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Original Article

Effects of modified atmosphere packaging (MAP) and natural edible coatings on controlling postharvest fungal infection, shelf life extension and quality retention of strawberry (*Fragaria* × *ananassa* Duch.)

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Strawberry, MAP, aloe vera, garlic, chitosan, fungus, shelf life, quality

A B S T R A C T

Strawberry is a nutritious but highly perishable fruit, which require appropriate technology to maintain postharvest quality. Hence, an experiment was conducted to develop a safe technology for controlling postharvest fungal infection, shelf life extension and quality retention of strawberry using modified atmosphere packaging (MAP) and natural edible coatings. Two factor experiment comprised three MAP viz. control (without packaging), low-density perforated polyethylene (LDPPE) and low-density perforated plastic box (LDPPB); and four natural edible coatings viz. control (no treatment), aloe vera @ 1%, garlic @ 1:1 and chitosan coating @ 0.2%, was conducted in completely randomized design with 3 replications. MAP and natural edible coatings were significant on all the parameters studied. Results revealed that combined treatment of LDPPB along with edible garlic coatings showed best external appearance among the treatments. The maximum weight loss (38.53%) and TSS content (8.23%) were recorded in without packaging plus control, while the minimum weight loss (21.17%) and TSS content (6.40%) were found with LDPPB plus garlic extract. Maximum disease incidence (90%) and severity (46.33%) was found in control fruits, while minimum disease incidence (30.0%) and severity (6.37%) was recorded in combined treatment of LDPPB and garlic coatings. The shortest shelf life was found from control treatment, whereas the longest shelf life (6.36 days) was obtained from combined treatment of LDPPB and garlic coatings. Therefore, combined treatment of LDPPB along with edible garlic coatings was found to be better in respect of significantly reducing postharvest fungal infection, shelf life prolongation and quality retention of strawberry.

Introduction

Strawberry (*Fragaria* × *ananassa* Duch.) is a very famous and highly appreciated fruit worldwide not only for its unique taste, distinct flavour, deliciousness and attractive colour, but also for its health benefits (Petriccione *et al*., 2015). The edible portion of the fruit is about 98% and contains various nutritionally important elements such as minerals and vitamins, and a diverse range of anthocyanins, flavonoids and phenolic acids with biological properties, such as antioxidant, anticancer and anti-inflammatory activities (Ayala-Zavala *et al*., 2004; Seeram *et al*., 2006). Strawberry is a highly perishable fruits with a very short shelf life because of high respiration rate (50-100 ml CO_2 per kg of fruits per hour at 20 °C) and relatively high postharvest losses (20-50%) due to infection by several pathogens during transport and storage, mechanical injury, physiological deterioration and water loss, which limit its popularity among the traders (Guerreiro *et al*., 2015; Nunes *et al*., 2006). About 20-50 per cent fruit loss occurs as post-harvest decay in strawberry depending upon harvesting month, fruit maturity, transportation distance and method of packaging (Mingchi and Kojimo, 2005).

Many preservation methods have been used to extend the shelf life and improve the quality of strawberry such as refrigeration, synthetic chemical fungicides, osmotic treatments, hypobaric treatments, heat treatment, controlled atmospheres and gamma irradiation (Bhat and Stamminger, 2015; Castello *et al*., 2010; Marina *et al*., 2015; Peerzada *et al*., 2012; Wang and Gao, 2013; Zhu and Zhou, 2007). However, consumers demand more natural, environmentally friendly food, with high quality and an extended shelf life, without any chemical preservatives (Lin and Zhao, 2007). Therefore, it is necessary to explore and utilize new techniques for maintaining postharvest quality of strawberry fruit.

Modified atmosphere packaging (MAP) using different films and boxes can be one of the best and low cost technology to have a better shelf life with proper quality for a soft fruit like strawberry, making the product more attractive to the retail customer (Panda *et al*., 2016). Modified atmosphere means an atmosphere composition around the fruit that is different from that of normal air i.e., 78.08% N2, 20.95% O2 and 0.03% CO2 (Kader, 1992). Such change in the gaseous atmosphere can be attributed to the factors like respiration and other biochemical processes of the produce and permeation of gases through the packaging film. It slows down the growth of aerobic microbes and speed of oxidation reactions. A well-known benefit of MAP is to reduce high water loss by creating high humidity inside the packaging and with that the produce maintains freshness comparatively for a longer period. However, good hygiene practices and temperature control throughout the chill-chain for perishable products are required to maintain the quality benefits and extended shelf life of MAP foods.

Edible coatings such as aloe vera, ethanolic extracts of garlic cloves, chitosan, etc. have been used as novel promising approach for the preservation and extending shelf life of fruits and vegetables (Neeta *et al*., 2013; Nur Fatima *et al*., 2018; Mondal *et al*., 2011; Sharmin *et al*., 2015; Zhang *et al*., 2016). Edible coating with semipermeable films can prolong the postharvest life and improve textural quality of strawberry fruits which helps retain volatile flavor compounds and reduce microbial growth through a reduction of moisture, gas exchange (oxygen and carbon dioxide), respiration and oxidative reaction (Lee *et al*., 2003; Pillai *et al*., 2009; Shiekh, 2013; Velickova, *et al*., 2013). Chitosan-based coatings are considered the best edible and biologically safe preservative coatings for different types of fruits, with functional advantages, such as slower respiration rates, extended storage periods, firmness retention and controlled microbial growth (Fan *et al*., 2009; Romanazzi, *et al*., 2015).

