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Effect of chitosan coating on postharvest diseases and fruit quality of mango *(Mangifera indica)*

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ABSTRACT

The objective of this work was to study the effect of postharvest chitosan coating on fruit quality and shelf life of mango (*Mangifera indica* L.) during storage at ambient condition. Physiologically mature freshly harvested mango fruits (cv. Langra) were treated with chitosan solutions (0%, 0.5%, 0.75% or 1.0%) containing 0.5% acetic acid for 1 min and stored at ambient condition (25 ± 2°C, 85 ± 5% RH). Treatment with 1.0% and 0.75% chitosan significantly reduced weight loss and disease incidence of fruit compared to control. These treatments also maintained higher ascorbic acid, total phenolics content and total antioxidant activity than other treatments.However, development of peel colour and total carotenoids content in the fruit pulp was suppressed by chitosan 1.0% treated fruits. Treatment of mango with chitosan also delayed increase in the total soluble solids and decrease in titratable acidity compared to control. No significant differences were recorded between chitosan 0.75% and 1.0% treated fruits except for peel colour development and total carotenoids content. These results indicated that 0.75% chitosan coating could preserve fruit quality, reduce disease incidence and extend shelf life of mango up to 12 days during storage at ambient condition.

Key words: Chitosan, Coating, Decay, Mango, Postharvest

Mango (*Mangifera indica* L.) is one of the most important tropical fruit of the world, known for its attractive colour, exotic flavour, delicious taste and rich nutritional properties. Owing to climacteric in mature mangoes ripen rapidly after harvest, showing respiration and ethylene evolution peaks on 3rd or 4th day of harvest at ambient temperature (Narayana *et al*. 1996). The fruit is highly perishable in nature having shelf life of only 4 – 8 days at ambient condition (Carrillo *et al*. 2000). Such a short postharvest life seriously limits the long distance commercial transport of this fruit (Gomer-Lim 1997). Moreover, mango fruits are also highly sensitive to postharvest decay which causes massive loss during transit or storage. The major postharvest diseases occur in mango are anthracnose (caused by *Colletotrichum gloeosporioides*) and stem-end rot (caused by *Lasiodiplodia theobromae*) which remain as latent infection at the time of harvest. Susceptibility of mango to the attack by these pathogens increases during storage owing to a series of ripening associated changes and environmental condition favouring establishment and colonisation of these pathogens (Eckert *et al*. 1996). Low temperature storage is a common postharvest technique to delay ripening, senescence and minimize spoilage. But, the problem with mango is its high chilling sensitivity while stored at low temperature (Barman and Asrey 2014). Generally, control of these postharvest diseases is achieved by combination of thermal and fungicide prochloraz treatment. However, owing to adverse effects of synthetic fungicides on human health and environment (Zeng *et al*. 2006) and also reports for development of pathogen resistance over synthetic fungicides (Cavelier *et al*. 1994) make a strong desire to explore new alternatives to reduce postharvest diseases and extend its shelf = life.

Chitosan is a high molecular weight cationic polysaccharide derived from deacetylation of chitin obtained from exoskeleton of crustacean shells such as shrimps, crabs and krills (Sandford and Hutchings 1987, Sandford 1989). Due to its excellent film-forming and barrier properties, biodegradable nature and antimicrobial property, chitosan has attracted attention as a potential food preservative in improving storability of several fruits (Arvanitoyannis *et al*. 1998, El-Ghaouth *et al*. 1991, Zhang and Quantick 1997, Jiang and Li 2001, Li and Yu 2001). United States Food and Drug Administration (USFDA) have also approved application of chitosan as a potential food additive (Hirano

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et al. 1990). In the present study, effect of chitosan on postharvest life, disease incidence, quality attributes and health promoting bioactive compounds of mango was investigated during storage at ambient condition.

