fruit were characterised by early senescence of the seed coat, which was accompanied by both a loss of transport into and out of the seed and premature maturation of the fruit. This premature seed coat senescence appeared similar to programmed cell death in tissues exposed to stress or elevated reactive oxygen species, stimuli that are often accompanied by elevated ABA levels. Callose was localised to the plasmodesmata and is proposed to play a role in the gating of, and hence movement through, these pores. Small fruit were characterised by a loss of symplastic continuity, as represented by fewer plasmodesmata, and reduced callose degradation. Comparison of callose content and rates of synthesis suggest that ABA-treatment, similarly, reduces callose catabolism. The association of ABA with both the premature senescence of the seed coat and a reduction in symplastic Continuity, and, hence, a reduction in solute transport, further cements the potential role of ABA in the occurrence of the small fruit phenotype.