Strawberry is a quick growing and exotic fruit in Bangladesh and suitable for adaptation in our cropping pattern. Recently, strawberry production in Bangladesh has been increased, however, very few researches have been conducted on the combined application of modified atmosphere packaging (MAP) and natural edible coatings on the reduction of postharvest losses and quality retention capacity of strawberry. The present study was, therefore, undertaken to develop an appropriate safe technology using MAP and natural edible coatings to reduce postharvest fungal infection, shelf life prolongation and quality retention of strawberry cv. RU-1 (Festival).

Materials and Methods

Experimental location and material

The present study was conducted to study the effects of modified atmosphere packaging (MAP) and natural edible coatings on controlling postharvest fungal infection, shelf life extension and quality retention of strawberry cv. RU-1 (Festival) at the Laboratories of the Departments of Horticulture and Agricultural Chemistry, Bangladesh Agricultural University (BAU), Mymensingh during the period from February to April 2019. Strawberry fruits were collected from the Landscaping section of BAU. Medium sized fruits of strawberry cv. RU-1 (Festival) were harvested at approximately 75 per cent colour development stage in early morning hours (6-8 am). The collected strawberry fruits were uniform in shape, size, weight (average 10-12 g) and without visible imperfections or quality defects. About 150 g fruits were initially packed in plastic punnets with due care to minimize the chances of injury were kept under ambient temperature (25 ± 1°C) prior to further treatments.

Treatments of the investigation and experimental design

The two-factor experiment consisted of three MAP viz., (i) P0= Control (Without packaging), (ii) P1= Low-density perforated polyethylene (LDPPE) and (iii) P1= Low-density perforated plastic box (LDPPB); and four natural edible coatings viz., (i) T0= Control (No treatment), (ii) T1= Aloe vera extract @ 1%, (iii) T2= Garlic extract @ 1:1 and (iv) T3= Chitosan coating @ 0.2%. The experiment was conducted in a completely randomized design with 3 replications.

Application of the natural postharvest treatments

The postharvest treatments were sequentially applied to the selected strawberry fruits, which were dipped in each natural edible coating solutions for 2 minutes, air-dried and then kept under ambient temperature (25 ± 1°C) using different MAP. For control treatment ten fruits under each treatment were selected randomly from a strawberry fruit lot, washed with distilled water, air-dried. The fruits were kept at 40,000cc/m^2 day atm oxygen transmission rate (OTR) packaging film as a MAP with 85% relative humidity and stored under ambient temperature (25 ± 1°C) to measure the fruit quality and shelf life (Islam *et al*., 2017). For aloe vera coating (T1), the selected fruits were dipped in 1% aloe vera solution for 2 minutes. Aloe vera gel was extracted from fresh aloe vera leaves and gel solution was prepared as described by Sharmin *et al*. (2015). For garlic treatment (T2), initially stock garlic extract (1 kg garlic cloves and 1 L water) was prepared by crushing the fresh cloves in distilled water using a blender through straining and then cheesed. The stock extract was then used to prepare treatment of 1:1 concentration. For chitosan coating (T3) the selected strawberry fruits were dipped at 0.2% chitosan solution for 2 minutes and air-dried. Chitosan solution (0.2%) was prepared by dissolving 0.2 g of chitosan in 90 mL of distilled water added with 2 mL of glacial acetic acid. The mixture was heated with continuous stirring for proper dissolution of chitosan. The final pH of the solution was adjusted to 5.6 with 2 N NaOH and made up to 100 mL with distilled water. After application of all the treatments, fruits were wrapped with the respective MAP and kept at 25 ± 1°C and all changes of the fruits were monitored every day started from 0 to 4 days of storage.

Parameters studied

External appearance

Changes in external appearance of strawberry fruits were carefully observed, visually examined and recorded at every day starting from 1st day up to 4th day of storage through scoring using a colour chart.

Fruit firmness

Firmness of strawberry fruit was measured by Fruit Penetrometer (Model PX-145, Panomex Inc.). The Fruit Penetrometer accurately measures fruit hardness by measuring the force required to push a plunger tip (of a certain size) into strawberry fruit. The instrument was equipped with a 3.5 mm pressure head that had 10 mm insertion depth of pressure head.

Weight loss

Weight loss of strawberry was measured by weighing the fruits every day using a top pan electric balance. Ten fruits per treatment were taken for this purpose and same fruits were used until the end of the experiment. The percentage of weight loss was calculated by using the following formula:

$$\% \text{ Weight loss} = \frac{W_1 - W_2}{W_1} \times 100$$

Where, W1 = Initial weight of fruit (0 days)

W2 = Fruits weight at various storage periods (0, 1, 2, 3, and 4 days)

Fruit juice pH

The pH of fruit juice was measured by using a Portable pH Meter (Model pHS-1701, Shanghai, China), which was standardized with the help of a buffer solution as described by Ranganna (1994).

