Enhancement of storage life and quality maintenance of papaya fruits using *Aloe vera* based antimicrobial coating

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Papaya is one of the main tropical fruits of India. The desiccation of fruits and perishable nature of papaya is a major drawback during transportation to distant markets and storage during glut in the market. *Aloe vera* gel, mainly composed of polysaccharides, has been recently explored as an edible coating owing to its antifungal activity. To improve the performance of edible coatings, various substances/chemical additives have been incorporated. Papaya leaf is a potential antifungal agent that could be used as a bio-based additive, especially, by papaya growing farmers. The present study was carried out to evaluate the ability of *Aloe* gel based antimicrobial coatings to reduce/control the loss of post harvest fruit quality in papaya and to compare the effects with a natural polysaccharide-chitosan, an established coating material with antifungal activity. Freshly harvested papaya fruits were coated with *Aloe* gel/AG (50%), papaya leaf extract/PLE incorporated *Aloe* gel (1:1) and 2.5% chitosan. The coated and uncoated (control) fruits were stored at 30±3°C and 42-55% RH for 15 d. Physical (PLW, fruit size), chemical (pH, titrable acidity and TSS), and sensory characteristics (colour, taste & firmness); fruit disease index (FDI), and marketability were analyzed at regular intervals during the storage period. The coated fruits survived the storage period of 15 d, whereas, all the uncoated controls decayed within 10 d. The uncoated/control fruits exhibited significantly greater changes in all the parameters tested. Among the coated fruits, PLEAG treated fruits exhibited least changes followed by AG and chitosan coated fruits. The coatings controlled the PLW, ripening process (chemical changes, colour development and softening of fruit tissue) and decay to a great extent and thereby extended the shelf life quality of the fruits. Marketability was also found to be better for PLEAG coated fruits among the 3 coatings, followed by AG and chitosan coated fruits. The effectiveness of AG coating was found to improve on incorporation of PLE. Shelf life could be further extended in low temperature storage. This is probably the first study on utilizing a natural alternative such as *Aloe*-gel and PLE to extend shelf life quality in papaya. On the basis of the overall physiological changes, *Aloe* gel based antimicrobial coating has been identified as a suitable method to extend the shelf life of papaya fruits.

Keywords: *Carica papaya*, edible coating, *Aloe* gel, chitosan, papaya leaf, post-harvest shelf life

Introduction

One of the major growth segments in the food retail industry is fresh and minimally processed fruits and vegetables. This new market trend has, thus, increased the demands for the food industry for seeking new strategies to increase storability and shelf life and to enhance microbial safety of fresh produce. The most important quality attributes contributing to the marketability of fresh and minimally processed produce include appearance, colour, texture, flavour, nutritional value, and microbiological safety. These quality attributes are determined by plant variety, stage of maturity or ripening, and the pre- and post-harvest conditions, and all can change rapidly during post-harvest storage. In general, desiccation and decay are the two major causes of the termination of commercial life span of fruits and vegetables. The water loss resulting from transpiration causes not only shrinkage and wilting, but also softening of produce. The main tropical fruits produced in India include banana, mango, guava, pineapple and papaya. India is the second amongst papaya producing countries and produces around 250 lakh quintals of papaya annually. The perishable nature of papaya is a major drawback for transport of the fruits to distant places and storage during glut in the market. The estimated post-harvest losses of papaya fruits in India has been reported to be between 40-100%. The different storage methods used to preserve papaya fruits include low temperature storage, controlled atmospheric storage, plastic film wraps and wax coatings. But there are many reports that papaya in refrigerated storage is susceptible to fungal decay. In controlled atmosphere storage, papaya fruits were benefitted only slightly from storage in low oxygen

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(1-1.5%) conditions, at 13°C. Gamma irradiation has also been used to increase the shelf life. However, higher doses caused excessive softening of the flesh, and low dose gamma irradiation though increased the shelf life but reduced the levels of ascorbic acid. Polymeric films and wax coatings were found to be effective in reducing water loss from papaya but were not effective in preventing fruit decay.