MATERIALS AND METHODS

Mango fruits (cv. Langra) were harvested at physiological maturity stage from the orchard of Horticulture Garden, Department of Horticulture (Fruit and Fruit Technology), Bihar Agricultural University, Sabour, Bhagalpur, Bihar (India) and immediately transported to the laboratory. Following that, fruits of uniform size, shape, colour and free from disease and pest were selected for the experiment. A total 200 fruits were selected for the experiment which was randomly divided into 4 lots containing 50 fruits in each lot for different treatments. The concentrations of chitosan solutions for the experiment were selected on the basis of initial screening trials. The chitosan solutions of 0.5%, 0.75% and 1.0% were prepared by dissolving chitosan in distilled water containing 0.5% acetic acid. The pH of the solution was then adjusted to 5.6 with 1 N sodium hydroxide. Fruits were then treated with chitosan by dipping in chitosan solutions for 1 min. The control mango fruits were treated with acidic solution (pH 5.6) without chitosan. Fruits were then air-dried and stored at ambient condition (25 ± 2°C, 85 ± 5% RH) for 12 days.At 4 days interval, fruits from each treatment were sampled at random and subjected to analysis for different physico-chemical attributes.

Weight loss of fruit during postharvest storage was determined by calculating the difference between the initial fruit weight and the weight at the time of recording data and expressed as percentage (%).Disease incidence was expressed as percentage (%) on the basis of fruit showing symptom of rots irrespective of severity out of total number of the fruits in each treatment (Sivakumar *et al*. 2002). Peel colour of mango fruit during storage was determined visually following a score ranging from 1 – 5 where, 1= 100% green, 2= 1 – 25% yellow, 3= 26 – 50% yellow, 4= 51 – 75% yellow and 5= 76 – 100% yellow. It was calculated by multiplying the number of fruits in each category by the respective score, summing the products and dividing by the total number of fruits.

Total carotenoids content in the fruit pulp was estimated by extracting the carotenoid pigments from the fruit pulp with a mixture of petroleum ether and acetone which was then estimated in a spectrophotometer at 452 nm and the results were expressed as mg 100/g (Roy 1973). Ascorbic acid content in mango during storage was estimated using the 2,6-dichlorophenol indophenol dye method (Jones and Hughes 1983). Fruit pulp of 10 g was homogenized with 3.0% metaphosphoric acid solution and volume was adjusted up to 100 ml. The mixture was then filtered and 10 ml filtrate was titrated against the dye solution to a pink colour that persisted for 15 s. The ascorbic acid content in fruit was expressed as mg 100/g. To determine total phenolics content, 0.1 ml of mango pulp extract in ethanol (80%) was added to distilled water (2.9 ml), foilin-ciocalteau phenol reagent (0.5 ml) and 20% sodium carbonate solution (2.0 ml). The absorbance of the mixture was then determined in a spectrophotometer at 760 nm and results were expressed as gallic acid equivalent (µg GAE/g) (Singleton *et al*. 1999). Total antioxidant activity in the edible portion of mango was estimated following CUPRAC (cupric ion reducing antioxidant capacity) method of Apak *et al*. (2004). The results were expressed as trolox equivalent (µmol TE/g).

Total soluble solids (TSS) content in fruit during postharvest storage was determined using refractometer, which was calibrated with distilled water prior to taking observations and results were expressed as °Brix. Titratable acidity (TA) was estimated by titration method (AOAC 2000). For this, 2 g of fruit sample was titrated against 0.1 N NaOH using phenolphthalein as indicator to a pink end point (pH 8.1). The results were expressed as the citric acid (%).

The experiment was conducted in a Completely Randomized Design (CRD) with four treatments and three replications.Data recorded from each treatment under different parameters were subjected to analysis of variance (ANOVA) using treatment and storage time as sources of variation. Duncan's multiple range test was used to compare means among different treatments at a significance level of $P \leq 0.05$. All analyses were performed using SAS 9.2 (SAS Institute, Cary, NC, USA).

