

## Effects of hot water treatments on the physiology and quality of 'Kluai Khai' banana

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**Abstract:** Sucrier bananas (*Musa cavendishii* [Musa acuminata] AA group), locally known as 'Kluai Khai', were dipped in water at 45°C for 5, 10 and 15 min and 50°C for 10 min and then cooled by dipping in water at 25°C for 30 min. Fruits dipped in water at 25°C for 30 min were used as control. Samples were stored at room temperature (25 ± 2°C and 70-75% RH). Respiration rate as well as chlorophyll degradation were reduced by dipping fruit in water at 50°C for 10 min. Peel color change was retarded and fruit firmness was maintained by hot water treatments. The storage life of bananas dipped in hot water (45°C for 15 min or 50°C for 10 min) was significantly extended. The best fruit overall quality was obtained at 50°C for 10 min. Catalase and peroxidase activities of banana dipped in 50°C for 10 min were higher compared to the other treatments.

**Keywords:** Banana cv. 'Kluai Khai', hot water treatment, quality, catalase, peroxidase

### Introduction

Banana is a commercially important fruit crop of the world. However, its short shelf life seriously limits the marketing of the fruit. Extending banana shelf life could be a considerable commercial benefit to both exporters and retailers. Banana is usually harvested at a full mature stage for domestic consumption, but harvested unripe for the export market. In most cultivars of banana, senescence-associated skin spotting occurs later than pulp softening. However, for some cultivars, especially 'Sucrier' (*Musa cavendishii* [Musa acuminata] AA group) locally known as Kluai Khai, senescence skin spots develop as soon as the fruit turn yellowish-green, a stage that coincides with optimum flavor and aroma of the fruit flesh. This early development of spots on the skin of this cultivar is negatively perceived by consumers who wrongly attribute this characteristic to overripening or diseased fruits (Ketsa, 1996).

Many storage techniques have been developed to extend the shelf life and prolong freshness of banana for exporting purposes. Heat treatment is one of these postharvest techniques which has been used as a plant quarantine procedure in mango, apple, avocado, and litchi (Fallik, 2004). Over the last few years there has been increasing interest on the use of postharvest heat treatments (Lurie, 1998; Ferguson *et al.*, 2000; Fallik, 2004). Indeed, the overall quality of fresh produce treated with optimal hot water temperatures is significantly better than untreated produce, as determined by a sharp reduction in decay incidence and maintenance of several quality traits (Reyes *et al.*, 1998; Keryl *et al.*, 2001). Moreover, hot water

treatment has been proposed for fruit fly infestation (Fallik, 2004).

At this time, the response of 'Kluai Khai' banana to hot water treatment has not been much studied and more information is needed to understand the effects of this treatment on the physiological and biochemical changes of the fruit. This investigation was undertaken to evaluate the effects of hot water treatments on banana cv. 'Kluai Khai' and to determine the optimum hot water temperature and duration of exposure necessary to minimize the loss of fruit quality and to extend postharvest life.

### Materials and methods

#### Plant material

Entire bunches of 'Kluai Khai' were harvested at commercial maturity from a plantation in Petchaburi province, Thailand. Bunches were cut into individual banana hands (8-9 fruits/hand) and transported to the laboratory in corrugated cardboard boxes, within 2 h of harvest. Upon arrival at the laboratory, banana hands were immediately sorted and evaluated for initial skin colour. Uniform, unblemished, equal size and colored fruit in hands were selected and cleaned.

#### Hot water treatment

Fruit hands were then dipped in water at 45°C for 5, 10 and 15 min and 50°C for 10 min. Fruit hands were then cooled by dipping in water, mixed with 1,000 mg L<sup>-1</sup> benomyl at 25°C for 30 min. Fruit dipped in 1,000 mg L<sup>-1</sup> benomyl solution at 25°C for 30 min were used as a control treatment. Fruit

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were randomly sampled every 3 d to determine the physiological and biochemical changes.

