Storage behaviour and quality responses of mango (*Mangifera indica* L.) fruit treated with chitosan and gum arabic coatings during cold storage conditions

1Khaliq, G., 1Mohamed, M.T.M., 1Ding, P., 2Ghazali, H.M. and 3Ali, A.

1Department of Crop Science, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia
2Department of Food Science, Faculty of Food Science and Technology, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia
3School of Biosciences, Faculty of Science, The University of Nottingham Malaysia Campus, 34500 Semenyih, Selangor, Malaysia

Abstract

Natural products are useful for delaying the ripening process, preserving quality and reducing biochemical changes in fruits. Effect of gum arabic (GA) 10% and chitosan (CH) 1% edible coatings on physiological and biochemical properties of mango (*Mangifera indica* L. cv. Choke Anan) fruit were investigated. Mango fruit were stored at 13°C and 80% relative humidity for 28 days. Significant (P≤0.05) differences were observed in fruits treated with GA 10% and CH 1% as compared to the control. The results showed that GA 10% and CH 1% treatments significantly reduced weight loss than the control fruits. The application of CH 1% coating effectively inhibited the increase in soluble solid concentration (SSC), respiration rate and ethylene production. But no significant differences were observed in terms of ascorbic acid loss between treated fruits and control during the entire storage period. Furthermore, the combined application of GA 10% + CH 1% alleviated decay incidence and retained high firmness of mango fruit. These results suggested that application of GA 10% coating combined with CH 1% as a bio preservative might be a simple and effective technique for delaying ripening and maintaining quality of mango fruit during cold storage without the use of fungicides.

Keywords

Mango
Gum arabic
Chitosan
Postharvest qualities
Cold storage

Introduction

Mango (*Mangifera indica* L.) is a tropical climacteric fruit and ripens very fast in harsh climatic conditions. Best quality is retained at the lowest possible storage temperature tolerated by the product. Postharvest diseases and short shelf life are the major problems of mango fruit, which affecting its quality (Zheng *et al*., 2012). Various biochemical changes during the ripening process affect fruit composition and quality. Soft texture of mango fruit limits the postharvest life and increase susceptibility to various pathogenic infections. Several techniques have been used to reduce deterioration, extend the shelf life and maintain quality of mango fruit, including low temperature, controlled or modified atmosphere storage, hypobaric storage, chemicals, irradiation and coatings (Ravindra and Goswami, 2007). Refrigeration storage has been shown to be an effective method to maintain postharvest quality and extend the shelf life of mango fruit (Mitra and Baldwin, 1997). However, mango fruit are susceptible to chilling injury, when stored below 13°C (Nair and Singh, 2003). Controlled atmosphere reduced physico-chemical changes and delayed the ripening process of mango fruit (Sudhakar Rao and Gopalakrishna Rao, 2008), but can cause poor colour, undesirable flavour and physiological disorders (Thompson, 2001). Continuous use of fungicides has been used to reduce postharvest decay and extend the shelf life of fruit. However, fungicide resistance by pathogens, consumer concerns about the residue of fungicides on the fruit surface and its impact on the environment, has needed the development of consumers and environment friendly bio preservative (Charles *et al*., 1994; Mari *et al*., 2014).

Natural products are useful and taking place as an alternative approaches for delaying ripening and reducing postharvest deterioration of fruit (Tripathi and Dubey, 2004). Edible coatings have been studied for keeping quality and extending the shelf life of fruits. Coatings act as a barrier on the fruit surface, and
control the internal gas atmosphere, decreasing water loss and delaying fruit ripening (Bourtoom, 2008). Edible coatings are used as postharvest managements to retain fruit quality and minimize the size of non-biodegradable packaging materials. Preservation of fruit quality has been attained by using some edible coatings, such as aloe vera gel in nectarine (Ahmed et al., 2009), pectin in mango (Moalemiyan et al., 2012) and alginate in plum (Valero et al., 2013).