Total soluble solids (TSS)

Total soluble solids concentration of strawberry fruit was determined by using a hand refractometer (Model N-1 α, Atago, Japan). The remaining juice from pH determination was used to measure the TSS of the fruit juice. Before measurement, the refractometer was calibrated with distilled water to give a zero reading. One or two drops of the filtrate were placed on the prism of the refractometer to obtain %TSS reading. The reading was multiplied by dilution factor to obtain an original %TSS of the fruit tissues. Since differences in sample temperature could affect the TSS measurement, temperature corrections were made by using the methods described by Ranganna (1994).

Titratable acidity (TA) and ascorbic acid (Vitamin C) contents

Using the filtrate prepared to determine TSS, titratable acidity of strawberry juice was determined by titration against 0.1 N sodium hydroxide. Ascorbic acid (vitamin C) was measured by 2,6-dichlorophenol-indophenol titration as described by Ranganna (1994).

Determination of reducing sugar

Reducing sugar content of strawberry juice was determined by Dinitrosalicylic acid method (Miller, 1972).

$$\% \text{ Reducing sugar (g/100 g of sample)} = \frac{\text{Amount of sugar obtained}}{\text{Weight of sample}} \times 100$$

Estimation of non-reducing sugar

Non-reducing sugar content of strawberry juice was estimated by using the following formula:

$$\% \text{ Reducing sugar (g/100 g of sample)} = \frac{\text{Amount of sugar obtained}}{\text{Weight of sample}} \times 100$$

Disease incidence (percentage of infected fruits)

Ten fruits for each treatment were critically examined every day for the appearance of the disease symptoms and the incidence was recorded. The first count was made at the 1st day of storage. The disease development was identified by the visual quality, which was observed on the scale of 1 to 5 (1 = very bad, 2 = bad, 3 = good, marketable, 4 = very good, and 5 = excellent) (Islam *et al*., 2017). A total of three fungal diseases like Grey mould, Rhizopus soft rot and Leather rot were identified by observing the typical symptoms of those fungal diseases which were caused by Botrytis cinerea, Rhizopus stolonifer and Phytophthora cactorum, respectively (Mass, 20). Number of fungus-contaminated strawberry was counted and they were converted to fungal incidence percentage by the following formula:

$$\% \text{ Disease incidence} = \frac{\text{Number of infected fruits}}{\text{Total number of fruits assessed}} \times 100$$

Disease severity (percentage of skin infected fruits by fungal diseases)

In order to measure disease severity level, the strawberry fruits were critically observed and the percent skin infected fruits was recorded every day starting from the 1st day of storage up to the 4th day. All the infected fruits were taken to determine the percent fruit area infected and carefully evaluated. This evaluation was determined by centimeter scale by calculating the mean values regarding the infected fruit areas.

Shelf life

Shelf life of fruits means the days required for fully ripe as to retaining optimum marketing and eating qualities. In order to determine the shelf life, ten fruits were taken for each treatment and then the treated fruits were kept under ambient temperature (25 ± 1°C). Shelf life was measured according to visual quality (≥3; good, marketable) and determinants such as mold growth, decay, shriveling, smoothness, shininess, and homogeneity (Islam *et al*., 2017).

Statistical analysis

The collected data on various parameters were analysed statistically using MSTAT computer programme. The means for all the treatments were calculated and analysis of variance (ANOVA) was performed by F-test. The mean difference between a pair of treatments was tested by least significant difference (LSD) at 5 and 1% levels of probability.

Results and Discussions

External appearance

The change in external appearance was noticeable due to MAP and natural edible coating treatments (Figure 1a-e). Peano *et al*. (2014) reported that various edible coatings films are able to create MAP storage condition maintaining along all the storage time gas values different from the normal atmosphere composition (20.8% O2 and 0.03% CO2). The results showed that the external changes in fruits of control and single treatments were observed earlier than the combined treatments under various MAPs. In the control, protoplast was changed into chromoplast normally, while in treated samples, this process was suppressed by the treatment effect (Rashid *et al*., 2019). The changes of these components affected the fruit colour from slight red into deep red in the skin. The results revealed that combined treatment of LDPPB along with edible garlic coatings (P2T2) showed better external appearance than those of the other treatments.

Fruit firmness

Firmness is an important physical parameter used to assess the quality of fruits during ripeness, storage and distribution (Pasquariello *et al*., 2013). At harvest, strawberry cultivars showed different firmness values that could be due to different lignin content. Statistically significant variation was observed between the MAP and natural edible coatings in reference to strawberry fruit firmness (Table 1). Fruit firmness was decreased gradually with the storage period irrespective of different packaging and coating treatments, and at 4 DAS, LDPPB plus garlic coating (P2T2) followed by 3.62% in LDPPB plus aloe vera coating (P2T1) was most effective (3.94%) in maintaining the firmness of strawberry fruits, while the lowest fruit firmness (2.25%) was recorded under control (P0T0). The previous results reported the beneficial effects of several MAP and coating applications, and indicated that natural edible coating treatments significantly inhibited the softening of strawberry fruit resulting from the degradation of the middle lamella of the cell wall of cortical parenchyma cells (Perkins-Viaziel, 1995). At the end of the storage period, strawberry lost firmness, but LDPPB plus edible-coated fruits remained still much firmer than the control in which firmness preservation was the criteria used by the authors to explain the delay of ripening support this result, since red colour developed normally.

