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Enhancing postharvest storage life of pointed gourd (*Trichosanthes dioica* Roxb.) fruits with edible coatings

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Abstract
In the present investigation the effectiveness of two edible coatings viz. Carnauba wax and chitosan on postharvest storage behaviour of pointed gourd fruits was evaluated under ambient storage conditions (temperature 27.4-32.3°C and 70-81% RH). There were seven treatments, replicated thrice and experiment was laid out in completely randomized design. The fruits treated with 1.0% Carnauba wax gained highest sensory score (7.33). The physiological loss of weight in Carnauba wax treated fruits remained lower (0.63%) than the chitosan coated fruits. The minimum spoilage (3.08%) was also reported in 1.0%Carnauba wax. The highest disease reduction index of 83.98 was recorded in 1.0% Carnauba wax. The highest retention of total chlorophyll (5.89 mg/g) was recorded in 1.0% Carnauba wax and lowest in chitosan treated fruits at the end of storage period. Thus, among the two coating materials, Carnauba wax has the potential to extend storage life and preserve other quality attributes.

Keywords: Carnauba wax, chitosan, edible coating, pointed gourd, postharvest, storage

Introduction

The major quality factors of fresh fruits and vegetables contributing to the consumer acceptance are texture, colour, flavour, appearance, nutritional value and microbial safety. These quality factors are attributed towards the pre-harvest and postharvest conditions. The postharvest losses of fresh produce are a matter of grave concern because it rapidly deteriorates them during handling, transport and storage leading to huge qualitative and quantitative loss. The application of edible coatings emerges to be a potential approach in reducing such postharvest deterioration and preserving the quality during storage. An edible film is a thin layer of material which can be eaten by the consumer, be applied on the vegetable by wrapping, dipping, brushing or spraying (Wu *et al*., 2002) [1]. The main purpose of edible coating is basically to increase the natural barrier, if already present and to replace it in the cases where handling and washing have partially removed or altered it. It can also be safely consumed as part of the product and do not add unfavourable properties to the produce. Edible coatings act as partial barriers to CO_2 and O_2, moisture exchange, aroma compounds, decreasing the respiration rate of the fruit, water loss and preserving texture and flavour (Olivas and Barbosa-Canovas, 2005) [2]. In addition, it also has certain functional ingredients such as antioxidants, antimicrobials, nutrients and flavours to further enhance food stability, quality and safety (Debeaufort *et al*., 1998; Min and Krochta, 2005) [3, 4].

Edible coatings are classified into three categories based on the components used for preparation: (i) Hydrocolloids such as proteins, polysaccharides and alginate, (ii) lipids such as fatty acids, acylglycerol, waxes and (iii) composites (Donhowe and Fennema, 1993) [5]. Carnauba wax is an edible coating material in the lipid group, is a wax from the Brazilian Carnauba or Carnaubeira palm (*Copernicia prunifera*, Family: Arecaceae), obtained from the leaves of the palm (Parish *et al*., 2002; Puttalingamma, 2014) [6, 7]. The carnauba wax is well known for retaining postharvest properties of several fruits and vegetables during storage (Eum *et al*., 2009; Khuyen *et al*., 2008; Koley *et al*., 2009a, Kore and Kabir, 2011) [8-11]. Chitosan used as edible films or coatings are polysaccharides that come under the hydrocolloids group, is derived by deacetylation from chitin which is the second most abundant naturally occurring biopolymer after cellulose and is found in the exoskeleton of crustaceans, in fungal cell walls and other biological materials (No *et al*., 2007; Xu *et al*., 2005; Maghsoudlou *et al*., 2012) [12-14]. Several studies have shown that chitosan is effective at extending the shelf life of fruits and vegetables (Jiang and Li, 2001; Li and Yu, 2000, Pereda *et al*., 2010; Mendes de Souza *et al*., 2010) [15-18].

Pointed gourd (*Trichosanthes dioica* Roxb.) is an important river bed crop grown extensively in the states of West Bengal, Bihar, Uttar Pradesh and Assam in India (Chadha, 2000) [19]. Now a day's pointed gourd has become a popular vegetable on health conscious consumers' platter due to its nutritive value. In spite of the abundant production, this nutrient packed vegetable fails to store under ambient conditions for longer period and loses its freshness within 2-3 days. Pointed gourd shows non-climacteric type of behaviour and produces lesser amount of ethylene (Koley *et al*., 2009b) [20]. So, edible can enhances storability and preserves qualitative characters. Hence, the experiment was designed with the hypothesis that coating with two edible coatings viz. Carnauba wax and chitosan can preserve the quality attributes and extend the shelf life as well as the marketability of pointed gourd fruits. The present investigation was therefore has been undertaken with the objective to study the relative effectiveness of Carnauba wax and chitosan as edible coatings on the storage behaviour of pointed gourd.