Gum arabic is a polysaccharide natural secretion from Acacia species and used in industries for film forming, emulsification, and encapsulation properties. Gum arabic coatings effectively delayed ripening and extended the storage life of tomato during storage (Ali et al., 2010). Coating with gum arabic plus natamycin potentially improved the quality and enhanced the storage life of shiitake mushroom as reported by Jiang et al. (2013). Chitosan is a natural biopolymer containing (1,4)-linked 2-amino-2-deoxy-β-d-glucan, derived by deacetylation of chitin (Aider, 2010). It has a wide range of uses in fruit and vegetable, because of its film forming, biochemical properties and antimicrobial activity (Devlieghere et al., 2004). Therefore, the objective of this work was to elucidate the effect of gum arabic and chitosan pre-storage treatments on delaying ripening, enhancing the shelf life and retaining quality of mango fruit during cold storage.

Materials and Methods

Plant materials

Mature and hard green mango (Mangifera indica L. cv. Choke Anan) fruit were obtained from a commercial orchard located in Ipoh, Perak state of Malaysia. Fruits were transported with minimal delay after harvest and brought to the postharvest laboratory within a day. Only the firm and well developed fruits of uniform size and maturity, free from pest, disease, injuries, bruises and blemishes were selected for the experiment. Chitosan from crab shells (high molecular weight, viscosity>400 mPa.s) and gum arabic (from Acacia tree) powder were purchased from Sigma Chemicals Co., USA.

Preparation of dipping solutions

Gum arabic solution 10% (w/v) was prepared by dissolving 10 g of gum arabic powder in 100 mL distilled water. The solution was heated at 40°C for 60 min, using a hot plate magnetic stirrer (Model SP 18420-26 Barnstead thermolyne 2555 Kerper Boulevard Dubuque, USA). Glycerol 1.0% was also added as a plasticizer to the coating solutions. The pH of the solution was adjusted to 5.6 with 1 mol L⁻¹ sodium hydroxide (NaOH) using a pH meter (GLP 21, Crison, Barcelona). Chitosan coating solution 1% (w/v) was prepared by dissolving 1 g of chitosan in 100 mL distilled water containing 1% (v/v) glacial acetic acid. The solution was agitated constantly using a magnetic stirrer for 3 h. The suspension was filtered through cheese cloth in order to eliminate the insoluble material. The pH of the solution was adjusted to 5.6 with 1 mol L⁻¹ NaOH, and 0.2 mL of Tween-20 also added to improve wettability. The composite coatings solution was prepared in the same way by dissolving 10 g of gum arabic powder plus 1 g of chitosan in 100 mL distilled water containing glycerol and glacial acetic acid. The solution was stirred for 30 min and the pH of solution adjusted to 5.6 with 1 mol L⁻¹ NaOH using a pH meter.

Treatments for the experiment were: (1) distilled water as a control, (2) gum arabic 10%, (3) chitosan 1%, and (4) gum arabic 10% + chitosan 1%.

Fruits were washed with 0.01% sodium hypochlorite for 2 min, and dried in air at ambient temperature. The mango fruit were randomly divided into four lots of 60 fruits each. All the treatments were conducted with three blocks. One of the four dipping treatments was then applied to each lot. The first lot was dipped in distilled water containing Tween-20 as a control. The other three lots were dipped in the corresponding solutions for three minutes. After dipping, fruits were air-dried for one hour. All fruits were packed in plastic boxes (40 × 30 × 12 cm) covered with polyethylene plastic film 0.02 mm thickness to maintain relative humidity (RH) of about 80%. To avoid modifying the atmosphere around the fruit, five holes of 7 mm in diameter were made in the plastic film, and then the fruits were stored at 13 ± 1°C for 28 days. The data were collected before treatment (day 0) and at 7 day intervals for 28 days during cold storage. Twelve fruits of each treatment were sampled at each interval.

Determination of weight loss

Three fruits in each block were separately marked before storage and weighed at the start of the experiment. The same fruits were consistently weighed at each sampling interval during the whole storage period. The result was expressed on a percentage basis.

Measurement of fruit firmness

Fruit firmness was assessed by using Instron Universal Testing Machine (Model 5543 PS995, USA) connected with computer. The force required to penetrate 10 mm inside the fruit using a probe diameter of 6 mm. The machine was set with
compression mode at the speed of 20 mm min⁻¹. Reading was recorded on 3 points in the equatorial region of the whole fruit with skin removed, and the result was expressed in term of force recorded in Newton (N).