There are few studies on the efficacy of edible coatings to reduce the perishability of papayas. Application of various semi permeable edible coatings has been found to be a satisfactory way of achieving modified atmospheres. These coatings include a mixture of fatty acids and carboxymethylcellulose, and commercial coatings. Fruits coated with these edible coatings were reported to extend the shelf life and delay the onset of fungal infection. Chitosan, a modified natural biopolymer, with broad antimicrobial activity and film forming property has also been reported to be effective in extending the shelf life of papaya.

There is, hence, scope to study economically viable alternatives to improve the shelf life of papaya fruits using other edible coatings that are biodegradable and eco-friendly. Recently, there has been increased interest in using Aloe vera gel as an edible coating material for fruits and vegetables driven by its antifungal activity. A. vera gel based edible coatings have been shown to prevent loss of moisture and firmness, control respiratory rate and maturation development, delay oxidative browning, and reduce microorganism proliferation in fruits such as sweet cherry, table grapes and nectarines. In addition to the traditional role of edible coatings as a barrier to water loss and delaying fruit senescence, the new generation coatings are being designed for incorporating and/or for controlled release of antioxidants, nutraceuticals, chemical additives and natural antimicrobial agents. Extracts of papaya leaves have been reported to possess antimicrobial activity against fungal pathogens. There are no reports presently on the post-harvest application of Aloe gel coating on papaya fruits or the use of papaya leaf extracts as an antimicrobial agent in coatings. A study was, hence, conducted to evaluate the effect of Aloe gel and papaya leaf extract incorporated Aloe gel coatings on the storage life of papaya fruits in comparison to chitosan coating.

**Materials and Methods**

**Materials**

Freshly harvested papaya (Carica papaya L.) fruits were procured from the local market of Anantapur. They were selected on the basis of size, colour and absence of external injuries. Chitosan was purchased from Panvo Organics Pvt. Ltd., Chennai. Fresh leaves of Aloe vera and papaya were obtained from the University campus garden.

**Methods**

**Preparation of Coating Solutions**

Aloe vera gel matrix was separated from the outer cortex of leaves and this colourless hydroparenchyma was ground in a blender. The resulting mixture was filtered to remove the fibres. The liquid obtained constituted fresh Aloe vera gel. The gel matrix was pasteurized at 70°C for 45 min. For stabilizing, the gel was cooled immediately to an ambient temperature and ascorbic acid (1.9-2.0 g L⁻¹) was added; citric acid (4.5-4.6 g L⁻¹) was then added to maintain the pH at 4. The viscosity of the stabilized Aloe gel and its coating efficiency was improved by using 1% commercial gelling agent and was used as Aloe gel (AG) coating. To prepare papaya leaf extract incorporated Aloe gel coating (PLEAG), 100 g of leaves were surface sterilized for 5 min in 0.1% HgCl₂, washed thoroughly with sterile distilled water, crushed along with Aloe gel and its extract was filtered and made up to 100 mL with Aloe gel.

To prepare chitosan coating, chitosan was first purified by the method given by El Ghaouth. Purified chitosan was prepared by dissolving chitosan in 0.25N HCl and the undissolved particles were removed by centrifugation. The viscous solution was then neutralized with 2.5N NaOH (pH 9.8). Precipitated chitosan was collected, washed extensively with deionised water to remove the salts and subsequently dried. Purified chitosan was redissolved in 0.25N HCl and the pH was adjusted to 5.6 with 2N NaOH. The solution was autoclaved at 121°C for 15 min at 15 lb/inch² and used.

**Application of Edible Coating Solutions**

Before coating, papaya fruits were washed thoroughly and dried. The coating solutions used for papaya fruits were 50% Aloe gel, PLEAG (1:1), and 2.5% chitosan (the highest concentration reported in literature for coating application and antimicrobial activity). The fresh fruits were dipped completely into the coating solutions at room temperature for 15 min. They were allowed to drain and then dried at room temperature with forced air drying to allow a thin film layer to be formed on the fruits. Fifteen fruits were used for each coating solution. The fruits were then...
weighed and stored in bamboo baskets lined with newspapers at room temperature (30±3°C) and at 42-55% RH. The experiments were replicated. Fruits without coating were used as controls and were stored under same conditions as those for coated fruits. Various parameters were determined initially and at 5 d interval. The parameters analyzed included physiological loss in weight (PLW), fruit size, pH, titrable acidity, total soluble solids (TSS), sensory analysis for fruit quality (peel colour, firmness, appearance and taste), marketability and degree and rate of fruit spoilage.