RESULTS AND DISCUSSION

Effect of chitosan on weight loss

In this study, chitosan coating significantly reduced weight loss in mango fruit compared to control (Fig 1). Irrespective of treatments, a progressive increase in weight loss was recorded with the advancement of storage period. After 12 days of storage, maximum weight loss of 20.97% was recorded in control fruits. Among the treatments, the

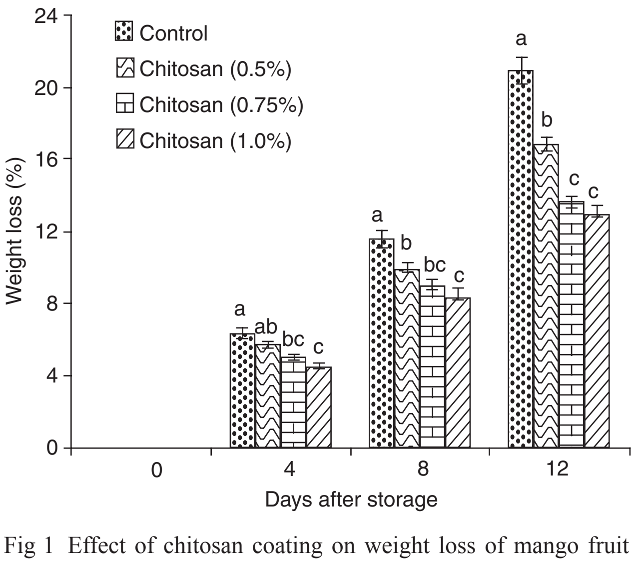

Fig 1 Effect of chitosan coating on weight loss of mango fruit during storage

minimum weight loss of 12.9% was recorded in fruits treated with 1.0% chitosan which was statistically at par with 0.75% chitosan treated fruits (13.68%). Loss in weight in mango fruit during postharvest storage was mainly due to loss of water from the fruit by transpiration and respiration process (Barman *et al*. 2011). Application of chitosan formed a protective coating over the fruit surface thereby reduced loss of moisture from the fruit. The higher concentration of chitosan was found more effective in reducing weight loss due to formation of thick physical barrier than lower concentration of chitosan. The finding of this study is in agreement with the previous findings in papaya (Ali *et al*. 2011), guava (Hong *et al*. 2012), longan (Jiang and Li 2001), strawberry (Hernández-Muñoz *et al*. 2008) and litchi (Kumari *et al*. 2015).

Effect of chitosan on disease incidence

Chitosan treatment was found highly effective against disease incidence in mango during postharvest storage (Fig 2). After 4 days of storage, disease incidence of 1.33% was observed in control fruits while all the chitosan treatments completely suppressed the onset of diseases in mango. After 8 days of storage, only 0.5% chitosan treated fruits showed symptoms of diseases (7.0%) in addition to control (12.33%). However at the end of the experiment, minimum disease incidence of 12.0% was noted in 1.0% chitosan treated fruits while, it was highest in control (43.0%).The inhibitory effect of chitosan coating on disease incidence of mango fruit was mainly attributed to antifungal property of chitosan (Bautista-Baños *et al*. 2006). It increased the activities of defence enzymes like ß-1,3-glucanase, chitinaseand synthesis of phytoalexins, thereby elicited host defence responses(Zhang and Quantick 1998). Moreover, onset of diseases (anthracnose and stem-end rot) in mango during storage from pre-harvest latent infection is usually associated with ripening of fruit (Barkai-Golan 2001; Sharma and Sharma 2016). Chitosan treatment of mango delayed ripening of fruit thereby indirectly suppressed onset of disease incidence. Apart from that, reduced O_2 level and elevated CO_2 level inside the fruit due to chitosan coating might also have direct effect on reduced disease incidence in mango (Jitareerat *et al*. 2010).