Decay incidence (DI)

The degree of decay incidence was evaluated as described by Zheng et al. (2007) with slight modification. The fungal or bacterial growth symptoms on the fruit surface were observed visually by using a scale where 0 = no signs of decay, 1 = 1–10% decay, 2 = 11–25% decay, 3 = 26–40% decay, 4 = 40–50% decay, 5 = > 50% decay. Decay incidence was calculated using the following formula:

Decay incidence
= \frac{100 \times \sum (DI \text{ level}) \times \text{(number of fruit at the DI level)}}{\text{(total number of fruit in the treatment \times the highest score)}}.

Determination of soluble solid concentration

Soluble solid concentration was measured by using a pocket Pal 1 refractometer (Atago, Japan) and expressed as a percentage (°Brix). Before taking reading, it was standardized with distilled water and adjusted to reading 0°Brix. Pulp tissue 10 g were homogenized with 40 mL distilled water in the blender jug, and filtered with cotton wool. Then, two drops of filtrate were placed on the glass prism of refractometer and recorded reading. The reading was corrected to a standard temperature of 20°C by adding 0.28% to get SSC at 27°C. SSC was determined according to the following formula:

SSC%
= \frac{(\text{refractometer reading} \times \text{dilution factor})}{0.28}.

Ascorbic acid

Ascorbic acid was measured following the procedure of AOAC (2000). Ten grams of the fruit pulp was homogenized with 40 mL of 3% metaphosphoric acid by using a blender. The mixture was filtered with cotton wool. Then, 5.0 mL of filtrate were taken and titrated against freshly prepared dye solution (2, 6-dichlorophenol-indophenol) until pink colour and the result was expressed as mg 100⁻¹ g on a fresh weight basis (FW).

Respiration rate and ethylene production

Three fruits from each block were randomly selected to measure respiration rate and ethylene production. Single fruit was kept in 1.9 L airtight container at 20°C for 2 h. For determination of CO₂ and C₂H₄ concentration, one millilitre of gas from the sealed container was injected into a gas chromatograph (Claru-500, Perkin Elmer, USA) fitted with a stainless steel Porapak Q column (3 m × 3.125 mm, 50/80 mesh). Thermal conductivity (150°C) and flame ionization detectors (150°C) were used for CO₂ and ethylene quantification, respectively. Nitrogen served as the carrier gas at a flow rate of 45 mL min⁻¹. One millilitre certified standard gas was used for calibration the instruments. The amount of CO₂ and C₂H₄ production was expressed in mL kg⁻¹ h⁻¹ and µL kg⁻¹ h⁻¹, respectively.

Statistical analysis

The experiment was conducted in a randomized complete block design. There were three blocks for each treatment and each block contained 20 fruits. At each sampling date 4 fruits were taken at random from each block. The data were subjected to analysis of variance (ANOVA) using the statistical analysis software (SAS, version 9.3, SAS Institute Inc., Cary, North Carolina, USA). Fisher’s least significant differences (LSD) were calculated following a significant (P≤0.05) F-test.

Results and Discussion

Weight loss

Weight loss is used as a quality index in the postharvest life of fruits. The results showed that weight loss gradually increased in all samples during storage (Figure 1A). Weight loss of treated mango fruit was significantly lower than untreated fruit during the entire storage period. The highest weight loss was observed in the control fruits at the end of storage period. These results are in agreement with the findings of previous studies, in which weight loss was reduced with gum arabic treatment in tomato (Ali et al., 2010), composite coating of gum arabic plus chitosan in banana (Maqbool et al., 2011) and chitosan combined with pectin in mango (de S. Medeiros et al., 2012). Weight loss reduction in mango fruit treated with carnauba wax coating could be due to the effects of coating which created a modified atmosphere, and restricted gas exchange and moisture (Baldwin et al., 1999). Loss of weight in fruits occurs due to transpiration process, depends on the gradient of water vapour pressure between the surrounding atmosphere and the fruit tissue. Edible coatings act as a barrier on the fruit surface, thereby reducing water transfer, sealing small wounds and thus delaying weight loss. Mango fruit coated with GA 10% and CH 1% may be formed a thin layer of
film on the fruit surface, and thus reduced moisture loss from the fruit surface. The use of CH alone or combined with GA reduced weight loss and avoided the fruits from shrivelling in this experiment.

