HortFlora Research Spectrum
Vol. 5, Issue 1; 28-33 (March 2016)
www.hortflorajournal.com
ISSN: 2250-2823

IMPROVEMENT IN SHELF-LIFE OF STRAWBERRY (*Fragaria* × *ananassa* Duch.) CV. WINTER DAWN WITH EDIBLE COATINGS ENRICHED WITH CHITOSAN

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ABSTRACT: A lab experiment was conducted during February-March, 2015 on strawberry (*Fragaria* × *ananassa* Duch.) to study the effect of Calcium Chloride, Carboxymethyl cellulose and Chitosan on physical and chemical characters having 14 treatments treated with calcium chloride and CMC (1%, 2% and 3% each) without adding Chitosan and with Chitosan 1%. Application of Carboxymethyl cellulose 2% + Chitosan 1% to the strawberry fruits helped to maintain all the characters attributing to quality. These treatments reduced the weight loss and spoilage during storage. Under these treatments strawberry could be stored for over 12 days (fruit still reddish in colour) compared to the control which started turning turbid yellow soon after 9 days. These treatments can be used satisfactorily by the fruit growers and the fruit merchants in order to prolong the storage life of strawberry fruits up to 12 days. However, these results are only indicative and require further experimentation to arrive at more consistent and final conclusion.

Keywords : *Strawberry, calcium chloride, carboxymethyl cellulose, chitosan*

Strawberry (*Fragaria* × *ananassa* Duch.) is one of the most fascinating fruit of the world. It is a cross between two species of American wild strawberry: a large fruited species, *Fragaria chiloensis*, originally from Chile and *Fragaria virginiana*, originally from Virginia, USA. The United States is the world's largest producer of strawberries accounting for about 30 per cent of the world strawberry production (Morgan, 20). Strawberry belongs to the family Rosaceae and its cultivated varieties are octaploid (2n=56) in nature. Botanically, the strawberry is an aggregate fruit. From the postharvest perspective, strawberries are extremely perishable and have a very demanding postharvest handling requirement. Their short postharvest life is mainly due to their susceptibility towards mechanical injury, physiological deterioration, water loss and microbial decay. Loss of the fruit quality starts with the loss of membrane integrity that generally becomes the leading event in the senescence.

Postharvest application of calcium prevents postharvest disorders, retards fruit ripening and decreases postharvest decay (Sadat *et al.,* 25). Moreover, strawberry fruits treated with calcium maintain increased calcium content in the cell wall of the fruit tissue which maintains fruit firmness and soluble solids content, without affecting their sensory quality (Guzman and Barrett, 14). CMC is a cellulose derivative that had received significant attention with several examples of applications in a variety of fruit and vegetables. Edible coatings based on cellulose derivatives have been extensively used to delay ripening and loss of quality in fresh products such as pears (Zhou *et al.,* 28), cherries (Yaman and Bayoindirli, 27) and many other fruits.These coatings may retard ripening and increase the shelf life of coated produce, without creating severe anaerobic conditions (Baldwin *et al.,* 5). Chitosan (poly b-(1, 4) N-acetyl-D-glucosamine) polymer is industrially produced by chemical de-acetylation of the chitin found in arthropod exoskeletons. Chitosan coatings have been reported to limit fungal decay and delay the ripening of several commodities, including strawberry (Ribeiro *et al.,* 24).

Article's History:
Received : 20-01-2016 ***Accepted : 19-02-2016***

Therefore the following work had been done regarding the effect of calcium chloride, Carboxymethyl cellulose and Chitosan on the postharvest quality characteristics of strawberry fruits.

MATERIALS AND METHODS

Freshly harvested, uniform maturity size and quality fruits of strawberry cv. 'Winter Dawn' free from insect pest and microbial infection were procured from the Hi-Tech unit of the College of Horticulture and Forestry, Jhalarapatan, Jhalawar. These were brought to the laboratory and sorted out. Bruised, off types and misshaped fruits were removed.