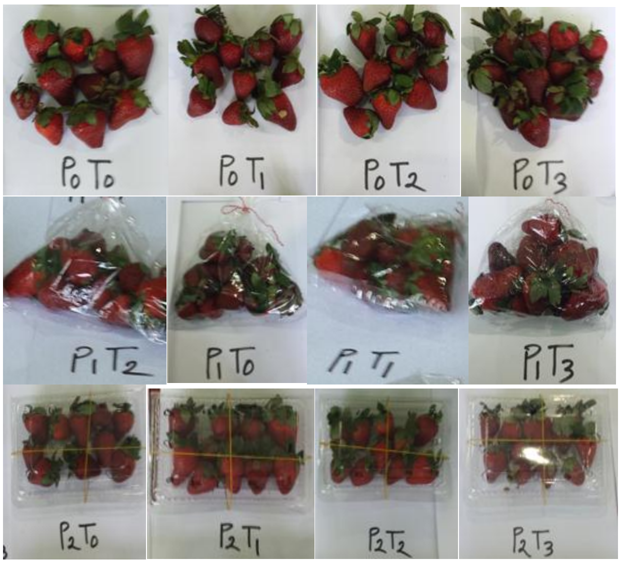

Figure 1a. Strawberry fruits at 0 DAS

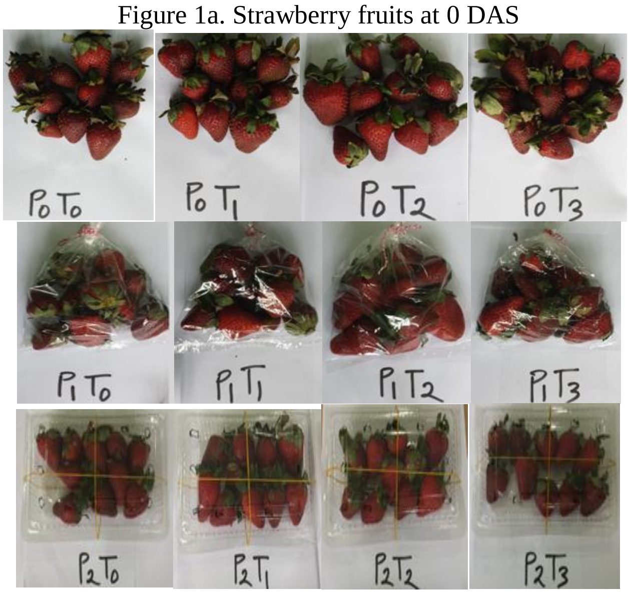

Figure 1b. Strawberry fruits at 1 DAS

Figure 1c. Strawberry fruits at 2 DAS

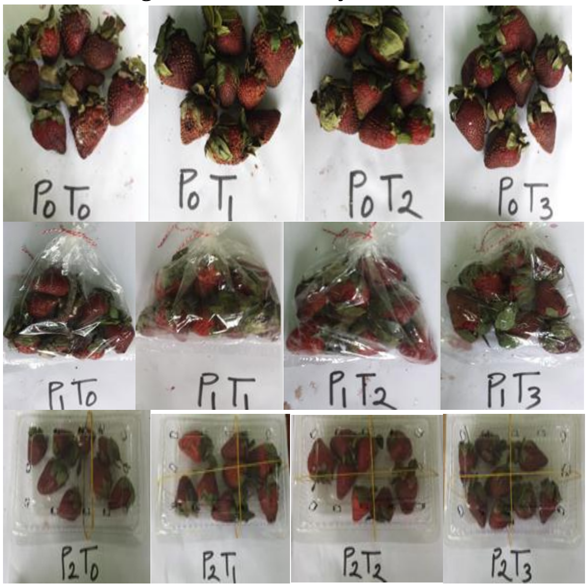

Figure 1d. Strawberry fruits at 3 DAS

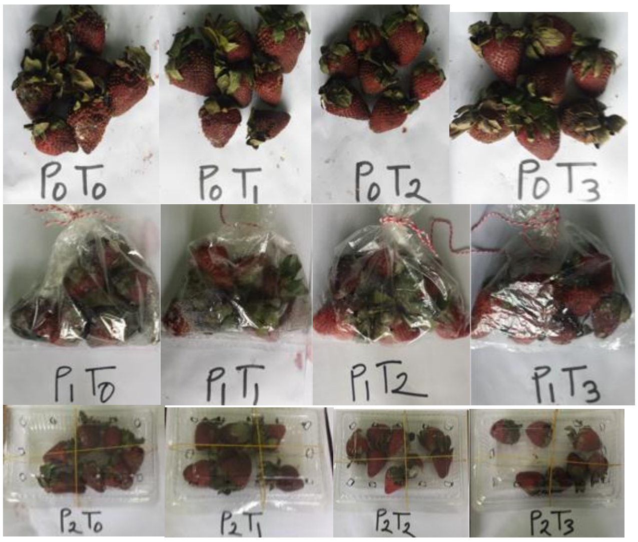

Figure 1e. Strawberry fruits at 4 DAS

Figure 1. Photograph showing the differences in external appearance of strawberry fruits at various days after storage (DAS) under different MAP and natural edible coating treatments. P_0= Control (Without packaging), P_1= Low-density perforated polyethylene (LDPPE), P_1= Low-density perforated plastic box (LDPPB), T_0= Control (No treatment), T_1= Aloe vera @ 1%, T_2= Garlic @ 1:1, and T_3= Chitosan coating @ 0.2%.