Figure 1. Effect of gum arabic and chitosan on weight loss (A) and decay incidence (B) of mango fruit during storage at 13°C for 28 days. Vertical bars indicate standard error of means for three replicates.

**Firmness**

Ripening of mango fruit is considered by textural softening during storage. Mango fruit firmness was reduced in all samples with the progress of storage period (Table 1). Firmness of the control fruit rapidly decreased with increasing the storage period. From day 7 up to the end of experiment, treated mango significantly retained higher firmness over control fruits. Several studies confirmed that edible coating retained high firmness in mango fruit (Kittur et al., 2001; Zhu et al., 2008; Moslemiyani et al., 2012). It has been observed that combined treatment of gum arabic and chitosan effectively retained high firmness of banana fruits (Maqbool et al., 2011). In the same way, shiitake mushroom treated with gum arabic 10% and natamycin maintained high firmness during storage (Jiang et al., 2013). Fruit texture properties are affected by cell turgidity, changes in cell structure and cell wall composition (Seymour et al., 1993). Fruit softening is related to water loss, which is responsible for the decrease of cell turgor of fresh fruit (Yang et al., 2014). The reasons for GA 10% and CH 1% coatings that reduced the moisture loss of mango could be explained by their capacity to work as barriers to water vapour and thus retained high firmness.

The pectin substances are responsible for the cohesiveness of the fruit and the main components of the middle lamella as well as structural elements in the primary cell wall. Degradation of cellular material and pectin results in textural softening of fruits (Cheng et al., 2009). It has been well known that softening of fruits can be related to the enzymatic hydrolysis of cell wall components. Controlled or modified atmosphere delayed the ripening process and reduced physico-chemical changes in fruits (Yahia, 2009). In this study, the fruits treated with GA 10% and CH 1% coatings showed more firmness than the control. These coatings might be created a modified atmosphere around the fruit surface as a result reduced changes in pectin substances and thereby delayed the textural changes.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>0</th>
<th>7</th>
<th>14</th>
<th>21</th>
<th>28</th>
</tr>
</thead>
<tbody>
<tr>
<td>Firmness (N)</td>
<td>Control 97.66a</td>
<td>80b</td>
<td>61 c</td>
<td>45.33c</td>
<td>20 c</td>
</tr>
<tr>
<td></td>
<td>GA 10% 98a</td>
<td>95.66a</td>
<td>85 ab</td>
<td>74 ab</td>
<td>65 a</td>
</tr>
<tr>
<td></td>
<td>CH 1% 99.66a</td>
<td>92.33a</td>
<td>80 b</td>
<td>63.66b</td>
<td>45 b</td>
</tr>
<tr>
<td></td>
<td>GA 10% + CH 1% 98.33a</td>
<td>97a</td>
<td>90.33a</td>
<td>80.66a</td>
<td>72 a</td>
</tr>
</tbody>
</table>

Means with the same letters within a column are not significantly different (P<0.05) using LSD. Each value is the mean of three replicates.

**Decay incidence**

Postharvest diseases and fast ripening are the main causes of mango fruit deterioration. The decay incidence was observed after 7 days of storage and gradually increased in all fruits with progressing the storage time (Figure 1B). From day 14 until the end of experiment, the decay incidence of treated mango was significantly inhibited than the control fruits. However, no significant differences can be seen between CH 1% and GA 10% + CH 1% treated fruits throughout the storage period. The highest decay percentage was recorded in the control samples at the end of storage period. It has been observed that coating has the ability to prevent the growth of fungi in wide horticultural produces (Tripathi and Dubey, 2004). Several strategies have been evaluated by using natural bio preservatives instead of fungicides to inhibit decay development and delay the ripening
process of mango fruit. Chitosan has strong antimicrobial and antifungal properties and helps to prevent postharvest fruit diseases (Elsabee and Abdou, 2013). Wongmetha and Ke (2013) reported that chitosan combined with 1-methylcyclopropene treatment extended the storage life of ‘Irwin’ mango at 10°C up to 32 days. Gum arabic in combination with calcium chloride reduced decay incidence of mango fruit during storage (Khaliq et al., 2015). Similarly, polysaccharide based treatment and carnauba wax coating reduced decay in ‘Tommy Atkins’ mango fruit (Baldwin et al., 1999). The reduction of decay incidence with GA 10% and CH 1% may be due to its film-forming property, which acted as a fence and thus reduced microbial activity.