Effect of chitosan on peel colour and total carotenoids content

The development of peel colour and total carotenoids content in the fruit pulp were significantly affected by the chitosan treatments. Up to 4 days of storage, no significant changes in mango peel colour was recorded among the treatments. After that, a rapid development of yellow colour on mango peel was noted in all the fruits under treatment. The maximum peel colour (2.2 score) development after 8 days of storage was observed in control while, it was minimum (1.26 score) in 1.0% chitosan treated fruits. After 12 days of storage, minimum change in peel colour was observed in 1.0% chitosan (1.43%) followed by 0.75% chitosan (2.0%) treated fruits. Likewise, total carotenoids content in the fruit pulp increased progressively with the increase in storage period in all the treated and control fruits (Fig 3). Rapid increase in carotenoids content was observed in control and 0.5% chitosan treated fruits while, 0.75% and 1.0% chitosan treatments delayed development of carotenoid pigments in the pulp during storage. Highest carotenoids content (7.51 mg 100/g) was recorded in control fruits while it was lowest (4.98 mg 100/g) in 1.0% chitosan treated fruits. Application of chitosan in mango formed a semipermeable film over the fruit surface which modified the internal atmosphere (O_2, CO_2 and ethylene) of the fruit (Banks 1984, El-Ghaouth *et al*. 1992). Chitosan coating provides selective permeability to O_2 than CO_2 thereby does not cause anaerobic respiration (Bai *et al*. 1988). Therefore, delayed peel colour development and increase in carotenoid pigments in the fruit pulp of chitosan treated fruits might be attributed to delay in fruit ripening process owing to

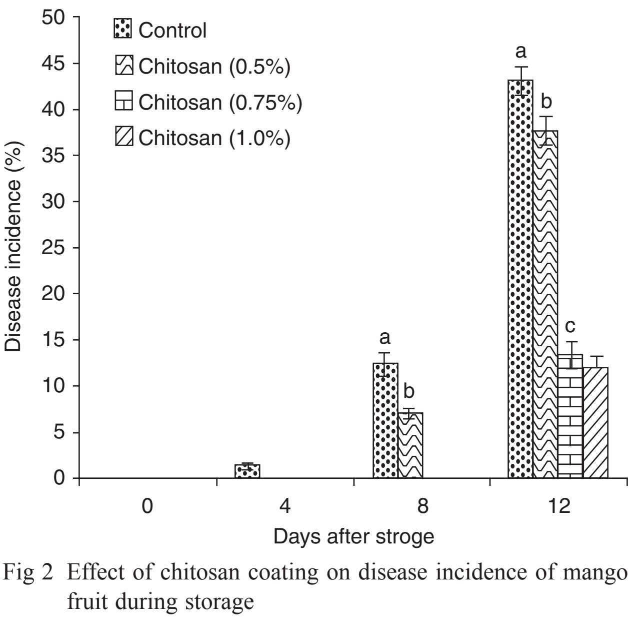

Fig 2 Effect of chitosan coating on disease incidence of mango fruit during storage

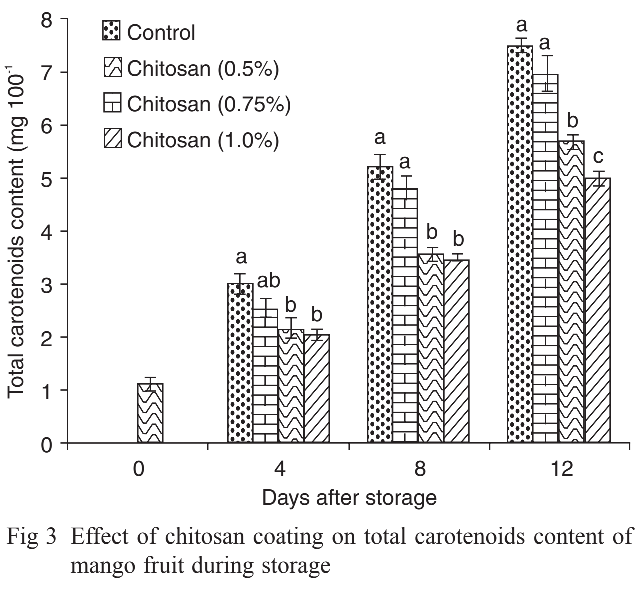

Fig 3 Effect of chitosan coating on total carotenoids content of mango fruit during storage

Table 1 Effect of chitosan coating on fruit quality attributes of mango during storage.