Table 1. Combined effects of MAP and natural edible coatings on weight loss and firmness of strawberry at different days after storage (DAS).

Treatment combination	Fruit firmness (%) at different DAS				Weight loss (%) at different DAS			
	1	2	3	4	1	2	3	4
P_0T_0	4.17	3.67	3.08	2.25	7.25	15.12	31.24	38.53
P_0T_1	4.56	4.11	3.52	2.91	5.35	13.68	24.50	33.52
P_0T_2	4.72	3.65	3.84	3.12	5.23	12.26	22.47	30.00
P_0T_3	4.37	4.10	3.68	2.96	5.59	12.30	23.17	31.61
P_1T_0	4.17	3.79	3.20	2.74	6.81	13.61	29.44	34.52
P_1T_1	4.90	4.37	3.81	3.40	5.22	13.10	25.18	30.61
P_1T_2	5.14	4.66	4.28	3.73	5.14	12.16	20.54	25.85
P_1T_3	4.93	4.47	4.04	3.56	5.47	12.42	21.50	27.35
P_2T_0	4.17	3.82	3.22	2.56	6.20	10.18	19.42	31.49
P_2T_1	5.32	4.84	4.26	3.62	5.12	9.89	23.84	26.27
P_2T_2	5.74	5.32	4.74	3.94	4.35	9.18	16.20	21.17
P_2T_3	4.88	4.43	3.89	3.40	4.32	9.44	19.06	23.53
$LSD_{0.05}$	0.160	0.341	0.151	0.199	0.266	0.399	1.086	0.843
$LSD_{0.01}$	0.217	0.462	0.204	0.270	0.361	0.540	1.471	1.142
Level of significance	**	**	**	**	**	**	**	*

**, *= Significant at 1 and 5% levels of probability, respectively, P_0= Control (Without packaging), P_1= Low-density perforated polyethylene (LDPPE), P_2= Low-density perforated plastic box (LDPPB), T_0= Control (No treatment), T_1= Aloe vera @ 1%, T_2= Garlic @ 1:1, and T_3= Chitosan coating @ 0.2%.

Weight loss

Weight loss in fresh fruits is mainly attributed to the loss of water caused by transpiration and respiration processes and is a major cause of quality deterioration (Hernández-Muñoz, *et al*., 2006). All strawberries showed a gradual loss of weight during storage and it was significantly influenced by combined effect of MAP and natural edible coating treatments (Table 1). Maximum weight loss was recorded with without packaging plus control in P_0T_0 (38.53%) followed by LDPPE plus control in P_1T_0 (34.52%), while minimum weight loss was found with LDPPB plus garlic extract in P_2T_2 (21.17%) followed by LDPPB plus chitosan coating P_2T_3 (23.53%). This might be due to the packaging films alter the CO_2 and O_2 concentration inside the packages hence, in a high respiring fruit like strawberry the respiration rate is reduced by keeping in low O_2 and/or high CO_2 atmosphere (Li and Kader, 1989). Panda *et al*. (2016) reported the fruits wrapped in different packaging films retain better quality for longer duration compared to the unwrapped fruits. Strawberry fruits are highly susceptible to a rapid loss of water due to the extremely thin skins of these fruits. These results are also consistent with those of previous studies demonstrating that chitosan coating acts as a semipermeable barrier against oxygen, carbon dioxide and moisture, thereby reducing respiration and water loss and counteracting the dehydration and shrinkage of the fruit (Veliockova *et al*., 2013).

Fruit juice pH

Combined effects of MAP and natural edible coating treatments exhibited significant variation in respect of juice pH during storage of strawberry. Results showed that the pH of strawberries decreased during storage and at 4 DAS, the highest juice pH was observed with no packaging plus chitosan coating in P_0T_3 (5.17) followed by without packaging plus garlic extract in P_0T_2 (4.27) (Table 2). In contrast, the lowest juice pH was recorded from LDPPB plus control in P_2T_0 (3.33) followed by without packaging plus control in P_0T_0 (3.57). The result was still above the average reported values for ripe strawberry of 3.3 (Elena *et al*., 2013).

Table 2. Combined effects of MAP and natural edible coatings on juice pH and TSS content of strawberry at different days after storage (DAS).

Treatment combination	Fruit juice pH at different DAS				TSS content (% brix) at different DAS			
	1	2	3	4	1	2	3	4
P_0T_0	6.83	5.73	4.43	3.57	3.67	4.20	6.40	8.23
P_0T_1	6.53	6.23	5.30	4.27	3.63	4.40	6.27	7.47
P_0T_2	6.70	6.17	5.13	4.27	2.27	3.97	5.87	7.03
P_0T_3	6.67	6.43	5.70	5.17	2.53	3.20	5.83	7.70
P_1T_0	6.90	6.27	4.77	3.53	3.80	5.10	6.77	8.13
P_1T_1	6.77	6.50	5.70	4.20	3.20	4.27	6.20	7.73
P_1T_2	6.70	6.43	4.17	3.70	3.23	4.23	6.17	7.20
P_1T_3	6.70	6.37	4.80	4.33	3.73	4.40	6.13	7.30
P_2T_0	6.83	5.63	4.10	3.33	3.40	4.20	6.17	7.87
P_2T_1	6.73	5.70	4.20	3.87	3.80	4.90	6.67	7.67
P_2T_2	6.73	6.10	4.37	4.10	2.43	3.13	5.37	6.40
P_2T_3	6.77	6.27	5.20	4.23	3.20	4.43	6.20	7.30
$LSD_{0.05}$	0.092	0.226	0.213	0.185	0.443	0.584	0.282	0.436
$LSD_{0.01}$	0.125	0.306	0.289	0.250	0.600	0.791	0.382	0.591
Level of significance	**	**	**	**	**	**	**	*