**Physiological Loss in Weight (PLW)**

Water loss was calculated in terms of PLW by the following equation: 

\[
\frac{(A-B)}{A} \times 100,
\]

where, A is the initial weight of fruits (0 d) and B is the fruit weight after the storage period.

**Size of Fruits**

The mean size of the fruits was measured by using digital Vernier calipers. Fruits were measured vertically and across the fruit to the nearest 0.01 mm and the average value calculated.

**Titrable Acidity, pH and Total Soluble Solids**

Fruits were homogenized and the resultant pulp was filtered. The pH of the fruit juice was determined using a digital pH meter. Ten mL of squeezed fruit juice was diluted to 50 mL with distilled water and titrated against 0.1N NaOH by using phenolphthalein as indicator. The results were expressed as % total acids (titrable acidity). Total soluble solids were determined for both control and coated fruits by using Abbe’s hand refractometer.

**Sensory Analysis for Evaluating Fruit Quality**

Sensory analysis was carried out by 10 selected panelists. The fruits were randomly selected from each batch and served on white plates. The sensory quality of each batch of fruits was evaluated visually in terms of peel colour (5-bright green, 4-<25% yellow, 3- > 30% to <50% yellow, 2-50% to <75% yellow, 1- >75% yellow); fruit firmness (5-very firm, 4-firm, 3-moderately firm, 2-soft, 1-very soft); and fruit taste (5 point hedonic scale from 5-sweet to 1-bland) as an indicator of respiration rate and ripening. Marketability (5-Excellent, 4-Good with slight defects, 3-Fair and moderate defects, 2-Marketability limited, 1-Not marketable) of the fruits was also determined.

**Degree and Rate of Fruit Spoilage**

The differently coated fruits were visually observed for fungal spoilage and fruit rots. The number of fruits infected or spoiled was recorded periodically to assess the effect of the different coating on retarding fruit spoilage. It was reported as percentage disease index and calculated as follows:

\[
\text{% Disease index} = \frac{(oxa)+(1xb)+(2xc)+(3xd)+(4xe)}{a+b+c+d+e} \times 100
\]

Where - 0,1,2,3,4,5 are infection categories (0 – no lesions, 1 – 5 to ≤15%, 2 – ≥15% to ≤25%, 3 – ≥25% to ≤50%, 4 – ≥50% to ≤75%, 5 – >75% ); a,b,c,d,e are number of fruits that fall into the infection categories and X = maximum number of infection categories.

**Statistical Analysis**

The data obtained from the experiments were expressed as mean ±SE. Duncan’s multiple range tests with p<0.05 were employed to analyze the results and to compare control and coated fruit samples.

**Results**

The control fruits started deteriorating before 5 d and only few fruits survived up to 10 d, whereas, shelf life was extended to 15 d (study period) for majority of the coated fruits. Therefore, data for uncoated control was given only till 10 d storage period.

**Physical Characteristics**

Abnormal loss of firmness due to over ripening and pitting of the skin causes changes in the weight and size of fruits. The effect of selected edible coatings on PLW and fruit size was observed. PLW during storage was found to be significantly (P<0.05) different among the papaya fruits treated with the three different coatings and from control fruits at the end of 10 d storage (Fig. 1). PLW was observed to be...
values of the fruits on the initial day was 5.7 and were found to increase to 7.2±0.03 after 10 d of storage in control. In AG, PLEAG, and chitosan coated fruits there was minimal change in the pH values. Titrable acidity in the fruit samples decreased with storage time in both control and treated fruits. However, the difference was to a lesser extent in coated fruit compared to control. Titrable acidity values were found to be 0.12, 0.16, 0.18 and 0.15 for control, AG, PLEAG and chitosan coated fruits. After 15 d storage the pH values of coated fruits increased further with the lowest value of 5.82 in PLEAG coated fruits. The same treatment recorded least change in TA (0.12%) compared to 0.5% in chitosan coated fruits. The TSS was also lowest (8.2) in PLEAG fruits indicating slight ripening.