**Soluble solid concentration**

Soluble solid concentration is a maturity index and also used for quality measurement. The ratio of sugar to acid plays a significant role in the determination of ripeness stage and taste of the fruit. Soluble solid concentration gradually increased in all samples regardless of dipping treatments with increasing the storage time (Figure 2). From day 7 up to the end of experiment, treated fruits significantly retained lower SSC than the control. These results revealed that CH 1% and GA 10% coating treatments efficiently reduced the rapid changes in SSC during the entire storage period. It has been reported that SSC increased with progressing the storage time in nectarine (Ahmed et al., 2009) and mango (Zheng et al., 2012; Li et al., 2014). These data support previous reports, where a minor increase in SSC has been reported in shiitake mushroom treated with gum arabic plus natamycin (Jiang et al., 2013) and chitosan coated mango (Kittur et al., 2001; de S. Medeiros et al., 2012). The increase in SSC could be attributed by the breakdown of carbohydrate into simple sugar and glucose (Kittur et al., 2001). During the ripening process, starch hydrolyzed into simple sugars, where glucose, fructose and sucrose are dominant in ripe fruits (Ito et al., 1997). The activities of sucrose synthase, invertase and amylase enzyme increased and hydrolysed the starch to sucrose (Kumar et al., 1994). Edible coating created a semipermeable film around the fruit and modified the internal atmosphere by increasing CO₂ and decreasing O₂ production (de S. Medeiros et al., 2012). The low respiration rate reduces the use of metabolites, resulting in lower SSC consumption and slow conversion of carbohydrates to sugars. A possible explanation for the lower SSC in mango fruit treated with GA 10% and CH 1% might be due to suppression of respiration and metabolic activity, thereby delayed the conversion of starch into sugars. Mango fruit treated with GA 10% and CH 1% reduced changes in SSC, thus these treatments retained the fruit quality and protected the fruit from quick deterioration during storage.

**Ascorbic acid**

Fruits are a natural source of ascorbic acid and it is known that its level decreases during the ripening process. In general, a gradual decline in ascorbic acid was observed during storage in both treated and untreated mango fruit as indicated in Table 2. The highest loss of ascorbic acid was recorded in the control fruits at the end of experiment. However, no significant differences were observed between treated fruits and the control during the whole storage period. It has been observed that edible coatings had no significant effect on ascorbic acid of mango fruit during storage (Hoa and Ducamp, 2008). Contrary to other organic acids, ascorbic acid is quite unstable. This instability is mainly due to the activity of ascorbate oxidase enzyme and the reaction with oxygen in the presence of heavy metal ions and light (Bode et al., 1990). Therefore, these dipping treatments might be insufficient to prevent losses of ascorbic acid in mango fruit at this storage temperature. Losses of ascorbic acid are common in different fresh fruits during storage. A sharp decrease in ascorbic acid was observed in fresh-cut and whole ‘Ataulfo’ mango during storage for 15 days (Robles-Sánchez et al., 2009). Similarly, in ‘Brokin’, ‘Julie’ and ‘Peter’ mango varieties, ascorbic acid continuously decreased during storage at ambient temperature stored for 12 days (Faasema et al., 2014). Zhu et al. (2008) reported that ascorbic acid was declined in control fruits and chitosan treated mango throughout the storage period.
Table 2 Effect of gum arabic and chitosan on ascorbic acid of mango fruit during storage at 13°C for 28 days.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Storage days</th>
<th>0</th>
<th>7</th>
<th>14</th>
<th>21</th>
<th>28</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ascorbic acid (mg 100⁻¹ g FW)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td>19.93 a</td>
<td>16.42 a</td>
<td>14.46 a</td>
<td>13.03 a</td>
<td>11.83 a</td>
</tr>
<tr>
<td>GA 10%</td>
<td></td>
<td>21.16 a</td>
<td>19.59 a</td>
<td>18.04 a</td>
<td>16.63 a</td>
<td>16.06 a</td>
</tr>
<tr>
<td>CH 1%</td>
<td></td>
<td>21.10 a</td>
<td>18.81 a</td>
<td>17.44 a</td>
<td>16.36 a</td>
<td>15.03 a</td>
</tr>
<tr>
<td>GA 10% + CH 1%</td>
<td></td>
<td>20.12 a</td>
<td>17.16 a</td>
<td>15.5 a</td>
<td>15.20 a</td>
<td>13.85 a</td>
</tr>
</tbody>
</table>