**, *= Significant at 1 and 5% levels of probability, respectively, P_0= Control (Without packaging), P_1= Low-density perforated polyethylene (LDPPE), P_2= Low-density perforated plastic box (LDPPB), T_0= Control (No treatment), T_1= Aloe vera @ 1%, T_2= Garlic @ 1:1, and T_3= Chitosan coating @ 0.2%.

Total soluble solids (TSS) content

The TSS of strawberry fruits packed in different packaging materials and coated with various natural edible extracts exhibited significant variation (Table 2). It was observed that TSS content in fruit juice gradually increased during storage period irrespective of all treatments, which was similar to the investigation of Panda *et al*. (2016) and Petriccione *et al*. (2015) where the authors reported that uncoated and no packaging fruits exhibited significantly ($p < 0.05$) higher TSS compared to all other edible-coated fruits. At 4 DAS, the highest TSS (8.23%) was observed in the treatment combination of without packaging plus control in P_0T_0 followed by LDPPE plus control in P_1T_0 (8.13%), whereas it was the lowest with LDPPB plus garlic extract in P_2T_2 (6.40%) followed by without packaging plus garlic extract in P_0T_2 (7.03%). Similarly, Magazine *et al*. (2015) and Li and Kader (1989) also reported, strawberry fruits packed in LDPE in room storage retain the TSS percent compared to the other packaging material. Strawberries are considered mature with approximately 7% of soluble solids (Kader, 1999). The increase in TSS values probably not due to conversion of starch to sugars, since strawberries accumulate very little starch, but due to solubilization of cell wall pectin as showed by the increases in anthocyanin and results are consistent with those of other studies concerning the effects of chitosan-coated treatment on mango, guava, banana, papaya, guava and sweet cherry (Ali *et al*., 2011; Hong *et al*., 2012; Kittur *et al*., 2001; Petriccione *et al*., 2015).

Titratable acidity (TA) content and ascorbic acid (vitamin C) content

The TA estimates the organic acid contents of fleshy fruits, and in strawberry fruit, the main organic acids are citric and malic acid (Kallio *et al*., 2000). This trait is an important component of fruit organoleptic quality and is different in each cultivar. Results revealed that titratable acidity of strawberry fruits packed in different MAPs went on decreasing with the advancement of storage period (Table 3). Significant variation in TA of fruits packed with different MAPs and treated with various natural coatings was found. At the end of storage (4 DAS) the highest TA (4.90%) was recorded from without MAP plus aloe vera extract in P_0T_1 which was statistically identical with no MAP plus garlic extract (4.70%) in P_0T_2, while the lowest TA (2.63%) was observed from LDPPE plus aloe vera extract in P_1T_1 followed by without MAP plus chitosan coating (3.02%) in P_0T_3. Such reduction in acidity might be due to the utilisation of different free acids present in the vacuole of cells during various metabolic processes like respiration and anthocyanin biosynthesis (Panda *et al*., 2016). Previous studies have suggested that the higher acidity loss in uncoated fruits might reflect the use of organic acids as substrates for respiratory metabolism during storage (Diaz-Mula *et al*., 2012; Diaz-Mula *et al*., 2009). Chitosan treatment plays an important role in delaying fruit ripening during cold storage, and chitosan-coated fruits showed a lower acidity loss, consistent with other studies on strawberry, peach, guava and litchi (Hernandez-Munoz *et al*., 2008; Li and Yu, 2001; Dong *et al*., 2004).

The combined treatment of MAP and natural edible coating treatments showed significant influence on vitamin C content of strawberry (Table 3). At 4 DAS the highest vitamin C (126.13 mg/100g) was observed in combined treatment of LDPPB plus garlic coated fruits (P_2T_2) followed by 120.10 mg/100g in LDPPE plus garlic coated fruits (P_1T_2), while the lowest vitamin C (83.97 mg/100g) was recorded in without MAP plus uncoated fruits (P_0T_0) followed by 41.40 mg/100g in LDPPE plus uncoated control (M_2T_0). Such results might be attributed to better modification of the atmosphere inside the packages by these materials with respect to the O_2 concentration and concomitant decrease in enzymatic oxidation of ascorbic acid (Agrahari *et al*., 2001). With the advancement of storage periods, the ascorbic acid content was decreased significantly, which might be due to the oxidation and irreversible conversion of ascorbic acid to dehydroascorbic acid in the presence of enzyme ascorbinase. Similar decreasing trend in ascorbic acid content was also obtained by Kirad *et al*. (2007), where the authors reported that polysaccharide and chitosan-coated fruits possessed relatively higher ascorbic acid contents than control which might reflect low oxygen permeability, which reduced the activities of enzymes involved in the oxidation of ascorbic acid.

Table 3. Combined effects of MAP and natural edible coatings on titratable acidity and vitamin C content of strawberry at different days after storage (DAS).

