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Original Research Article

A New Approach of Hurdle technology to preserve Mango fruit with the application of *Aloe vera* gel and Calcium chloride

Shweta Chauhan¹*, K.C.Gupta¹ and Mukesh Agrawal²

¹Department of Microbiology, Vijya Raje Government Girls Post Graduate College, Morar, Gwalior (M.P), India ²Defense Research Development and Establishment, Jhansi Road Gwalior (M.P), India **Corresponding author*

ABSTRACT

Keywords Aloe vera gel, calcium chloride, mango, hurdle technology The application of preservatives is necessary to boost the global economy which is essential for the prevention of the post-production and transportation losses of fruit world widely. The use of plant products, as natural preservatives, is increasing interest because it is non toxic to the human health. The present study aim to investigate the efficacy of natural preservative (Aloe vera gel coating) and chemical preservative (calcium chloride spray) separately and in the combination (hurdle technology) as an effective preservative for the increase of the shelf life period of mango (Mangifera indica) during storage. Fruits were harvested from mango-farm of Gwalior (M.P.), India, at the matured stage. Treated and controls fruits were stored at 15±1 °C and 85% RH with Aloe vera gel and calcium chloride separately and 60 days shelf life was recorded. But 90 days shelf life period was noticed when hurdle technology was used. Fruit firmness, weight loss, skin color, microbial counts, total soluble solids (TSS), and total titratable acidity (TTA) were evaluated. Significant inhibition of the total microbial count was observed when the 1%,5% and 10% Aloe vera gel were applied ; however Aloe vera gel was analysed to be inhibitory in action when applied in combination of calcium chloride. But in all the controls, the decaying process was started after 7 day in similar conditions.

Introduction

In India, mango is known as the king of fruits and has unique taste, prominent flavor, strong aroma, and contain high amount of vitamin C, beta-caroteniods and trace amount of minerals (Shweta et al, 2014). Mango (*Mangifera indica* L.), as an emerging tropical export crop is produced in about 90 countries in the world with a production of over 25.1 million tones. Asia is the main producer with 76.9% of the total world production, followed by

America with 13.38%, Africa with 9% and less than 1% each for Europe and Oceania (Sauco, 2002). Ripen mango fruits are highly perishable with short shelf life. After harvesting, the ripening process in mature green mango takes 9-12 days (Herianus *et al.*, 2003). Depending on storage conditions shelf life of mango varies, ranges from 4 to 8 days at room temperature and 2 to 3 weeks in cold storage at 13° C (Carrillo *et al.*, 2000). Short shelf life of mango restricts the long distance commercial transport (Gomer-Lim, 1997). Substantial quantities of mangoes are wasted because of poor postlack harvest management and of appropriate facilities in developing countries. Development and application of inexpensive preservation techniques to produce high quality and acceptable products of mango could be valuable, allowing a better use of the fruit (Ulloa et al., 2008) . Application of modified (MA) atmosphere controlled or atmosphere (CA) has extended the shelf life of mango (Bender et al., 2000; Noomhorm & Tiasuwan, 1995). MA storage reported slow ripening, but accompanied by high CO2 and off mango flavor (Gonzalez-Aguilar et al., 1997). Therefore there is a need of effective preservative technique.

Being the cheapest among several methods of preservation, use of *Aloe vera* gel coating and calcium chloride spray preservation is the cheapest method and can be used to prevent the food spoilage due to microbial attack and effectively applied in combinations for better preservation. No single preservative is completely effective against all microorganisms (Chipley, 1983).

Aloe vera is a tropical and subtropical plant that is in application since ancient time for medicinal and therapeutic properties. Aloe vera belongs to liliaceae family and is a very short-stemmed succulent plant. Aloe vera gel is significantly used in the food industry. It is a resource of functional foods in drinks, beverages and ice creams (Shweta et al., 2014).

Calcium chloride has been widely used as preservative and firming agent in the fruits

and vegetables industry for whole and fresh-cut commodities (Chardonnet, Charron, Sams, and Conway, 2003).

This study focused on the development of methods of bio preservation to evaluate their efficacy in extending the shelf life and improving the microbial safety of mango fruit products. The present study has recorded the significant and the effective natural and chemical antimicrobial agents to increase the shelf life period of the mango.

