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Ultrastructural Studies of 'Kensington' Mango (*Mangifera indica* Linn.) Heat Injuries

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Abstract. Mature green 'Kensington' mango fruit were submerged in hot water at 46°C until the fruit center reached 45°C and then held for 30 minutes. The fruit were allowed to ripen for 7 to 10 days after the hot water treatment, and then damaged areas of skin and mesocarp tissue were prepared for observation by scanning and transmission electron microscopy. Heating-related injuries included rupturing the patterned cuticle and exocarp and exposing the underlying cells and hollow cavities (which varied in size and shape) randomly distributed within the mesocarp beneath the skin. Starch deposits still were present in the mesocarp parenchyma cells. The cell walls of damaged mesocarp parenchyma cells were convoluted and thickened in places. The injury suggested disruption of enzymes involved in carbohydrate metabolism.

Mangos are heat-disinfested in many countries to meet import-country requirements. Mexico, Florida, and Haiti treat 'Francis', 'Oro', 'Ataulfo', 'Tommy Atkins', 'Keitt', and 'Haden' with hot water for export to the U.S. mainland (Sharp, 1986; Sharp et al., 1988). External and internal injuries to a range of mango varieties have been reported following hot water and hot air treatments, including skin scalding, starchy layers forming beneath the skin, cavities within the mesocarp, and 'ricey' spots forming in the mesocarp around the seed (Esguerra et al., 1990; Jacobi and Wong, 1992). Smith and Chin (1989) found 'Haden' and 'Kensington' the most susceptible and 'Irwin' the least susceptible varieties to heat injury following hot water treatments of 42 to 48°C for 30 to 90 min.

To our knowledge, descriptions of such heat injuries to mango have been solely macroscopic. Our study was undertaken to examine the external and internal heat injuries to 'Kensington' mango using scanning and transmission electron microscopy.

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then cooled in ambient air. Each treatment group consisted of a minimum of 30 fruit. After treatment, fruit were stored at 22°C for 7 to 10 days until ripe. Tissue then was sampled for microscopic examination. Thin slices of damaged areas were excised and immediately immersed in 3% glutaraldehyde buffered in 0.066 M sodium cacodylate for 12 h. The tissue was postfixed in 1% osmium tetroxide for 1 h, dehydrated in a graded series of acetone, dried to the critical point, mounted on aluminium stubs, and sputter-coated with either platinum or gold before being viewed on a scanning electron microscope (SEM) (model 505; Philips, The Netherlands, or model 820; JEOL, Japan). Tissues for transmission electron microscopy were fixed in a similar manner to that for SEM. After dehydration in acetone, the tissues were embedded in Spurr's resin. Thin sections were stained with uranyl acetate and Reynolds lead citrate and viewed on a transmission electron microscope (TEM) (model H-7000; Hitachi, Japan, or model 400 T; Philips).

Results and Discussion

The nontreated, 'Kensington' mango fruit surface was covered by a thick, waxy, highly patterned cuticle (Fig. 1A). Pantastico et al. (1975) found that the thick and complex mango cuticle varied in thickness at various fruit development stages. The hot water treatment caused brown, damaged areas scattered over the mango surface. The patterned cuticle and exocarp of the scalded mango skin was ruptured, and underlying cells within the fruit mesocarp were exposed (Fig. 1B). The brown coloration might have been due to the action of the enzyme polyphenoloxidase (PPO), which increases in mango peel during ripening

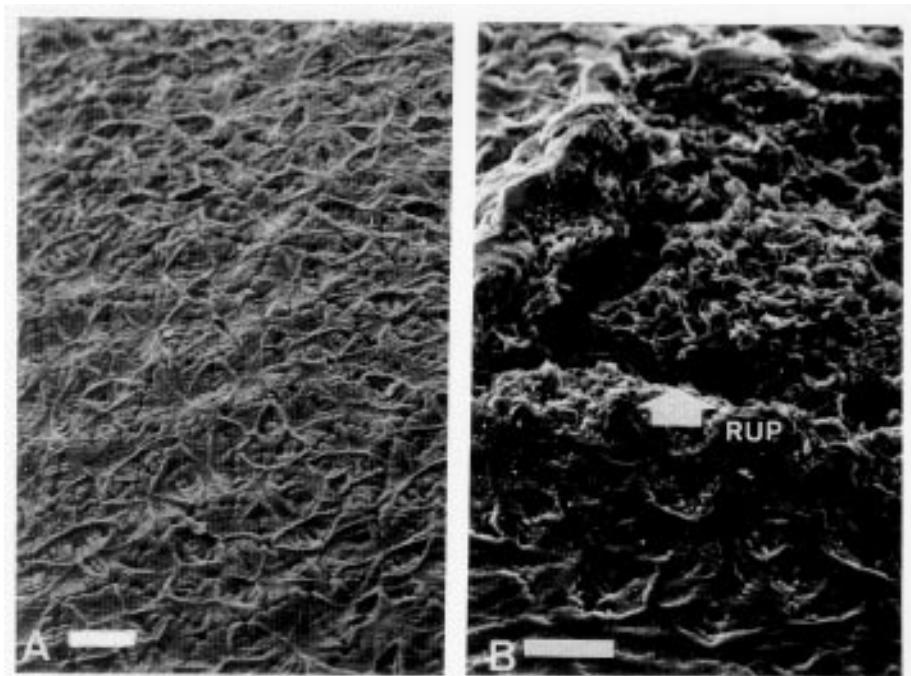


Fig. 1. Scanning electron micrographs of the patterned cuticles covering the skin of (A) a ripe nontreated 'Kensington' mango (bar = 0.1 mm) and (B) a ripe 'Kensington' mango skin ruptured by a hot water treatment until the fruit core was at 45°C for 30 min (bar = 0.1 mm) (RUP, rupture of cuticle).

