



# Changing of physiochemical properties and color development of mango fruit sprayed methyl Jasmonate



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## ABSTRACT

A key physical characteristic of mango cv. Manhachanok is its red color. The eye catching appearance good first impressions and can serve as a quality guide for consumers. This study was carried out to investigate the effectiveness of methyl jasmonate for enhancing uniform red color development in peel and for improving the quality of fruit in mango cv. Mahachanok. Methyl jasmonate at five concentrations (20, 40, 80 and 120  $\mu\text{L}/\text{mL}$ ) sprayed onto Trees 90 days after anthesis and compared with water. Fruit was harvested at 110 days after anthesis and stored at 15 °C. Physiochemical properties were then determined every three days. Mango fruit sprayed with 80  $\mu\text{L}/\text{mL}$  methyl jasmonate had significantly higher phenylalanine ammonia lyase activity (PAL), higher concentration of phenolic compounds and greater total anthocyanin concentration than the control fruit. Physiochemical changes measured during the postharvest period indicated that the application of MJ increased  $L^*$  and  $a^*$  values of peel and decreased the hue angle values compared with those of the control. Application of MJ enhanced  $\beta$ -carotene, vitamin C, glucose, fructose, sucrose and SS/TA concentrations compared with the control while firmness was increased and weight loss was delayed. Sensory evaluation panelists detected changes in color of fruit during storage.

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## 1. Introduction

Within the group of red mango (*Mangifera indica* L.) species, 'Mahachanok' is one cultivar which can be forced for off-season production in Thailand through the use of paclobutrazol. One problem in production of 'Mahachanok', however is that the red color of skin is uneven and fruit do not meet export standard. The red peel in 'Mahachanok' is caused by the synthesis and accumulation of anthocyanin. Color is a very important indicator of fruit quality and of freshness and also for estimating the stage of maturity. A number of factors and signals influence the accumulation of pigments, such as carotenoids and anthocyanins, including light, temperature, mineral, nutrition and the impact of plant growth regulators. In addition apart orchard management practices such as bagging, pruning and fertilization can also strongly impact on fruit color pigmentation. Pre-harvest and postharvest application of plant

growth regulators such as methyl jasmonate is a new approach to effectively maintain the quality of fruit. Specific plant growth regulators can increase anthocyanin concentration in the peel of mango and other fruits. The synergistic or additive responses have been found between ethylene and methyl jasmonate in apple peel pigment synthesis pathways (Kondo et al., 2001; Rowan, 2009). Such changes in synthesis can cause changes in physical appearance, mechanical properties and in the composition of bioactive compounds in fruits (Fan et al., 1997). Methyl jasmonate is an important cellular regulator that is involved in diverse developmental stages including fruit ripening, accumulation of pigments, phenolic compounds, antioxidants, and sugars (Cheong and Choi, 2003; Rohwer and Erwin, 2008; Heridia and Cisneros-Zevallos, 2009; Burhan et al., 2013) and specifically to increase  $\beta$ -carotene accumulation in ripening tomatoes (Saniewski and Czapski, 1983) and in mango cv. Nam Dok Mai (Buanong and Kanlayanarat, 2010). Dipping or spraying methyl jasmonate either before or after harvest influenced the development of red color through an increase in anthocyanin concentration in both apple (Rudell et al., 2005) and strawberry (Abolfazl et al., 2013). Dipping mango cv. Mahachanok in methyl jasmonate also enhanced the ability of skin to change to be more red in color (Chanwijit et al., 2010). In addition, fruits sprayed with

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methyl jasmonate, had a greater rate of growth, higher yield and higher fruit weight (Yilmaz et al., 2003). The aim of this study was to investigate the effectiveness of methyl jasmonate sprays, applied in a range of concentration pre-harvest, for enhancing red color development in peel, and increasing phenylalanine ammonia lyase (PAL) enzyme activity, the concentrations of phenolic compounds and of anthocyanins in mango cv. Mahachanok. Improving the quality of mango cv. Mahachanok fruit would increase the economic value of this product for both local sales and for export.

## 2. Materials and methods

### 2.1. Plant material and MJ treatment

Mango trees on rootstock Kaew varieties, approx. 10 year-old trees of 3–4 m canopy diameter size, cultivation distance 4 × 4 m, pruning type is open-center pattern, were selected within mango cv. Mahachanok export orchard, in the Noenmaprang district of Phitsanulok Province, Thailand. The experiment was arranged as a randomized complete block design (RCBD) divided into five treatments, with each treatment consisting of six replications (one tree each, total of 30 trees). Methyl jasmonate (Sigma–Aldrich, St. Louis, Mo, USA) was applied at five concentrations (20, 40, 80 and 120 µL/mL) was sprayed onto Trees 90 days after anthesis and compared with a control (water). Fruit were harvested 20 days later (110 days after anthesis) and processed using the same management practices that are used for export (i.e., cleaning, dipping in fungicides, placing in a net sleeve, and packing into a paper box) then stored at 15 °C, RH 90%. Physicochemical properties were determined at 90 (early immature stage), 100 (immature stage) and 110 (mature) days after anthesis and then every three days after harvest were randomly sampled and measured from each treatment (six replications, three fruits at each sampling time).