Treatment combination	Titratable acidity (%) at different DAS				Vitamin C (mg/100 g) at different DAS			
	1	2	3	4	1	2	3	4
P_0T_0	11.25	9.34	6.18	4.05	177.33	157.80	128.90	83.97
P_0T_1	9.44	7.24	6.70	4.90	190.33	180.77	140.30	101.27
P_0T_2	8.56	7.08	6.50	4.70	203.30	189.80	147.60	109.80
P_0T_3	10.00	8.55	5.51	3.02	184.67	172.43	138.10	97.67
P_1T_0	10.91	9.10	6.07	3.70	184.23	162.73	130.63	98.37
P_1T_1	8.87	7.20	4.97	2.63	195.60	181.63	142.30	109.20
P_1T_2	8.10	6.93	4.70	3.70	215.27	196.40	153.03	120.10
P_1T_3	9.49	8.12	6.65	3.86	200.60	177.46	143.03	103.43
P_2T_0	10.31	8.69	7.24	4.25	195.20	172.73	137.67	106.37
P_2T_1	8.53	6.90	4.90	3.80	208.17	187.33	151.17	119.03
P_2T_2	7.75	6.48	4.48	3.20	223.07	203.20	165.50	126.13
P_2T_3	9.03	7.67	5.78	3.62	203.07	183.17	143.03	113.43
$LSD_{0.05}$	0.075	0.092	0.388	0.250	4.89	1.91	4.94	1.63
$LSD_{0.01}$	0.102	0.125	0.526	0.339	6.62	2.59	6.70	2.20
Level of significance	**	**	**	**	*	**	**	**

**, *= Significant at 1 and 5% levels of probability, respectively, P_0= Control (Without packaging), P_1= Low-density perforated polyethylene (LDPPE), P_2= Low-density perforated plastic box (LDPPB), T_0= Control (No treatment), T_1= Aloe vera @ 1%, T_2= Garlic @ 1:1, and T_3= Chitosan coating @ 0.2%.

Reducing and non-reducing sugar contents

Glucose, fructose and sucrose represent the main soluble metabolites in the fruits (Makinen and Soderling, 1980). Combined effect of MAP and natural edible coating treatments showed significant result in terms of both reducing sugar content of strawberry fruits. Reducing sugar content was increased gradually during the storage period (Table 4). At the end of the storage period (4 DAS) the maximum reducing sugar (4.27%) was found from without MAP plus aloe vera coated fruits (P_0T_1) followed by 4.13% in LDPPE plus garlic extracts, while the minimum reducing sugar (3.50%) was recorded in LDPPB plus garlic coated fruits (P_2T_2) followed by 3.51% in without MAP plus chitosan coated fruits (P_0T_3). Similar result was also reported by Peano *et al*. (2014). It was also found the significant influence of MAP and natural edible coatings on non-reducing sugar content of strawberry fruits (Table 4). The highest non-reducing sugar (2.70%) was observed in the combination of LDPPB plus garlic coating (P_2T_2), while the lowest non-reducing sugar (2.15%) was recorded from the combination of LDPPE plus garlic coated fruits ((P_1T_2). The result showed that non-reducing sugar content of strawberry fruits slightly increased with ripening. The non-reducing sugar content, remain more or less constant, after attaining peak, it was increased rapidly along with reducing sugar content during the first 1-2 days of storage and then leveled off as reported by Joshi and Roy (1988).

Table 4. Combined effects of MAP and natural edible coatings on reducing and non-reducing sugar content of strawberry at different days after storage (DAS).

Treatment combination	Reducing sugar (%) at different DAS				Non-reducing sugar (%) at different DAS			
	1	2	3	4	1	2	3	4
P_0T_0	3.30	3.36	3.42	3.54	2.19	2.21	2.25	2.31
P_0T_1	3.25	3.38	3.42	4.27	2.15	2.18	2.20	2.23
P_0T_2	3.29	3.42	3.46	3.52	2.15	2.17	2.20	2.22
P_0T_3	3.28	3.37	3.40	3.51	2.12	2.15	2.17	2.18
P_1T_0	3.41	3.46	4.18	3.62	2.14	2.16	2.18	2.20
P_1T_1	3.54	3.62	3.71	3.87	2.07	2.11	2.14	2.16
P_1T_2	3.51	3.65	4.00	4.13	2.12	2.13	2.14	2.15
P_1T_3	3.43	3.52	3.61	3.71	2.10	2.13	2.15	2.16
P_2T_0	3.41	3.47	3.53	3.62	2.11	2.14	2.15	2.18
P_2T_1	3.37	3.42	3.50	3.54	2.06	2.10	2.12	2.18
P_2T_2	3.24	3.30	3.38	3.50	2.25	2.37	2.54	2.70
P_2T_3	3.38	3.44	3.53	3.60	2.12	2.14	2.16	2.18
$LSD_{0.05}$	0.053	0.053	0.107	0.169	0.053	0.053	0.092	0.107
$LSD_{0.01}$	0.072	0.072	0.144	0.228	0.072	0.072	0.125	0.144
Level of significance	**	**	**	**	**	**	**	**

**, *= Significant at 1 and 5% levels of probability, respectively, P_0= Control (Without packaging), P_1= Low-density perforated polyethylene (LDPPE), P_2= Low-density perforated plastic box (LDPPB), T_0= Control (No treatment), T_1= Aloe vera @ 1%, T_2= Garlic @ 1:1, and T_3= Chitosan coating @ 0.2%.