Parameters	Days after storage	Control	Chitosan (0.5%)	Chitosan (0.75%)	Chitosan (1.0%)
Ascorbic acid	4	30.45 ± 0.419 a	30.77 ± 0.403 a	31.94 ± 0.422 a	31.64 ± 0.836 a
	8	25.51 ± 0.690 a	27.07 ± 0.825 ab	29.35 ± 0.732 b	29.42 ± 0.694 b
	12	19.31 ± 0.930 a	21.47 ± 0.811 ab	23.88 ± 0.556 bc	24.87 ± 0.962 c
Total soluble solids	4	17.36 ± 0.726 a	15.70 ± 0.404 b	14.60 ± 0.360 bc	13.70 ± 0.288 c
	8	23.66 ± 0.409 a	21.30 ± 0.321 b	19.00 ± 0.208 c	18.80 ± 0.200 c
	12	24.66 ± 0.392 a	24.16 ± 0.317 a	22.43 ± 0.375 b	22.00 ± 0.378 b
Titratable acidity	4	0.57 ± 0.048 a	0.62 ± 0.014 ab	0.73 ± 0.074 ab	0.76 ± 0.054 b
	8	0.36 ± 0.047 a	0.44 ± 0.026 ab	0.58 ± 0.063 bc	0.67 ± 0.047 c
	12	0.25 ± 0.051 a	0.26 ± 0.054 ab	0.42 ± 0.054 bc	0.47 ± 0.028 c

Data are means ± standard error of three replicate determinations (n=3). Mean values in each row followed by the same letter are not significantly different ($P \leq 0.05$).

reduced rate of respiration and ethylene production. Previous workers have also reported that chitosan application delay fruit ripening by reduced rate of respiration and ethylene evolution (Martínez-Romero *et al*. 2006, Hong *et al*. 2012, Hewajulige *et al*. 2009).

Effect of chitosan on ascorbic acid content

Ascorbic acid content in the fruit pulp of mango declined during storage irrespective of treatments (Table 1). No significant difference in ascorbic acid content was recorded among the treatments after 4 days of storage. After 12 days, highest retention of ascorbic acid (24.87 mg/100 g) was recorded in chitosan 1.0% treated fruits while, it was lowest (19.31 mg/100 g) in control fruits. This result showed that chitosan coating slowed down the loss of ascorbic acid in mango during storage. It might be due to the fact that chitosan treatment reduced O_2 permeability in fruit and maintained higher concentration of CO_2 by providing physical barrier surrounding the fruit which protected ascorbic acid from deteriorative oxidation reaction (Ayranci and Tunc 2004). Our results are in line with those of Mathooko (2003), Ayranci and Tunc (2004) and Hong *et al*. (2012), where a slower loss of ascorbic acid was reported in tomato and guava due to higher CO_2 and lower O_2 levels around fruit after chitosan treatment.

Effect of chitosan on total phenolics content and total antioxidant activity

Chitosan treatment of mango also influenced total phenolics content and total antioxidant activity in the fruit during postharvest storage. Control fruits exhibited rapid decline in phenolics content with the advancement of storage period however, chitosan treatments delayed decrease in phenolics in the fruit pulp. After 12 days of storage, lowest total phenolics content was recorded in control (148.26 µg GAE/g) which did not differ significantly with 0.5% chitosan treated fruits (162.44 µg GAE/g). However, fruits treated with 1.0% chitosan retained maximum content of total phenolics (239.12 µg GAE/g) up to end of the experiment. The total antioxidant activity did not differ significantly up to 4 days of storage among the treatments (Fig 4). After 12 days of storage, lowest antioxidant activity (5.18 µmol TE/g) was recorded in control fruits which did not differ significantly with 0.5% chitosan treated fruits. While the maximum antioxidant activity (5.70 µmol TE/g) was recorded in fruits treated with 1.0% chitosan followed by those treated with 0.75% chitosan (5.56 µmol TE/g).In this study, decrease in total phenolics content in mango during storage might be attributed to onset of fruit ripening (Barman *et al*. 2015, Sharma *et al*. 2012). However, delayed decline in phenolics content in chitosan treated fruit may be ascribed to suppressed ripening of mango by chitosan coating. The antioxidant activity of mango is mainly due to presence of ascorbic acid, phenolic compounds and carotenoid pigments (Asrey *et al*. 2013). In this experiment, chitosan treated fruits retained higher antioxidant activity than control. This was due to higher content of ascorbic acid and phenolic