Materials and Methods

Sample Collection

Fully ripened, sound and healthy mangoes (Chaunsa variety) were purchased from local fruit market in Gwalior, India. The fruits were thoroughly washed with potable water for 5 minute remove dirt, dust, pesticide residues and surface microbial load. All the samples were individually stored in washed and sanitized air tight plastic container. Each treatment consists of five (5) mango sample with one (1) replicate per treatment.

Treatment with calcium chloride and Aloe vera gel

Calcium chloride was purchased from market (Gwalior). The mango fruits were treated with 1.0%, 3.0%, 5.0% and 7.0 % calcium chloride (CaCl2.2H2O), solutions by spraying for 10 seconds, control (0.0%) in which fruits were sprayed with distilled water for 10 seconds. Aloe vera was collected from botanical garden of V.R.G college Gwalior, India and fresh gel was used for application. Scheme of study

In this study, the samples of mango fruit

were immersed (for 10 seconds) in respective solutions in following manner:

 $\begin{array}{l} M1 = 0.0 \ \% \ Aloe \ vera \ gel \\ M2 = 1.0\% \ Aloe \ vera \ gel \\ M3 = 5.0\% \ Aloe \ vera \ gel \\ M4 = 10\% \ Aloe \ vera \ gel \\ M5 = 1.0\% \ calcium \ chloride \\ M6 = 3.0\% \ calcium \ chloride \\ M7 = 5.0\% \ calcium \ chloride \\ M8 = 7.0\% \ calcium \ chloride \\ M9 = \ 5.0\% \ calcium \ chloride \ and \\ 5.0\% \ Aloe \ vera \ gel \end{array}$

M 10 = control

Parameters evaluated Physical characteristics

Weight loss (%):

Samples (5) were weighed at the start of experiment and at the end of each storage interval. The difference between initial and final fruit weight was considered as total weight loss during that storage interval. The calculations were made in percentages on fresh weight basis. Fruit weight was recorded on weekly interval by using digital balance.

Sensory Evaluation

The samples with edible coating were evaluated for its acceptability, during the process of optimization and storage studies. For sensory evaluation paneer samples were served to a panel of five panelists consisting of faculty from the department of Microbiology in the VRG College. The panelists were asked to evaluate the sensory quality of paneer samples as per sensory score card. Panel members were directed to judge each samples on the basis appearance, flavor, body and texture and overall acceptability, and indicate their degree of liking on a 9point Hedonic Scale (Lawless and Hayman, 1998).

Organoleptic evaluation

The visual characteristics of mango appearance for skin colour, aroma, pulp colour, flavour and taste were scored in daylight, by a panel of 5 trained judges (Xu et al. 2007).

Table -1

Quality	Methods of evaluation				
Parameter	and units				
Skin colour	Visual index for mango:				
(shown in	1 = excellent, $2 = $ good, $3 =$				
table -3)	slightly dull, $4 = <50\%$				
	brownish, $5 = >50\%$				
	brownish.				
	Visual index for mango:				
Pulp color	1 = 100% good (yellow), 2				
	= 75 % good (pale yellow),				
	3 = 50 % good (light				
	brown), $4 = 25 \%$ good				
	(brown), $5 = poor quality$				
	(dark brown and black)				
Flavor	Flavor acceptability using a				
(Aroma),	5-point scale:				
shown in table	1 = excellent, $2 = $ good, 3				
-3	= acceptable, 4 $=$ poor, 5 $=$				
	unacceptable.				

Microbial analysis

Daily observations were taken and microbial counts were estimated by the microbial limit test (MLT) along with pathogen estimation. For this purpose 90 mm sterile petri plates were required. Four-petri plate were required and labeled as two plates for Bacteria and remaining two for fungi count. Transferred 1 ml quantity of each pretreated dilution sample solution to each of four petri plates. Added 15 ml of sterile liquefied SCDA (soyabean casein digest agar) at not more than 450C, in to two plates labeled for bacterial count. Then added 15 ml of sterile

liquefied SCA at not more than 450C, in to two plates labeled for fungal count. Allowed to solidify the plates at room temperature, inverted and incubated at 30 to 350C for 5 days and 20 to 250C for 5 days respectively. Counted the number of colonies that were formed. Calculated the number of cfu per gm or per ml of the sample being examined.