**Sensory Characteristics**

Colour, firmness and taste, the major sensory attributes, were scored by panel members (Table 2). Colour was evaluated based on the peel colour with ‘5’ as a score for green to a score of ‘1’ for complete yellowness. Bright green colour of papaya peel changed to yellow during storage in both control and coated fruits except in PLEAG coated fruits. The control fruits had shown a greater degree of yellowness by 5th d. Colour values were given as 1, 2.5, 5 and 2.5 for control, AG, PLEAG and chitosan coated fruits, respectively, after 10 d of storage. Firmness values were observed to be 1.0, 2.5, 4 and 2.2 for control, AG, PLEAG and chitosan coated fruits, respectively, stored up to 10 d. Taste values were found to be 5±0, 4.5±0, 2±0.35, and 4±0 for control, AG, PLEAG and chitosan coated fruits, respectively. The mean values of coated fruits for these parameters were significantly (P<0.05) different from control.

![Fig. 2—Effect of edible coatings on fruit size of papaya fruits (AG: Aloe gel coated fruits, PLEAG: Papaya leaf extract incorporated Aloe gel coated fruits).](image)

**Table 1—Effect of edible coatings on pH, titrable acidity and TSS of papaya fruits stored at 30±3°C**

<table>
<thead>
<tr>
<th>Storage Period (d)</th>
<th>Control pH</th>
<th>AG pH</th>
<th>PLEAG pH</th>
<th>Chitosan pH</th>
<th>Control TA (%)</th>
<th>AG TA (%)</th>
<th>PLEAG TA (%)</th>
<th>Chitosan TA (%)</th>
<th>Control TSS (°Brix)</th>
<th>AG TSS (°Brix)</th>
<th>PLEAG TSS (°Brix)</th>
<th>Chitosan TSS (°Brix)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>5.7±0</td>
<td>5.7±0</td>
<td>5.7±0</td>
<td>5.7±0</td>
<td>0.2±0</td>
<td>0.2±0</td>
<td>0.2±0</td>
<td>0.2±0</td>
<td>8.1±0.07</td>
<td>8.1±0.07</td>
<td>8.1±0.07</td>
<td>8.1±0.07</td>
</tr>
<tr>
<td>5</td>
<td>5.86±0</td>
<td>5.77±0</td>
<td>5.69±2</td>
<td>5.77±0</td>
<td>0.19±0.0</td>
<td>0.2±0</td>
<td>0.19±0.0</td>
<td>0.2±0</td>
<td>9.4±0.03</td>
<td>8.12±0.01</td>
<td>7.94±0.03</td>
<td>8.34±0.03</td>
</tr>
<tr>
<td>10 #</td>
<td>7.2±0.03a</td>
<td>5.85±0</td>
<td>5.72±0</td>
<td>5.91±0</td>
<td>0.12±0.06</td>
<td>0.16±0.06</td>
<td>0.18±0.08</td>
<td>0.15±0.04</td>
<td>11.1±0.04a</td>
<td>8.7±0.15b</td>
<td>8±0.02c</td>
<td>8.8±0.02b</td>
</tr>
<tr>
<td>15</td>
<td>*</td>
<td>5.96±0</td>
<td>5.82±0</td>
<td>6.13±0</td>
<td>*</td>
<td>0.07±0</td>
<td>0.12±0.0</td>
<td>0.05±0.02</td>
<td>9.1±0.04</td>
<td>8.2±0.02</td>
<td>9.2±0</td>
<td></td>
</tr>
</tbody>
</table>

*Indicates complete decay of fruits.

# Significant test carried out at the end of storage period of control fruits. Mean ± S.E. values in a row followed by different letters are significantly (p<0.05) different (DMRT).

(AG: Aloe gel coated fruits, PLEAG: Papaya leaf extract incorporated Aloe gel coated fruits)
Marketability

The commercial quality of control fruits decreased greatly by 5th day itself, whereas, coated fruits had maintained the quality (Figs 3 & 4). Marketability values were found to be 1.5, 3.5, 4 and 3.2 for control, AG, PLEAG and chitosan fruits, respectively, stored for 10 d. From the analysis it was observed that control fruits were having least marketability and PLEAG coated fruits the maximum. At the end of 15th day, marketability was found to be better for PLEAG coated fruits among the 3 coatings, followed by AG and chitosan coated fruits.