#### *Fruit firmness measurement*

Fruit firmness was measured in Newtons (N) using a texture analyzer (Model TA-XT2). The fruit peel was removed before pressing a probe on the fruit (3 positions per fruit). Three fruits were used per replication.

#### *Determination of peel color changes*

Peel color changes were determined by measuring the hue angle value using a colorimeter (Minolta, Japan). Measurement was taken at the mid portion of each fruit.

#### *Determination of respiration rate*

Fruit respiration rate was measured by GC-8A Gas Chromatography (Shimadzu, Japan) with 80/100 mesh Porapak Q column and a thermal conductivity detector (TCD). Banana fruits in sealed plastic container were incubated at room temperature for 1 h. A 1 mL gas sample was withdrawn from the head space with syringe for carbon dioxide determination. The respiration rate was expressed as milligram of CO<sub>2</sub> kg<sup>-1</sup> hr<sup>-1</sup>.

#### *Determination of chlorophyll content*

Chlorophyll content was determined by the method of Arnon (1949). A 5 g sample of banana peel tissue was homogenized with 20 mL of 80% acetone. The mixture was filtered through Whatman No. 2 filter paper, adjusted to 100 mL volume with 80% acetone, spectrophotometric reading at 663 nm and 645 nm were taken. Total chlorophyll content was then calculated following the standard formula.

#### *Enzyme preparation and assay*

Two g of banana pulp were pulverized in a chilled homogenizer for 1 min with 12 mL of 0.1 M phosphate buffer (pH 6.1) containing 0.9 g polyvinylpyrrolidone (PVP) and 30 mg sodium ascorbate. The homogenate was centrifuged at 12,000×g for 15 min at 4°C. The supernatant was used for the catalase and peroxidase assay.

#### *Catalase assay*

Catalase activity was determined by the method of Aebi (1984) with minor modifications. Briefly, the reaction mixture contained 0.1 mL of enzyme extract, 0.1 mL of 10 mM H<sub>2</sub>O<sub>2</sub> and 2 mL of 30 mM potassium phosphate buffer (pH 7.0). After addition of the enzyme extract for 30 s, the absorbance at 240 nm was taken using a spectrophotometer model UV-1601 (Shimadzu, Japan). The enzyme activity was

expressed in units per mg protein.

#### *Peroxidase assay*

The reaction mixture consisted of 0.8 mL of enzyme extract and 2 mL of 0.4 M guaiacol. The reaction was started with the addition of 2 mL of 0.3 M H<sub>2</sub>O<sub>2</sub> and the reaction was carried for 5 min. The enzyme activity was recorded at an absorbance wavelength of 420 nm. The blank consisted of phosphate buffer 0.8 mL addition with 2 mL of distilled water and 2 mL of guaiacol. The rate of change in absorbance at 470 nm was measured with a spectrophotometer and the level of enzyme activity was expressed as ΔOD<sub>420</sub> min<sup>-1</sup> mg<sup>-1</sup> on a protein basis (Shannon *et al.*, 1966).

#### *Total protein analysis*

The protein content was measured according to the method of Bradford (1976). The Bradford dye reagent was prepared by diluting the commercial dye concentrate in a 1:4 ratio with distilled water. Five milliliters of diluted dye reagent was added to 100 μL of the supernatant and 100 μL extraction buffer was used as blank. After vortexing and incubating for 5 min, the absorbance was measured at 595 nm. The sample's protein content was determined by comparing to a standard of bovine serum albumin.

#### *Experimental design*

The experiment was conducted in a completely randomized design with three replicates, each consisting of 3 fruits per observation period. Standard error of the mean was calculated.