CMC (1%, 2% and 3%) was prepared by solubilizing 1 g, 2 g and 3 g of CMC powder in 100 mL of water-ethyl alcohol mixture (3:1 L/L) at 75°C under magnetic stirring for 15 min. Ethyl alcohol was used to reduce the drying time and to obtain the transparent and shiny methyl cellulose coating. Then glycerol monostearate (0.75%) was added and the solution was stirred for 10 min under the same conditions.CH (1%, w/v) was dispersed in the solution of glacial acetic acid (0.5%, v/v) prepared in water at 40°C. The solution was heated and agitated constantly for 12 h. The pH of the solution was adjusted to 5.6 with 1 N NaOH and 0.1 mL of glycerol monostearate (0.75%) was added to the solution as a plasticizer to improve the strength and flexibility of the coating solutions. The solution was stirred by a magnetic stirrer at room temperature to achieve complete dispersion.For calcium chloride treatments, first desired quantity of the calcium chloride was weighed and dissolved in small quantity of water, then required volume was made up by addition of distilled water.Fruits were dipped in different concentrations of calcium chloride (1%, 2% and 3%) and CMC (1%, 2% and 3%) without adding Chitosan and with Chitosan 1% for 5 minutes followed by placing them on newspaper sheet for drying in shade for 30 minutes. After treatment the fruits of each lot were packed in specially designed "punnets" made up of propylene material. The fruits were stored in refrigerated condition at 4±1°C and 85-90 per cent relative humidity.

Observations regarding physiological loss in weight (PLW), spoilage, overall organoleptic score, acidity, ascorbic acid, total sugars and anthocyanin content were recorded at 3 days interval during the storage period of 12 days. PLW was calculated by weighing the fruits on physical balance, spoilage was calculated on % basis, anthocyanin content, acidity and ascorbic acid content of the sample was determined by titrometric method as described by Ranganna (23) and total sugar was determined byanthrone reagents method (Dubois *et al.,* 9). Organoleptic evaluation was carried out by a panel of 7 judges who scored on 9 poin thedonic scale (Amerine *et al.,* 3). The initial values as observed at the start of the experiment (0 days) were- sensory score: 8.11, acidity: 0.85 %,ascorbic acid: 52.5 mg/100 g, total sugars: 5.52 % and anthocyanin content: 36.51 mg/100 g.

Statistical analysis : The experiment was arranged in completely randomized design (CRD), with 14 treatment combinations T_0 : Distilled water control, T_1 : $CaCl_2$ 1%, T_2 : $CaCl_2$ 2%, T_3 : $CaCl_2$ 3%, T_4 : CMC 1%, T_5 : CMC 2%, T_6 : CMC 3%, T_7 : Chitosan 1%, T_8 : $CaCl_2$ 1% + Chitosan 1%, T_9 : $CaCl_2$ 2% + Chitosan 1%, T_{10} : $CaCl_2$ 3% + Chitosan 1%, T_{11} : CMC 1% + Chitosan 1%, T_{12} : CMC 2% + Chitosan 1% and T_{13} : CMC 3% + Chitosan 1% each having 3 replicates. Each replicate was comprised of 10 uniform sized fruits of strawberry cv. Winter Dawn. Data were subjected to analysis of variance (ANOVA) using statistical software OPSTAT, CCS HAU, Haryana, India and the critical difference (C.D. P=0.05)was used to compare the means (Gomez and Gomez, 13). Data expressed as percentage were transformed in to arc sine square root values to normalize the distribution before analysis of variance; however, the percentages are shown as untransformed data.

RESULTS AND DISCUSSION

PLW and Spoilage : Treatments had statistically significant effect on PLW and spoliage during 12 days of storage (Table 1) . It is evident from the table that the PLW and spoilage increased continuously till the end of storage period. On the 12th day the maximum PLW and spoilage was observed in control (23.38 % and 61.22 %), whereas, the minimum PLW and spoilage was observed in CMC 2% + Chitosan 1% (8.80 % and 21.75 %) treated fruits.The maximum physiological weight loss in control fruits (T_0) might be due to high rate of respiration and transpiration from the fruit skin. However, PLW was found to be very slow in fruits treated with CMC 2% + Chitosan 1% as compared to control and other treatments, which could be due to the fact that composite coatings serves as a semi-permeable barrier against oxygen, carbon dioxide and moisture, thus reducing respiration, water loss and oxidation reactions (Maqbool *et al.,* 19; Deval *et al.* 8). These results are in line with the observations as reported by Ribeiro *et al.* (24) in strawberry and Abbasi *et al.* (1) in mango.