Materials and Methods

The experiment was carried out in the laboratory conditions of the Department of Post Harvest Technology of Horticultural Crops, Bidhan Chandra Krishi Viswavidyalaya, West Bengal, India. Fresh fruits of pointed gourd cv. Kajli were used for the present experiment. Fruits of uniform colour, size and maturity, without injuries were selected. The fruits were washed with chlorine (100 ppm) water for 10 minutes using sodium hypochlorite (4.4% w/w, as a source of chlorine). Then they were surface dried by keeping under fan in an airy place. The fruits were dipped in Carnauba wax and chitosan solutions respectively for 10 minutes. All the treatments were kept on trays and stored in normal room condition. The temperature and relative humidity of the atmosphere during the study period ranged from 27.4-32.3°C and 70-81% respectively. There were seven treatments viz., T_1-Control, T_2-Carnauba wax 0.25%, T_3-Carnauba wax 0.50%, T_4-Carnauba wax 1.0%, T_5-Chitosan 0.25%, T_6-Chitosan 0.50% and T_7-Chitosan 1.0%. The Carnauba wax was prepared in the laboratory by solubilizing it in trimethyl amine and oleic acid with the help of boiled water at a temperature of about 100°C. For Chitosan coating, the solution was prepared by dissolving 1% Chitosan (Sigma Chemical Co.) in a 0.5% glacial acetic acid and distilled water. The pH value of the Chitosan solution was then adjusted to 5.6 using 0.1M NaOH (Bal, 2013) [21]. From the stock solution three different concentration of coatings viz., 0.25, 0.50 and 1.0% were prepared. The analysis of data obtained in the experiment was analyzed by Completely Randomized Design with three replications, by adopting the statistical procedures of Gomez and Gomez (1984) [22]. The means between treatments were compared by Duncan's multiple range tests (DMRT) (Duncan, 1955) [23].

Sensory evaluation

During the period of study, observations on sensory properties were estimated by using 9-point Hedonic scale for their sensory characteristics like appearance, texture and overall acceptability. The scores were assigned from extremely liked (9) to disliked extremely (1) (Kaur and Aggarwal, 2015) [24].

Physiological loss in weight (PLW)

The weight of individual fruit in the experiment was taken on the day of observation and the percentage of loss in weight on the day of observation was calculated on the basis of the initial weight and expressed in percentage (Koraddi and Devendrappa, 2011) [25].

$$\text{PLW (\%)} = \frac{\text{Initial fruit weight} - \text{Weight of fruit on observation day}}{\text{Initial fruit weight}} \text{ X } 100$$

Spoilage

Spoilage percentage was observed after every 48 hours and was calculated as described below (Bhat *et al*., 2014) [26].

$$\text{Spoilage (\%)} = \frac{\text{Number of decayed fruits at the time of sampling}}{\text{Initial number of fruits}} \text{ X } 100$$

Disease reduction index (DRI)

The disease reduction index was estimated from the numbered fruits of each experimental lot at each date of observation and disease reduction index was calculated by the following formula (Gutter, 1969) [27].

$$\text{DRI} = \frac{\text{Percent disease in control} - \text{Percent disease in treatment}}{\text{Percent disease in control}}$$

Chlorophyll content

Chlorophyll a, b and total chlorophyll was extracted in 80% acetone and absorption was measured at 663 nm and 645 nm by spectrophotometer (Systronics Spectrophotometer 166) and expressed as mg chlorophyll per gram of fresh tissue at regular time interval. Using the absorption coefficients, the amount of chlorophyll is calculated using the following equations (Sadasivam and Manickam, 1996) [28]:

$$\text{mg chlorophyll a/ g tissue} = 12.7\ (A663) - 2.69\ (A645) \text{ x } \frac{V}{1000 \text{ X } W}$$

$$\text{mg chlorophyll b/ g tissue} = 22.9\ (A645) - 4.68\ (A663) \text{ x } \frac{V}{1000 \text{ X } W}$$

and mg total chlorophyll/ g tissue = 20.2 (A645) + 8.02 (A663) x $\frac{V}{1000 \text{ X } W}$

where,
A = absorbance at specific wavelengths
V = final volume of chlorophyll extract in 80% acetone
W = fresh weight of tissue extracted.