### 2.2. Fruit quality assessments

At each sampling, the color of peel was assessed on each individual fruit using a colorimeter (Minolta Model DP-1000) and expressed as  $L^*$ ,  $a^*$ ,  $b^*$  values along with color hue angle ( $^{\circ}H$ ). Firmness was measured using a texture analyzer (model QTS 25, Brookfield, USA) and results expressed in grams (g). For the determination of soluble solids content (SSC) and titratable acidity (TA), 200 g of mesocarp tissue was homogenized in blender and filtered through filter cloth. SSC was determined in the juice using a pocket refractometer (PAL-1, Atago, Japan) and expressed as %Brix. TA was determined in the juice with 0.1 NaOH to pH 8.2 using an automatic titrator (East Plus Titration, Mettler Toledo) and expressed as citric acid content, g per 100 g FW. The results were expressed as SSC/TA ratio. Juice was subsequently stored at 80 °C for analysis of glucose, fructose and sucrose content by high performance liquid chromatography (HPLC), using column (Inertsil® 5 µm NH<sub>2</sub> with guard column, 4.6 × 250 mm), RI detector, mobile phase: Acetonitrile: H<sub>2</sub>O (75:25).

### 2.3. β-Carotene and vitamin C assay

β-Carotene was measured using the method modified from Craft (2001). Each 10 g of pulp mango was homogenized with 15 mL 90% ethanol and 1 mL 60% KOH and sonicated for 15 min. Then 5 mL acetone, 15 mL hexane and 5 mL of 10% ascorbic acid were added and the mixture was incubated at 50 °C in a water bath for 30 min, shaken for 15 min, 50 mL deionized water added then shaker for 5 min, and allowed to settle then settled for 5 min. The hexane layer of each sample was transferred to a corresponding vial containing 50 mL methanol in 10% NaCl. Vials were capped and placed on an orbital shaker for 2 h. The hexane layer was then transferred to

another vial containing 50 mL deionized water, shaken and the process repeated twice more to remove KOH. Duplicate samples of the hexane layer were filtered using 0.45 µm PTFE syringe filters and the sample was assessed using HPLC column (Inertsil ODS-3) 5 µm 4.6 × 250 nm, mobile phase using acetonitrile: dichloromethane: methanol (70:20:10).

Vitamin C was assessed using HPLC (modified from Zapata and Dufour, 1992), using a column (Inertsil® ODS-3 5 µm 4.6 × 150 mm with guard column Inertsil ODS-3 4.0 × 10 mm), UV detector 248 nm, mobile phase: 3 mM potassium dihydrogen phosphate in 0.35% v/v ortho-phosphoric acid, with a flow rate of 1.0 mL/L, injector volume 60 µL, column temperature 40 °C.

### 2.4. Cell wall analysis

#### 2.4.1. Characteristics of anthocyanin in the cell wall

Observations of anthocyanin in the peel of mango fruit at different stages of fruit development were made using hand-sectioning with a razor blade and mounting with a drop of distilled water. The slides were examined in bright field using a light microscope (Carl Zeiss, Gottingen, Germany) equipped with a digital camera.

#### 2.4.2. Assay of phenolic compounds

Total phenolic compounds were extracted from mango fruit skin using the method described by Dewanto et al. (2002). Each 1 g of each sample was homogenized with 10 mL 80% methanol. The aqueous extracts were centrifuged at 12,000 × g for 20 min at 4 °C to remove the pellet. After that, the concentration of phenolic compounds in the alcohol extract was assessed using the Folin–Ciocalteu method of Slinkard and Singleton (1977). To 2 mL of sample or gallic acid, 10 mL of Folin–Ciocalteu reagent was added, mixed and incubated for 6 min at room temperature prior to addition of 8 mL of 7.5% Na<sub>2</sub>CO<sub>3</sub> solution. This mixture was then allowed to stand for 120 min at room temperature, and the absorbance of the supernatant was measured at 765 nm, using a UV mini-1240 spectrophotometer (Shimadzu, Japan). Total phenolic compounds were estimated as gallic acid equivalents (mg/100 g fresh weight).

#### 2.4.3. Assay of PAL enzyme activity

PAL enzyme activity in peel was measured using the method of Zucker (1965) and Lister et al. (1996) with slight modification. The reaction mixture comprised 1 mL of enzyme extract 1 mL 50 mM phenylalanine and 1 mL 150 mM Tris buffer (pH 8.4) and incubated at 37 °C in a water bath for 2 h. PAL enzyme activity was assessed by measuring absorbance at 290 nm using a UV mini-1240 spectrophotometer (Shimadzu, Japan). Triplicate assays were performed for each enzyme extract. One unit of PAL was defined as the yield of 1 µmol of *trans*-cinnamic acid per mg of protein.

Protein content in the enzyme extracts was measured using the method of Lowry et al. (1951), using bovine serum albumin (BSA) as a standard (0, 100, 200, 300, and 400 mg/mL). Specific activity of the enzyme was expressed as units per mg protein per hour.

#### 2.4.4. Anthocyanin determination

Total anthocyanins were extracted from mango fruit skin using the method described by Ranganna (1977). Each 3 g sample was homogenized with 30 mL of ethanoic HCl (85:15 v/v) and then shaken overnight at 4 °C in darkness and then adjusted to a final volume of 50 mL with ethanoic: HCl. The aqueous extracts were centrifuged at 10,000 for 10 min at 4 °C to remove the pellet. The absorbance of the supernatant was measured at 535 nm, using a UV mini-1240 UV-vis spectrophotometer (Shimadzu, Japan). Total anthocyanin concentration was expressed as mg/100 g fresh weight.

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