The spoilage was mainly brought about by rotting caused by pathogens. Reduction in spoilage with the use of CMC and chitosan was probably due to the anti-microbial activity of chitosan which induces chitinase, a defense enzyme and catalyze the hydrolysis of chitin, a common component of fungal cell-walls thus preventing the growth of fungi on the fruits (El Ghaouth *et al.,* 11 and Butler *et al.,* 6). The present results are in conformity with those of Hernandez-Munoz *et al.* (15) and Nadim *et al.* (21) in strawberry.

Sensory Evaluation : The overall organoleptic score of strawberry fruits was analyzed (Table 2) which indicated that the overall organoleptic rating based on colour, aroma and taste of strawberry fruits decreased with the advancing period of storage. This could be

Table 1: Effect of calcium chloride, carboxymethyl cellulose and chitosan on physiological loss in weight (%) and spoliage (%) of strawberry.

Treatments	PLW (%)				Spoilage* (%)		
	3 Days	6 Days	9 Days	12 Days	6 Days	9 Days	12 Days
T_0	4.72	12.44	18.66	23.38	34.21 (35.80)	45.00 (42.13)	61.22 (51.48)
T_1	4.20	10.40	15.10	19.30	33.00 (35.06)	39.23 (38.78)	55.07 (47.91)
T_2	4.05	10.05	14.55	18.60	30.93 (33.79)	37.23 (37.60)	59.00 (50.18)
T_3	4.00	9.95	14.40	18.40	33.21 (35.19)	43.08 (41.02)	57.00 (49.02)
T_4	3.25	6.80	8.85	12.10	31.12 (33.91)	35.22 (36.40)	55.07 (47.91)
T_5	2.47	5.90	7.83	10.30	30.11 (33.28)	33.34 (35.27)	51.14 (45.65)
T_6	3.00	6.50	8.50	11.50	30.98 (33.82)	37.22 (37.60)	53.15 (46.81)
T_7	4.34	10.42	15.00	19.34	24.00 (29.33)	29.91 (33.15)	35.40 (36.51)
T_8	4.10	9.90	14.20	18.30	21.61 (27.70)	25.42 (30.28)	28.78 (32.44)
T_9	3.98	9.70	13.92	17.90	23.52 (29.01)	29.43 (32.85)	33.82 (35.56)
T_{10}	3.90	9.40	13.40	17.30	22.82 (28.54)	27.52 (31.64)	31.62 (34.22)
T_{11}	2.14	5.50	7.30	9.50	19.64 (26.31)	22.77 (28.50)	25.68 (30.45)
T_{12}	2.02	5.15	6.78	8.80	16.58 (24.03)	19.09 (25.91)	21.75 (27.80)
T_{13}	2.08	5.20	7.00	9.08	18.58 (25.53)	21.52 (27.64)	24.17 (29.45)
C.D. (P=0.05)	0.55	0.37	0.63	0.62	1.18 (0.78)	1.41 (0.89)	0.94 (0.57)

*Figures in parenthesis are re-transformed values

Table 2 : Effect of calcium chloride, carboxymethyl cellulose and chitosan on overall organoleptic score, acidity (%) and ascorbic acid (mg/100 g) of strawberry.