(Prabha and Patwardhan, 1986). The SEM technique revealed tissue disruption in heated mango, presumably exposing cells to the PPO action, manifested as brown skin discoloration.

Nontreated ripe mango mesocarp consisted of thin-walled parenchyma cells with no starch granules (Fig. 2A). The starch granules present at the earlier stage of fruit development were metabolized into sugar during ripening

(Krishnamurthy and Subramanyam, 1973). Cavities and fissures formed in the mesocarp of heat-treated mango (Fig. 2B). The SEM showed that the cavities consisted of hollow spaces, randomly distributed within the mesocarp, 1 to 2 cm beneath the skin and of varying sizes and shapes. Parenchyma cells surrounding a cavity remained intact and appeared to be pushed aside throughout the tissue. Many cells

within the disrupted layer surrounding these cavities still had starch granules.

Mesocarp tissue from nontreated ripe fruit revealed turgid, polygonal parenchyma cells closely aligned to one another with thin walls (Fig. 3A). Cells walls were uniform in thickness, with the cytoplasm in a thin layer adjacent to the wall under a TEM. Heat-damaged mango mesocarp tissue showed parenchyma cells with cell walls convoluted and thickened in places (Fig. 3B). Starch granules were present freely within many cells. Such granules were absent in mesocarp tissue of non-treated mangos (Fig. 3A), confirming our SEM observations of the same tissue (Fig. 2).

The symptoms of mango heat injury documented in this study closely resemble the leatheriness in peaches caused by chilling injury (Luza et al., 1992). Injured peach fruit had markedly deformed internal mesocarp cells that had collapsed completely, leaving sharp protrusions of the cell walls, which became thickened. However, no evidence of separation of the plasma membrane from the cell wall or disintegration of the vacuolar membrane as in chill-injured peach cells was found in mango.

The ultrastructural changes occurring within tissues during ripening and the development of heat injury along with the redistribution of cellular components needs to be determined. Biochemical studies of the effect of hot water treatments on carbohydrate conversion could contribute to the understanding of the physiology of heated-treated fruit and the development of a nondamaging, commercial, heat disinfection treatment.

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Fig. 2. Scanning electron micrographs of (A) ripe 'Kensington' mango nontreated mesocarp (bar = 0.1 mm) and (B) ripe 'Kensington' mango mesocarp damaged by a hot water treatment until the fruit core reached 45°C for 30 min; cavitation evident (bar = 0.1 mm) (CUT, cuticle; P, parenchyma cell; C, cavity).

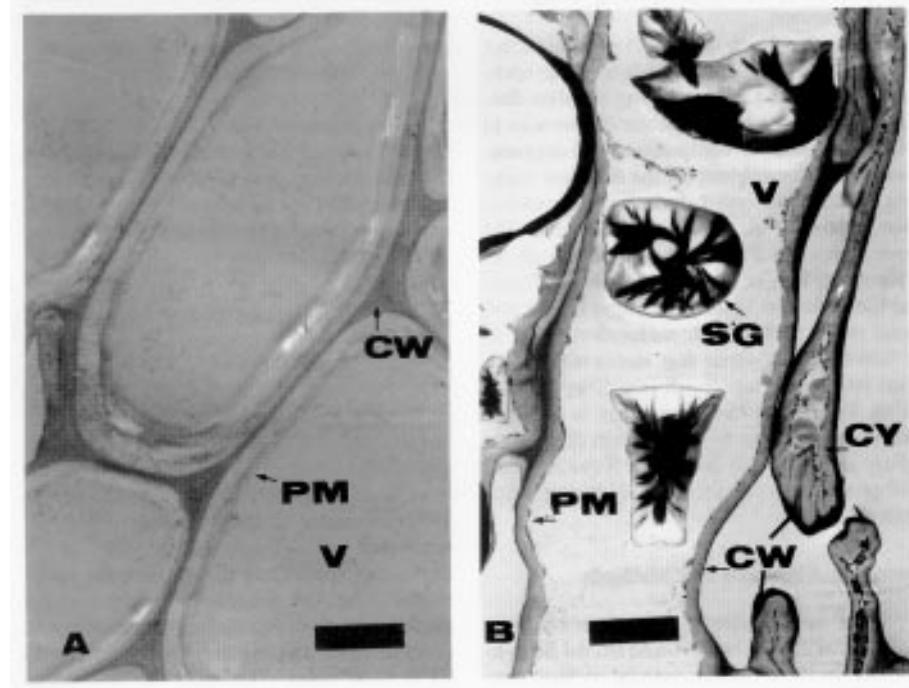


Fig. 3. Transmission electron micrographs of (A) ripe 'Kensington' mango nontreated mesocarp parenchyma cells (bar = 1.0 μm) and (B) ripe 'Kensington' mango mesocarp parenchyma cells following a hot water treatment until 45°C fruit core temperature for 30 min; starch granules and some cell wall convolution evident (bar = 1.0 μm) (CW, cell wall; PM, plasma membrane; V, vacuole; CY, cytoplasm; SG, starch granule).