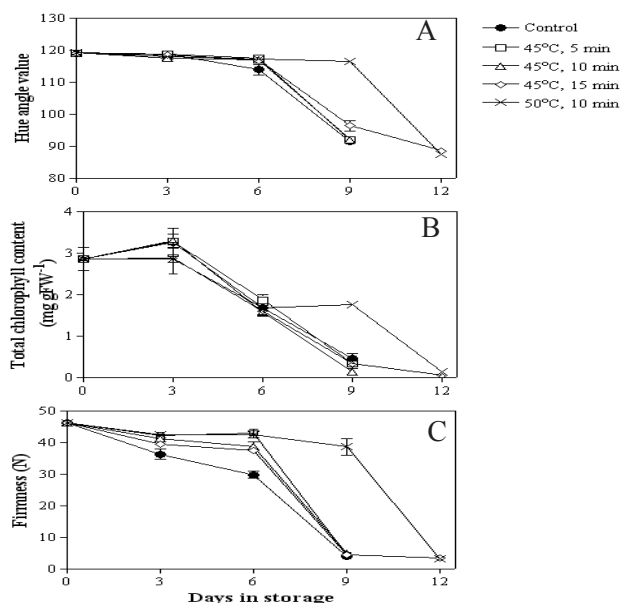
## **Results and Discussion**

#### *Peel color and firmness*

Fruit peel colour expressed as hue angle values, did not change until day 6 regardless of treatment (Figure 1A). Thereafter, hue values of fruit dipped in 45°C water for 5 and 10 min, and untreated fruit sharply declined indicating increased degree of yellowing. In contrast, fruits dipped in 45°C water for 15 min or in 50°C water for 10 min maintained higher hue values and more green colour than the other treatments. Hot water treatment at 50°C for 10 min was more effective maintaining higher hue values and hence delaying yellowing than at 45°C for 15 min. This was evident on day 9. Changes in hue values did not coincide well with those of chlorophyll content which started to decrease on day 6 in all treatments (Figure 1B). However, fruit dipped in 50°C water for 10 min had much higher chlorophyll content than the other treatments on day 9 coinciding with its hue

value which was highest among treatments.

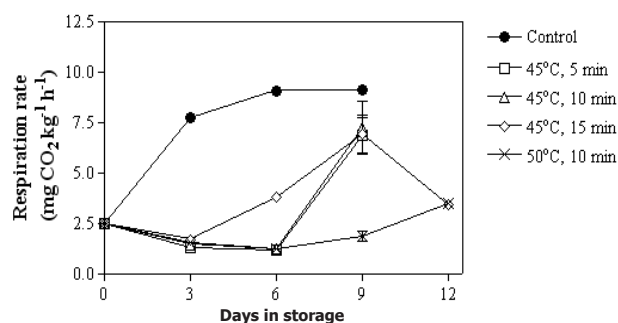
Fruit firmness decreased with storage at a faster rate in the untreated control than hot water-treated fruit (Figure 1C). Among hot water treatments, 50°C for 10 min was most effective in delaying firmness loss or softening. The fruit remained firm for 9 days and softened 3 days later whereas other hot water-treated fruit softened to the level of the control after 6 days of storage. It thus appears that hot water treatments, particularly 50°C for 10 min, effectively delay fruit softening and could be used as a treatment to extend the firm-fruit life for export purposes. Fallik *et al.* (2000) and Ilic *et al.* (2001) found that hot water treatment inhibited color development in melons and tomatoes and delayed their ripening as well. Polygalacturonase activity is decreased by the heat treatment in papaya over prolonged storage (Chan *et al.*, 1981). Lurie (1998) suggested that the reduction in fruit softening caused by hot water treatment was caused by the inhibition of pectin cell wall hydrolysis, indicative of a reduced level of cell wall degrading enzyme activity or of the inhibition of ethylene production due to a reduction in the activity of ethylene-forming enzyme. However, Jacobi and Giles (1997) reported that 'Kensington' mango fruit firmness decreased when the temperature or exposure time to the heat treatment were inappropriate. Our results confirm these observations whereby heated fruit at 45°C softened faster than those at 50°C (Figure 1C). Thus, the efficacy of heat treatment is time-temperature dependent. Furthermore, the results of the present study illustrate that during ripening of 'Kluai Khai' banana, loss of firmness (Figure 1C) preceded dramatic changes in peel color from green to yellow (Figure 1A).