Treatments	Overall organoleptic score				Acidity (%)				Ascorbic acid (mg/100g)			
	3Days	6 Days	9 Days	12 Days	3 Days	6 Days	9 Days	12 Days	3 Days	6 Days	9 Days	12 Days
T_0	6.00	5.11	3.67	2.11	0.73	0.62	0.50	0.38	44.00	37.70	30.70	18.70
T_1	6.78	5.89	5.11	4.00	0.74	0.64	0.53	0.42	44.20	38.10	31.30	19.40
T_2	6.78	5.89	5.33	4.11	0.75	0.65	0.55	0.43	44.30	38.30	31.60	19.75
T_3	6.78	5.89	5.33	4.11	0.74	0.63	0.52	0.41	44.40	38.50	32.00	20.16
T_4	6.78	6.00	4.89	3.89	0.76	0.68	0.59	0.49	44.70	39.10	33.20	21.37
T_5	6.66	5.78	5.11	4.00	0.77	0.71	0.63	0.56	44.90	39.50	33.70	21.97
T_6	7.11	6.33	5.33	4.33	0.75	0.68	0.58	0.51	45.10	39.90	34.30	22.60
T_7	6.67	5.89	4.89	3.89	0.78	0.70	0.65	0.53	45.50	40.50	35.50	24.81
T_8	6.56	5.78	5.00	4.00	0.79	0.73	0.67	0.60	46.10	41.30	36.50	24.85
T_9	7.33	6.33	5.44	4.67	0.80	0.74	0.69	0.61	45.90	41.31	36.75	25.12
T_{10}	7.11	6.33	5.33	4.67	0.79	0.73	0.67	0.61	46.30	41.90	37.55	25.97
T_{11}	6.67	5.78	4.78	4.22	0.81	0.76	0.72	0.66	47.10	43.30	39.60	28.07
T_{12}	7.56	6.89	6.33	5.67	0.83	0.80	0.78	0.73	47.70	44.00	41.00	30.05
T_{13}	7.10	6.33	5.44	4.44	0.82	0.78	0.75	0.70	47.30	43.50	40.15	29.12
C.D. (P=0.05)	0.58	0.54	0.59	0.99	0.04	0.05	0.04	0.02	0.23	0.55	0.37	0.67

probably due to occurrence of ripening process in the fruits followed by senescence and spoilage. Color is related to the presence of different pigments; aroma is attributed to esters, alcohols, acids and carbonyl compounds and taste is mainly due to sugar acid ratio. During ripening, transition of chlorophyll into carotenoids, biochemical conversion of starch into sugar and loss of organic acid through oxidation are responsible for the changes in these sensory parameters.

Among all the coatings used, CMC 2% + Chitosan 1% improved the shelf life of strawberries by maintaining the overall organoleptic score at higher levels as compared to control and other treatments.On the 12th day of storage, the maximum organoleptic score (5.67) was reported in CMC 2% + Chitosan 1% treated fruits and the minimum score was obtained in control (2.11). In control, the fruits had lowest score due to faster degradative changes in carbohydrate, acids, phenolic compounds and spoilage which accounted for the loss of colour and flavour of the fruits (Malundo *et al.,* 18) whereas, the highest score in coatings might be due to their ability to retard ripening processes.

Acidity : It is evident from Table 2 that all the treatments had a significant difference pertaining to acidity (%) of the stored strawberry cv. Winter Dawn. It was observed that the acidity decreased with the advancement of storage period in all the treatments.On the 12th day of storage, the maximum acid content (0.73 %) was observed under the treatment of CMC 2% + Chitosan 1% and the minimum acidity (0.38 %) was recorded in the control fruits followed by $CaCl_2$ 3% (0.41 %).The acidity of the fruit was highest at the start of the storage and it decreased with the advancement of storage period. It may be due to the fact that the strawberries were harvested at red stage, when accumulation of organic acid was over followed by rapid utilization of acid of the fruit pulp in respiration process and degradation of citric acid during subsequent storage which in turn might have influence on reduction in acidity due to their conversion into carbon dioxide and water and further utilization in metabolic process in the fruit (Maftoonazad *et al.,* 17). These results are in conformity to that obtained by El Ghaouth *et al.* (11), Garcia *et al.* (12) and Hernandez-Munoz *et al.* (15) in strawberry.

Ascorbic acid : The chemical treatments had significant effect on the ascorbic acid content of the fruits. On the 3rd day of storage, the maximum ascorbic acid content (47.70 mg/100 g) was recorded in fruits treated with CMC 2%+ Chitosan 1% which got reduced up to (30.05 mg/100 g) on the 12th day of storage. On the other hand in control fruits, the ascorbic acid content sharply declined from an initial value of 30.05 mg/100 g to 18.70 mg/100 g on 12th day of storage. In general, a trend of gradual decline in ascorbic acid content of the fruits was observed as the storage period progressed in all the treatments. The rate of decrease in ascorbic acid was significantly higher in untreated fruits as compared to treated fruits. This

Table 3: Effect of calcium chloride, carboxymethyl cellulose and chitosan on tTotal sugar (%) and anthocyanin content (mg/100 g) of strawberry.