The number of colonies should be in limit as:

For Bacterial Count = NMT 1000 cfu per ml

For Fungal Count = NMT 100 cfu per ml

Disease Severity Assessment (Shweta et al., 2014)

Disease severity assessment was performed according to the following empirical scale:

0 = healthy Mango;

- 1 =one very small lesion (beginning of infection);
- 2 =one lesion,
- 3 =several lesions or 25% of the mango infected;
- 4 =50% of the mango surface infected, sporulation present;
- 5 = more than 50% of the mango surface infected, sporulation present,
- 6 = 100% of the mango surface infected, sporulation present

Chemical Parameter

pH and titratable acidity (TTA)

Whole fruit was passed through an electric juicer (Inalsa, India) and filtered through cheese cloth. pH was measured by digital pH meter (WTW 526, Germany). For the free titratable acidity, 1 gm of peel powder was boiled for 10 minutes in 20 ml of distilled water and filtered through a Buchner funnel. The free titratable acidity was measured according to AOAC guidelines (Method 942.15.b, 2000).

Total phenolic content (TPC) and Total suspended solids (TSS)

Firstly mango was cut in halves and squeezed using an automatic juicer (Inalsa). The TSS of mango samples was determined according to AOAC method (Anon., 1990) by using hand refractometer. The total phenolic content (TPC) was assessed using Folin-Ciocalteu assay (Singleton et al., 1999). Volumes of 0.5 mL of distilled water and 0.125 mL of sample were added to a test tube. A volume of 0.125 mL of 2.0 N Folin-Ciocalteu reagents was added and allowed to react for 6 min. Then, 1.25 mL of a 7% sodium carbonate solution (v/v) was added to the mixture and allowed to stand for 90 min in the dark, for colour development. Before reading the absorbance at 760 nm in a spectrophotometer, the mixture was diluted up to 3 mL with distilled water. Gallic acid solutions were used for the standard calibration curve and the total phenolic content was expressed as mg gallic acid equivalents (GAE)/g or 100 g peels (dry weight or fresh weight basis, DW or FW). All measurements were carried out in triplicate.

Results and Discussion

Physical parameters

The author has studied Aloe vera gel for the first time in India as an edible coating over mango fruit and recorded the 60 days of storage period with calcium chloride and Aloe vera gel coating separately and 90 days of storage period at 15 ± 1 °C and 85% RH, shown in graph 1. During such storage applied physical, chemical and assessment methods microbial were evaluated. Controlled fruits showed a rapid deterioration with an estimated shelf life period of 7 days at 15°C with higher weight loss, color changes, accelerated softening and ripening, browning, and

high incidence decay. On the contrary, the treated samples significantly delayed these parameters significantly after post harvesting. This may be due to the semipermeability created by coatings on the surface of the fruit, which might have modified the internal atmosphere i.e. O_2 and CO_2 concentrations in the fruit and retards ripening (Lowings and Cutts, 1982; Bai et al., 1988).



*CC = calcium chloride, AG = *Aloe vera* gel, CCAG = combine effects of calcium chloride and

S.No	Treatment		Skin colour	Visual index	Pulp	Flavour
					color	(Aroma)
1	Cacl ₂	M1	5	4	5	5
		M2	1	2	1	2
		M3	1	1	1	2
		M4	1	1	1	2
2	Aloe vera	M5	5	4	5	5
		M6	1	1	1	1
		M7	1	1	1	1
		M8	1	1	1	1
		M9	1	1	1	1
3	Control	M10	5	4	5	5

Table -3 Sensory Evaluation (Organoleptic evaluation)

Table.4 Effect of different treatments on the turbidity of mango juice

Concentration(g/l)	***CCAG	**AG	*CC
1.00	0.0103	0.226	0.284
0.70	0.0477	0.2873	0.3684
0.50	0.0846	0.3147	0.3573
0.10	0.0953	0.2163	0.298
Untreated	0.526	0.528	0.532

Spectrophotometric results showed maximum turbidity with untreated controlled samples, where as all the treated mango juice samples were observed with minimum turbidity.