Results and Discussion

The sensory properties assessed on the basis of sensory score are presented in Table 1. Up to 4th day of storage, the sensory quality of fruits treated with Carnauba wax 1.0% was very good to good as indicated by sensory score of 9.00 and 7.67 respectively while in other treatments the sensory scores indicated were good. On 6th day, more or less the results were fair to non-acceptable in all treatments except in T_4 i.e. Carnauba wax 1.0%, (7.33) followed by T_3 (Carnauba wax 0.50%) (6.00), was significantly superior to other treatments. The sensory properties were superior in 1.0% and 0.50% Carnauba wax which were in accordance with the studies of Patel *et al*. (2013) [29] in pointed gourd treated with Carnauba wax (1 and 2%) with or without 6N-Benzyladenine 25 ppm. Wax dry up on the surface to produce a membrane which is differentially permeable to gases which manipulates levels of oxygen and carbon dioxide and create modified atmosphere rich in carbon dioxide, which delayed ripening (Chakraborty *et al*., 2002; Smith *et al*., 1987) [30, 31].

Table 1: Effect of edible coatings on sensory properties of pointed gourd fruits in storage

Treatments	Sensory properties		
	Days in storage		
	2	4	6
T_1	6.33 a	3.67 a	2.00 a
T_2	8.33 cd	6.33 b	5.33 cd
T_3	8.67 cd	6.67 bc	6.00 de
T_4	9.00 d	7.67 c	7.33 e
T_5	7.67 bc	5.00 a	4.33 bc
T_6	8.00 cd	4.33 a	3.33 ab
T_7	6.67 ab	4.00 a	3.00 ab
C.D. (0.05)	1.091	1.280	1.391
SEm ±	0.356	0.418	0.454

(Means in the column followed by the same alphabet do not differ significantly by DMRT at 5%)
(T_1-Control, T_2-Carnuba wax 0.25%, T_3-Carnuba wax 0.50%, T_4-Carnuba wax 1.0%, T_5-Chitosan 0.25%, T_6-Chitosan 0.50%, T_7-Chitosan 1.0%)

Physiological loss of weight increased gradually in all the treatments with advancement of storage period (Table 2). The physiological loss of weight in fruits after 2 days storage was noted least in T_4 (Carnauba wax 1.0%) (0.06%) followed by the highest in control (15.73%). On 4th day of storage also similar trend of weight loss prevailed and it differed from 0.63% in T_4 (Carnauba wax 1.0%) to 22.73% in control. However, per cent increase in weight loss was recorded significantly low in 1.0% Carnauba wax (0.63%) treated fruits followed by 0.50% Carnauba wax (0.69%) and highest in control (24.60%) after 6 days in storage. Thus, it was observed that the physiological loss of weight in Carnauba wax treated pointed gourd fruits remained lower than the chitosan coated fruits. Edible coating works by restricting the oxygen intake through the skin of fresh fruit and carbon dioxide out, thus delaying maturity or ripening process by slowing down respiration, without causing anaerobiosis (Curtis, 1988) [32]. Low water loss in Carnauba wax treated fruit may be attributed to the water and fat binding properties of Carnauba wax augmented with lower activities of enzymes (Koley *et al*., 2009a) [10]. Koley *et al*. (2009a) [10] and Chakraborty *et al*. (2002) [30] also recorded low physiological loss in weight with Carnauba wax and Semperfresh respectively in pointed gourd.

Table 2: Changes in the physiological loss in weight of pointed gourd fruits in storage as affected by the edible coatings

Treatments	Physiological loss in weight (%)		
	Days in storage		
	2	4	6
T_1	15.73 d (23.36)	22.73 d (28.46)	24.60 d (29.72)
T_2	0.81 a (4.16)	1.05 a (5.87)	1.44 a (6.79)
T_3	0.17 a (2.32)	0.49 a (3.66)	0.69 a (4.46)
T_4	0.06 a (1.316)	0.63 a (4.46)	0.63 a (4.46)
T_5	3.39 b (10.61)	18.81 b (25.69)	20.53 b (26.93)
T_6	3.22 b (10.32)	19.87 b (26.45)	23.22 c (28.79)
T_7	13.32 c (21.39)	21.46 c (27.59)	24.16 cd (29.43)
C.D. (0.05)	2.598	1.714	1.933
SEm ±	0.848	0.560	0.631