Treatments	Total sugar (%)				Anthocyanin content (mg/100 g)			
	3 Days	6 Days	9 Days	12 Days	3 Days	6 Days	9 Days	12 Days
T_0	7.07	8.20	8.00	6.66	42.17	43.67	46.17	47.67
T_1	6.47	7.39	7.13	5.82	42.12	43.52	45.87	47.27
T_2	6.36	7.26	6.97	5.70	41.99	43.48	45.73	47.09
T_3	6.23	7.11	6.82	5.56	41.67	43.00	45.15	46.48
T_4	6.19	7.02	6.69	5.43	41.67	42.97	45.07	46.37
T_5	6.15	6.94	6.59	5.31	39.78	41.03	43.07	44.32
T_6	6.13	6.92	6.58	5.28	39.54	40.69	42.67	43.82
T_7	6.07	6.82	6.39	5.21	38.88	39.98	41.78	42.88
T_8	6.05	6.75	6.30	5.10	38.87	39.92	41.64	42.69
T_9	6.02	6.70	6.22	5.05	38.23	39.25	40.87	41.89
T_{10}	6.00	6.66	6.17	5.00	37.94	38.91	40.41	41.38
T_{11}	5.97	6.58	6.03	4.90	37.88	38.87	40.21	41.11
T_{12}	5.91	6.38	5.69	4.67	37.24	37.84	38.82	39.42
T_{13}	5.95	6.53	5.94	4.83	37.57	38.43	39.71	40.57
C.D. (P=0.05)	0.04	0.10	0.19	0.19	1.53	0.89	1.25	0.66

might be due to rapid loss of L-ascorbic acid by oxidation because of greater availability of oxygen. Another reason might be due to rapid conversion of L-ascorbic acid into dehydro-ascorbic acid in the presence of enzyme ascorbinase (Atress *et al.,* 4). The maximum retention of ascorbic acid (30.05 mg/100 g) with CMC 2% + Chitosan 1% treatment might be due to influence on reducing respiration as well as oxidation in the fruits. The present findings confirm the observations of Dhaka *et al.* (7) in mango, Atress *et al.* (4) in strawberry and Adetunji *et al.* (2) in sweet orange.

Total sugars : The data presented in Table 3 showed that total sugar content of stored fruits was affected significantly by the different chemical treatments. On the 12th day, the maximum total sugar content was recorded in control (6.66 %) and the minimum total sugar (4.67 %) was observed in CMC 2%+ Chitosan 1% treated fruits. The total sugar content of fruits showed an initial rise and then decline during storage. The increase in total sugar might be due to the hydrolysis of polysaccharides into simple sugars and decrease in total sugars at the end of storage period might be due to the utilization of simple sugars in respiration process and their conversion into carbon dioxide and water. The treatment involving CMC 2% + Chitosan 1% reported the least increase in total sugar due to their ability to retard ripening process as compared to control. These results are in conformity with those of Kumar and Nath (16) in aonla and Garcia *et al.* (12) and Tanada-Palmu *et al.* (26) in strawberry.

Anthocyanin content : It is evident from the Table 3 that the anthocyanin content of strawberry fruits increased with the advancement of the storage period. On the 12^{th} day of storage, the maximum anthocyanin content was in untreated fruits (47.67 mg/100 g) and the minimum anthocyanin content (39.42 mg/100 g) was observed in CMC 2% + Chitosan 1% treated fruits. This increase might be due to synthesis of anthocyanin, a pigment contributing to the red colour in strawberry. From the data it can be concluded that control fruits were having high level of anthocyanin than the coated ones due to the fact that the edible coatings act as gas barriers thus modifying the internal atmosphere in the fruits (higher levels of CO_2 and low levels of O_2) and this in turn may retard the biochemical reactions leading to anthocyanin synthesis.These results are in conformity with those of El Ghaouth *et al.* (10) and Perez and Sanz (22) in strawberry.

From the findings of the experiment it may be conducted that the application of Carboxymethyl cellulose 2% + Chitosan 1% to the strawberry fruits helped to maintained all the characters attributing to quality.

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Citation : Bola P.K., Jain S.K. and Choudhary A. (2016). Improvement in shelf-life of strawberry (*Fragaria* × *ananassa* Duch.) cv. Winter Dawn with edible coatings enriched with chitosan. *HortFlora Res. Spectrum,* **5**(1) : 28-33.