Graph.2A Microbial limit test



Microbial analysis showed higher microbial counts with controlled sample where as all the treated samples showed lesser microbial counts with in limit of 1000 cfu per ml. M9 samples were observed with minimum microbial counts.

Graph 2B

CCAG samples were observed with healthy with no lesion and fungal spore and were scaled with highest 1 point, whereas controlled samples were observed with full of fungus sporulation and lesions, pointed with poor 6 scale pointer value.



Graph -3A

There was a decreasing scale in firmness in both coated and controlled fruits during the course of storage. However, the control exhibited higher loss of firmness than the experimental. This was due to the effects of *Aloe vera* gel coating which delayed the softening. The similar results were also obtained by Hagenmaier and Baker (1995).







However, the controlled fruits, exhibited higher loss in firmness, graph 1 showed higher percentage of firmness and weight loss (100%) in controlled samples. The authors results are in agreement with the observation of Hagenmaeir and Baker (1995) who found that coating reduces of fruits. The retention of shrinkages firmness in coated fruits was due to reduction in degradation of insoluble proto-pectins to more soluble pectic acid and pectin. It was found that during fruit ripening, depolymerization or shortening of chain length of pectin substances occurs with an increase in pectin-esterase and polygalacturonase activities (Yaman and Bayoindirli, 2002). Hence low oxygen and high CO₂ concentrations reduce the activities of these enzymes and allows retention of the firmness during storage (Salunkhe et al., 1991, Yaman and Bayoindirli, 2002; Patricia et al., 2005).

Where as treated sample showed lesser loss, this was due to the effects of *Aloe vera* gel coating which delayed the softening. The similar results were also obtained by Hagenmaeir and Baker (1995). The retention of firmness in coated fruits was due to the reduction in degradation of insoluble proto-pectins, to more soluble pectic acid and pectin. It was found that during post harvest fruit depolymerization or length ripening. shortening of pectin chain occurred with an increase in pectin-esterase and polygalacturonase activities (Yaman and Bayoindirli, 2002). Hence low oxygen and high CO₂ concentrations reduced the activities of these enzymes and allows retention of the firmness during storage (Salunkhe et al., 1991, Yaman and Bayoindirli, 2002; Patricia et al., 2005). Combine effects of calcium chloride and Aloe vera gel showed better results.

Sensory Evaluation (Organoleptic evaluation)

The sensory analyses revealed beneficial effects in terms of delaying mango fruit skin browning and dehydration and maintenance of the visual aspect of the fruit without any detrimental effect on taste, aroma, or flavors. Combine effects of CCAG showed best scale point of 1 for all the parameters, where as controlled sample showed poor scale point. Aloe vera gel treated mango showed best score for aroma / flavour and taste , while untreated or control samples showed rancid smell and poor taste , due to the biochemical changes in carbohydrates, proteins , amino

acids, lipids and phenolic compounds that are active component of natural additives, can influence the pleasant aroma, flavour and taste. additives, can influence the pleasant aroma, flavour and taste (Nadeem et al., 2009). Edible coatings used to protect perishable food products from deterioration by retarding dehydration, suppressing respiration, improving textural quality helping retain volatile flavor compounds, and reducing microbial growth.

Microbial Analysis

Controlled fruits showed higher fungal growth than the treated fruits. It has been reported that the antifungal activity of *A*. *vera* is based on the suppression of germination and inhibition of mycelial growth (Ali et al., 1999).

Chemical parameter

The maximum rate for TSS was observed with coated mango and apple. But such significant values of TSS were not recorded in coated grapes. Results showed the effects of Aloe vera coatings on TTA during storage. TTA is directly related to the concentration of organic acids present in the fruits. The decreasing acidity at the end of storage is due to the metabolic changes in fruits resulting from the use of organic acids in respiratory process; this observation was in agreement with the findings of Echeverria and Valich (1989).

In Conclusion, Mango samples were applied separately with chitosan, calcium chloride and Aloe vera gel. The good scale with higher TSS and TPC was recorded for application. Better results were noticed when mango samples were applied with hurdle technology in combination of chitosan (microbial product) and calcium chloride (chemical preservative). Hence this study aimed to investigate the effect of an edible coating over the mango but other factors are also responsible for extending the shelf life such as careful harvesting, handling without injuring the product, and hygienic storage conditions.

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