*figures in parenthesis indicate angular transformed values
(Means in the column followed by the same alphabet do not differ significantly by DMRT at 5%)
(T_1-Control, T_2-Carnuba wax 0.25%, T_3-Carnuba wax 0.50%, T_4-Carnuba wax 1.0%, T_5-Chitosan 0.25%, T_6-Chitosan 0.50%, T_7-Chitosan 1.0%)

The spoilage of pointed gourd fruits was minimum with Carnauba wax treatment followed by chitosan from second to six days of storage (Table 3). On 2nd day of storage, spoilage was recorded in control and chitosan treated fruits. The highest spoilage was observed in T_1 (53.26%) whereas, no spoilage was noted in T_3 (Carnauba wax 0.50%) and T_4 (Carnauba wax 1.0%) respectively on 4th day of storage. But with the advancement of storage period, the effectiveness of coating materials failed. On 6th day, the minimum spoilage was reported in T_4 (Carnauba wax 1.0%) (3.08%) followed by T_3 (Carnauba wax 0.50%) (5.82%) with complete spoilage in control and T_7 (Chitosan 1.0%). Fresh fruits and vegetables are susceptible to a variety of postharvest decays that can be reduced by treatment with a coating or wax which results in inhibiting mould growth. Carnauba wax prevent the incidence of moulds by sealing the open the surface on the fruit there by controlled the spoilage (Torres *et al*., 2009) [33].

Table 3: Influence of edible coating materials on spoilage of pointed gourd fruits

Treatments	Spoilage (%)		
	Days in storage		
	2	4	6
T_1	18.82 d (25.70)	53.26 d (46.85)	100.00 c (90.00)
T_2	0.00 a (0.81)	3.56 b (10.76)	6.33 b (14.56)
T_3	0.00 a (0.81)	0.00 a (0.81)	5.82 b (13.94)
T_4	0.00 a (0.81)	0.00 a (0.81)	3.08 a (10.11)
T_5	15.63 b (23.28)	47.88 c (43.77)	99.03 c (84.36)
T_6	15.96 bc (23.54)	48.42 c (44.08)	99.11 c (86.85)
T_7	16.57 c (24.01)	49.31 c (44.59)	100.00 c (90.00)
C.D. (0.05)	0.663	1.439	3.781
SEm ±	0.216	0.470	1.235

*figures in parenthesis indicate angular transformed values
(Means in the column followed by the same alphabet do not differ significantly by DMRT at 5%)
(T_1-Control, T_2-Carnuba wax 0.25%, T_3-Carnuba wax 0.50%, T_4-Carnuba wax 1.0%, T_5-Chitosan 0.25%, T_6-Chitosan 0.50%, T_7-Chitosan 1.0%)

The DRI of pointed gourd fruits were presented in Table 4, which revealed that the disease incidence started from 2nd day onwards in storage with minimum DRI in control (0.00) and T_7 (Chitosan 1.0%) (17.62). The DRI recorded in all treated fruits were significantly higher as compared to control till the end of storage. T_3 (Carnauba wax 0.50%) and T_4 (Carnauba wax 1.0%) recorded highest DRI on 4th day of storage. However, on 6th day, highest DRI was recorded in T_4 (Carnauba wax 1.0%) (83.98) followed by lowest being 0.89 (Chitosan 0.50%) and 0.00 (Chitosan 1.0% and control) respectively. The fruits treated with Carnauba wax maintained significantly higher DRI values than that treated with chitosan. The pointed gourd fruits coated with 1.0% Carnauba wax abridged spoilage and sustained fairly high DRI compared to uncoated fruits throughout the storage period. Coatings act as lubricants to reduce surface injury, scarring, and chafing (Hardenburg, 1967; Hartman and Isenberg, 1956) [34, 35]. The decay due to opportunistic wound pathogens is lessened due to less wounding of the fruit. Similar results were observed in waxed citrus and cucumber (Waks *et al*., 1985; Baldwin *et al*., 1997; Mack and Janer, 1942) [36-38].