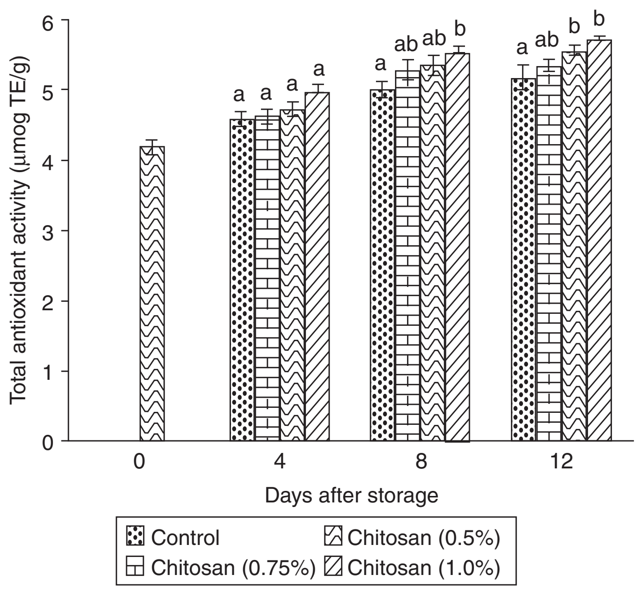

Fig 4 Effect of chitosan coating on total antioxidant activity of mango fruit during storage

compounds in chitosan treated fruits over control. Previous studies have also reported higher antioxidant activity owing to presence of higher amount of ascorbic acid and phenolic compounds in pomegranate (Barman *et al*. 2014a) and litchi (Barman *et al*. 2014b).

Effect of chitosan on total soluble solids (TSS) and titratable acidity (TA)

Total soluble solids content in the treated mango fruits increased with storage period (Table 1). Rapid increase in TSS content was recorded in control fruits than other treatments. After 12 days of storage, highest TSS content of 24.66°Brix was recorded in control fruits which did not differ significantly with 0.5% chitosan treated fruits (24.16°Brix). The lowest TSS content (22°Brix) was noted in 1.0% chitosan treated fruits. Titratable acidity in mango fruits declined with advancement of storage period (Table 1). Control fruits showed maximum decline in TA up to 0.25% after 12 days of storage. The maximum retention of TA (0.47%) was recorded in 1.0% chitosan treated fruits which was at par with 0.75% chitosan treated fruits (0.42%). The lower content of TSS and higher content of TA in chitosan treated fruits than control was probably due to slowing down of respiration rate and metabolic activity thereby retarding the ripening process of mango fruits. Owing to film forming property of chitosan, it modified the internal atmosphere by elevating CO_2 and reducing O_2 levels surrounding the fruit which supressed the respiration and ethylene evolution rates of mango (Dong *et al*. 2004). The reduced respiration rate slowed down the synthesis as well as use of metabolites. In addition, it also delayed the ripening of mango. As a result, hydrolysis of carbohydrates into sugar took place slowly thereby caused delayed increase in fruit TSS treated with chitosan. Similarly, slower increase in TSS in response to chitosan coating has also been reported previously in mango, banana and papaya (Kittur *et al*. 2001, Ali *et al*. 2011). The delayed ripening in chitosan treated fruits also slowed down the decrease in TA than control fruits as previously reported in strawberry and raspberry (Han *et al*. 2004).

The present study showed that treatments of mango fruit with chitosan (0.75% and 1.0%) are highly effective in reducing weight loss, decay loss, preserving quality attributes and bioactive compounds of fruit during postharvest storage at ambient condition. Significant differences were not recorded among the above treatments except in development of desired peel colour and total carotenoids content in the fruit pulp, which were delayed in 1.0% chitosan treated fruits. Therefore based on the findings, it can be suggested that 0.75% chitosan can be effectively used to control postharvest diseases, preserve fruit quality and prolong shelf life of mango during storage at ambient condition without any adverse effects on human health and environment.

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