**Figure 1** Changes in hue angle colour (A), chlorophyll content (B) and fruit firmness (C) of banana fruit dipped in hot water at 45°C for 5, 10 and 15 min, and 50°C for 10 min. Controls were dipped in room temperature water.

### Respiration rate

Respiration rate increased with storage in the untreated control (Figure 2). All hot water treatment delayed the onset of the respiratory rise by 3-6 days. Treatment with 50°C water for 10 min depressed the rate of respiration so that no distinct climacteric peak was evident during the 12-day storage period. However, the fruits were observed to ripen normally after day 12 (data not shown). The inhibitory effect of hot water treatment on respiration coincided with that on softening which further indicates retardation of ripening. Earlier, hot water treatment was shown to significantly lower the respiration and ethylene production rates of fresh produce (Fallik *et al.*, 1999). It has been suggested that heat treatment delay ripening and senescence in banana by activating stress-related oxidative defense enzymes (Jacobi *et al.*, 2001). Consequently, stress-associated changes in physiology and quality (e.g. respiration and chlorophyll degradation) are affected by the heat treatment. Indeed, dipping the banana in 50°C water for 10 min delayed the increases in both respiration rate and chlorophyll degradation resulting to increased and storage life of the fruit.



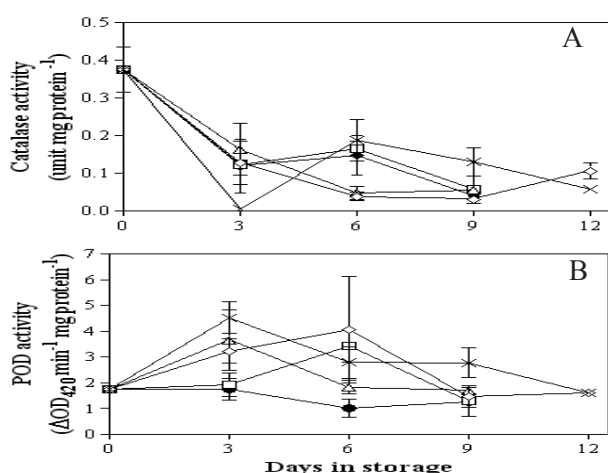
**Figure 2** Changes in respiration rate of banana fruit dipped in hot water at 45°C for 5, 10 and 15 min, and 50°C for 10 min. Controls were dipped in room temperature water.

### Catalase and peroxidase activity

Catalase activity in all treatments fell sharply after 3 days of storage (Figure 3A). Fruit treated with 50°C water for 10 min had the lowest catalase activity at this period but after 6-9 days of storage, catalase activity increased to levels comparable to that of the control. On the other hand, peroxidase (POD) activity increased in all hot water-treated fruit relative to that of the control (Figure 3B). The increase in POD activity was sustained throughout storage, particularly with 50°C water treatment.

The results showed no direct relationship between catalase activity and ripening changes while POD activity seemed to correlate well with ripening-associated physiological changes, in particular respiration rate and softening in response to hot water treatment. Increased POD activity has been reported

to be indicative of an increased capacity for free radical scavenging, reduced lipid peroxidation and diminished cellular membrane degradation, which possibly contributed to the ripening-delaying effect of heat shock or short heat treatment (Fallik, 2004). Optimum water temperature and time combination to induce such effect was 50°C, for 10 min. Thus, it could be a suitable treatment to retard ripening and prolong shelf life of the fruit.



**Figure 3** Changes in catalase (A) and peroxidase (POD) (B) activities of banana fruit dipped in hot water at 45°C for 5, 10 and 15 min, and 50°C for 10 min. Controls were dipped in room temperature water.

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