Fruit Disease Index (FDI)

FDI was used as a measure to indicate the effect of selected coatings on the microbial quality of fruits. Over ripening and decay were greater in non-coated than in coated fruits (Fig. 5). FDI was found to be 100, 40, 30 and 50 for control, AG, PLEAG and chitosan coated fruits, respectively, after 10 d storage period at 33±3°C. FDI was found to be similar (40) in AG and PLEAG coated fruits with a higher FDI of 60 in chitosan coated fruits after 15 d of storage.

Discussion

The results have proved the ability of the various bio-based coatings used in the present study to extent the shelf life of papaya fruits. From the analysis, it can be observed that PLEAG coating had shown the maximum effect in retarding the change in pH, titrable acidity and total soluble solids, and had controlled colour development and softening of fruit tissue even at the end of 15th day to a greater extent than AG and chitosan coatings. Maintenance of fruit firmness could be related to lower PLW, as reported earlier in sweet cherry treated with edible coating and Aloe gel coated nectarine fruits. The retardation
of physiological changes in the fruits indicate the ability of the selected coatings to modify the respiration rate and delay the ripening process, thereby extending the shelf life and maintaining the quality of the fruits. A delayed and smaller increase in TSS as seen in the present study has also been reported in Aloe vera gel coated sweet cherry\textsuperscript{11} and table grapes\textsuperscript{12}. This could be again attributed to the delayed ripening process. In climacteric fruits such as papaya, an increase in ethylene production during ripening is a normal physiological process. Earlier studies have shown a reduction in ethylene production in A. vera gel coated fruits\textsuperscript{13}. The retardation of the ripening process observed in the present study may be attributed to reduced ethylene production caused by the modified atmosphere. Surface coating has been reported to increase resistance of fruit skin to gas permeability, creating a modified internal atmosphere\textsuperscript{19} and reducing the respiration rate. Reduced respiration rate has been reported in A. vera gel coated sweet cherry\textsuperscript{11} and table grapes\textsuperscript{12}; in peach fruits coated with wax emulsions\textsuperscript{20}, and in avocado fruits coated with methyl cellulose\textsuperscript{21}. AG coated fruits had shown optimal colour development and softening of pulp followed by chitosan. Since, PLEAG had greatly delayed the physiological changes, it can be used for fruits that needs to be transported over longer distances and when longer shelf life period is necessary. The effect on the fruit physiology could be altered by changing the proportion of papaya leaf extract to Aloe gel. In the present study equal proportion of PLE and AG was used.

In the present study, Aloe based coatings showed the least disease index. A. vera gel based coatings have been reported earlier to reduce microorganism proliferation in sweet cherries and table grapes\textsuperscript{11}. Earlier in vivo studies in our laboratory had shown even unstabilised aqueous extracts of A. vera gel to have antifungal activity against Colletotrichum gloeosporioides, a major post-harvest pathogen of papaya fruit\textsuperscript{15}. Antimicrobial activity of A. vera inner gel against Gram-positive and Gram-negative bacteria has been also demonstrated by Habeeb et al\textsuperscript{22}. Coating of fruits with PLEAG resulted in better control of fruit decay compared to AG and chitosan coating during storage. In the present study, incorporation of PLE into AG had only marginally improved the antimicrobial activity of AG coating. The effectiveness could be improved by incorporating solvent extracts of papaya leaf than the aqueous extracts, as the former are reported to have better antimicrobial activity than aqueous extracts\textsuperscript{15,23}. Preservation of the coated papaya fruits at their ambient storage temperature (10-13°C) is likely to further extend the shelf life of this tropical fruit. In the present study, shelf life extension could be observed even at a higher storage temperature of 30°C. Studies are in progress to evaluate the efficacy of Aloe and chitosan composite coatings on different papaya varieties for a longer period (>15d).

The present investigation is probably the first to report the efficacy of using A. vera gel based antimicrobial coating in tropical fruits such as papaya with promising results. The utilization of papaya leaf extracts as an antifungal natural additive in coatings has been also presented here for the first time. These are bio-based substances and are thereby eco-friendly. Being available easily in the tropical regions, they could be used as a cost-effective simple method for extending the post harvest shelf life and quality of fruits and vegetables.

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References