Means with the same letters within a column are not significantly different (P≤0.05) using LSD. Each value is the mean of three replicates.

Respiration rate and ethylene production

Respiration rate and ethylene production are indicator of metabolic activity and give signals of the possible shelf life of the product. Respiration rate decreased, initially in the first 7 days of storage in all treated and untreated mango fruit, and afterward a sharp increase of respiration rate was observed (Figure 3A). The respiration peak was observed in the control fruits after 14 days of storage. On the other hand, fruit treated with GA 10% + CH 1% showed the same peak height after 21 days of storage and afterward the respiration rate decreased up to the end of experiment. GA 10% and CH 1% treated fruits exhibited a minor increase in respiration rate during the entire storage period. These results are in line with the findings of previous studies, where edible coatings reduced respiration rate in mango fruit (Kittur et al., 2001; Moalemiyan et al., 2012). It has been reported that respiration rate and ethylene production were suppressed by aloe vera gel coating in nectarine (Ahmad et al., 2009) and chitosan plus gum arabic in banana (Maqbool et al., 2011). In climacteric fruits, sudden increase in respiration and the burst of ethylene production occurs, after which it declines, which is the major cause of short shelf life. GA 10% and CH 1% treated fruits suppressed climacteric peak could be due to insufficient permeability and accumulation of carbon dioxide in the fruits.

Ethylene has a major role in ripening of climactic fruits, and biosynthesis of this hormone increase with the ripening process. Changes in ethylene production of mango fruit are shown in Figure 3B. The climacteric ethylene peak of control fruits was reached after 14 days of cold storage. Conversely, fruit treated with GA 10% + CH 1% showed the climacteric ethylene peak after 21 days of storage. Fruits treated with GA 10% and CH 1% showed a small rise in ethylene production during the whole storage period. Composite coating of gum arabic and chitosan reduced ethylene and respiration rate in banana (Maqbool et al., 2011). In the same way, alginate coating reduced ethylene production in plum fruits (Valero et al., 2013). Mango is a climacteric fruit and during ripening increase in ethylene production is a usual physiological process. However, gum arabic and chitosan coatings decreased ethylene production. This reduction could be by forming a semipermeable membrane around the fruits that modified the internal atmosphere, thus delayed the metabolic activity and possibly reduced the ethylene production.

Figure 3. Effect of gum arabic and chitosan on respiration rate (A) and ethylene production (B) of mango fruit during storage at 13°C for 28 days. Vertical bars indicate standard error of means for three replicates.

Conclusion

Results of this study demonstrated that application of gum arabic and chitosan coatings delayed the ripening process and reduced physico-chemical changes of mango fruit during cold storage. Both coating treatments decreased weight loss, respiration rate, ethylene production and changes in SSC. Chitosan plus gum arabic coatings efficiently inhibited decay incidence and maintained high firmness. The coating treatments had no significant effect on ascorbic acid, however maintained the overall quality of mango fruit. This study suggested that combine application of gum arabic and chitosan coatings can be used for reducing postharvest deterioration, extending the shelf life and maintaining quality of mango fruit during cold storage.
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References