Table 4: Disease reduction index at different days in storage of pointed gourd fruits

Treatments	Disease reduction index		
	Days in storage		
	2	4	6
T_1	0.00	0.00	0.00
T_2	100.00	77.83	76.39
T_3	100.00	100.00	82.13
T_4	100.00	100.00	83.98
T_5	39.24	23.75	4.22
T_6	28.40	19.32	0.89
T_7	22.42	17.62	0.00

(T_1-Control, T_2-Carnuba wax 0.25%, T_3-Carnuba wax 0.50%, T_4-Carnuba wax 1.0%, T_5-Chitosan 0.25%, T_6-Chitosan 0.50%, T_7-Chitosan 1.0%)

The change in colour of pointed gourd fruits from green to orange continued over the storage period as presented in Table 5. The initial chlorophyll a, chlorophyll b and total chlorophyll content of pointed gourd fruits were 6.10 mg/g, 2.91 mg/g and 9.01 mg/g respectively. This reference value decreased significantly with the storage time. On 2nd day of storage, maximum chlorophyll a (5.98 mg/g), chlorophyll b (2.85 mg/g) and total chlorophyll (8.82 mg/g) were recorded in T_4 (Carnauba wax 1.0%) and minimum in control. The same trend prevailed on 4th day as well as on 6 days after storage with highest retention of chlorophyll a (3.99 mg/g), chlorophyll b (1.90 mg/g) and total chlorophyll (5.89 mg/g) in T_4 (Carnauba wax 1.0%) and lowest chlorophyll a (1.63 mg/g), chlorophyll b (0.77 mg/g) and total chlorophyll (2.41 mg/g) in T_7 (Chitosan 1.0%). The fruits in control were no longer available for analysis. In addition, significant differences in chlorophyll contents were found in Carnauba wax coated pointed gourd fruits compared to chitosan coated samples. The efficacy of T_4 (Carnauba wax 1.0%) and T_3 (Carnauba wax 0.50%) treatment might be due to low activity of pectin methyl esterase and delayed chlorophyll degradation in parallel with enzymatic action (Koley *et al.*, 2009a) [10]. Such observation are in conformation with those of Olivas and Barbosa-Conovas (2005) [2], on use of edible coating in fresh cut fruit, Dabrowski *et al.* (1989) [39] in pumpkin and Machado *et al*. (2012) [40] in tangor.

Table 5: Chlorophyll a, chlorophyll b and total chlorophyll content at different days of storage of pointed gourd

Treatments	Pigment concentration (mg/g)								
	Days in storage								
	2			4			6		
	Chlorophyll a	Chlorophyll b	Total chlorophyll	Chlorophyll a	Chlorophyll b	Total chlorophyll	Chlorophyll a	Chlorophyll b	Total chlorophyll
T_1	3.34 a	1.59 a	4.93 a	2.16 a	1.03 a	3.19 a	-	-	-
T_2	4.94 c	2.35 c	7.30 d	4.21 e	2.00 d	6.21 d	3.05 e	1.45 de	4.50 de
T_3	5.19 c	2.47 c	7.66 d	4.68 f	2.23 e	6.91 e	3.51 f	1.67 ef	5.18 ef
T_4	5.98 d	2.85 d	8.82 e	5.16 g	2.46 f	7.62 f	3.99 g	1.90 f	5.89 f
T_5	4.21 b	2.01 b	6.22 c	3.90 d	1.86 cd	5.76 cd	2.56 d	1.22 cd	3.78 cd
T_6	4.15 b	1.98 b	6.13 bc	3.58 c	1.70 c	5.29 c	2.10 c	1.00 bc	3.10 bc
T_7	3.90 b	1.86 b	5.75 b	2.81 b	1.34 b	4.14 b	1.63 b	0.78 b	2.41 b
C.D. (0.05)	0.361	0.260	0.407	0.258	0.220	0.570	0.198	0.243	0.771
SEm ±	0.118	0.085	0.133	0.084	0.072	0.186	0.065	0.079	0.252

(T_1-Control, T_2-Carnuba wax 0.25%, T_3-Carnuba wax 0.50%, T_4-Carnuba wax 1.0%, T_5-Chitosan 0.25%, T_6-Chitosan 0.50%, T_7-Chitosan 1.0%)

(Means in the column followed by the same alphabet do not differ significantly by DMRT at 5%)

Conclusion

The present investigation revealed that coating of pointed gourd fruits with Carnauba wax was more promising in preserving the postharvest storage behaviour than chitosan. It could be concluded that Carnauba wax was efficient in delaying ripening, reducing the weight loss, decay incidences, maintaining pigment concentration and enhanced the shelf-life of pointed gourd fruits during storage. Carnauba wax can be easily applied, cost effective and hence can be used commercially to prolong the shelf life of pointed gourd fruits.

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