Disease incidence and severity

Strawberry postharvest diseases were observed from the 1st DAS and it increased with the progress of storage period. During storage period, a total of three fungal diseases like Leather rot, Grey mould and Rhizopus soft rot were identified by observing the typical symptoms of those fungal diseases which were caused by *Phytophthora cactorum, Botrytis cinerea* and *Rhizopus stolonifer*, respectively (Figure 2).

a b c

Figure 2. Photograph showing (a) Leathery rot (*Phytophthora cactorum*), (b) Grey Mould (*Botrytis cinerea*) and (c) Rhizopus soft rot (*Rhizopus stolonifer*) of strawberry in control treatment.

Assesment of percent disease incidence

The combined effect of MAP and natural edible coating treatments had significant influence on disease incidence of strawberry (Table 5). The disease incidence increased with the storage period and at 4 DAS the highest disease incidence (90%) was found in control (P_0T_0) and the lowest (30%) was recorded in LDPPB plus garlic coating (P_2T_2). This might be due to the synergistic effects of combined treatments, which could give less detrimental influence to quality attributes of the strawberry fruits.

Table 5. Combined effects of MAP and natural edible coatings on disease incidence and severity of strawberry at different days after storage (DAS).

Treatment combination	Disease incidence (%) at different DAS				Disease severity (%) at different DAS			
	1	2	3	4	1	2	3	4
P_0T_0	1.67	36.70	63.30	90.00	3.23	10.00	21.73	46.33
P_0T_1	0.00	20.00	33.30	50.00	0.00	5.40	16.00	26.63
P_0T_2	0.00	6.70	23.30	36.70	0.00	1.10	5.50	10.00
P_0T_3	0.00	20.00	30.00	43.30	0.00	4.77	15.83	19.07
P_1T_0	1.33	30.00	50.00	70.00	2.17	8.77	16.80	36.20
P_1T_1	0.00	13.300	33.30	56.70	0.00	5.23	16.27	20.50
P_1T_2	0.00	13.30	26.70	40.00	0.00	2.77	5.30	7.67
P_1T_3	0.00	16.70	23.30	30.00	0.00	4.73	14.73	18.13
P_2T_0	0.00	20.00	46.70	70.00	0.00	6.43	16.90	31.00
P_2T_1	0.00	20.00	33.30	43.30	0.00	4.10	11.83	9.03
P_2T_2	0.00	6.70	20.00	30.00	0.00	3.10	5.17	6.37
P_2T_3	0.00	16.70	26.70	40.00	0.00	3.77	4.90	7.63
$LSD_{0.05}$	0.169	1.341	2.111	4.416	0.141	0.920	3.134	1.513
$LSD_{0.01}$	0.228	1.817	2.861	5.985	0.191	1.247	4.247	2.050
Level of significance	**	**	**	**	**	**	**	**

**, *= Significant at 1 and 5% levels of probability, respectively, P_0= Control (Without packaging), P_1= Low-density perforated polyethylene (LDPPE), P_2= Low-density perforated plastic box (LDPPB), T_0= Control (No treatment), T_1= Aloe vera @ 1%, T_2= Garlic @ 1:1, and T_3= Chitosan coating @ 0.2%.

The combined effect of MAP and natural edible coating treatments had also significant influence on disease severity of strawberry fruits (Table 5). Like disease incidence, the disease development also increased with the storage period and the highest disease severity (46.33%) was found at the end of the storage period (4 DAS) under control packaging and uncoated fruits (P_0T_0) followed by 36.20% in LDPPE plus uncoated strawberry fruits (P_1T_0) and the lowest disease severity (6.37%%) was observed from LDPPB plus garlic extract (P_2T_2) followed by (7.63%) in LDPPB plus chitosan coated fruits (P_2T_3). This might be due to the effects of ethanolic extract and fresh garlic cloves and chitosan coating contain biologically natural fungicide substances which are potentially used for the control of many fungal diseases of fruits (Dong *et al*., 2004; Hernandez-Munoz *et al*., 2008; Li and Yu, 2001; Mondal *et al*., 2011; Nur Fatima *et al*., 2018).

Shelf life

Shelf life is the period from harvesting up to the last edible stage. This is the basic quality of fruits, which helps marketing duration, and it is the most important aspect in loss reduction technology of fruits (Mondal *et al*., 2011; Rashid *et al*., 2015). The extension of shelf life of fruit has been one of the prime concerns of marketing throughout the record of history. The combined effect of MAP and natural edible coatings had significant influence on shelf life of strawberry (Figure 3). The longest shelf life (6.36 days) was obtained in LDPPB plus gralic coated fruits (P_22_4) and the shortest shelf life (2.33 days) was recorded in control (P_0T_0). The increase in shelf life was probably due to the changes in the concentrations of various gases like the increase of O_2, the reduction of CO_2 and ethylene as well as the slowing down of the processes leading to delayed ripening and reducing decay by LDPPB and garlic extract treatments. The delay in ripening on MAP and coated fruits can occur due to the lower capacity of these fruits in producing ethylene, since this hormone has a stimulation role in the general metabolism, and seems to be implicated in the activation and regulation of some enzymes involved in ripening (Gomez *et al.*, 1999).

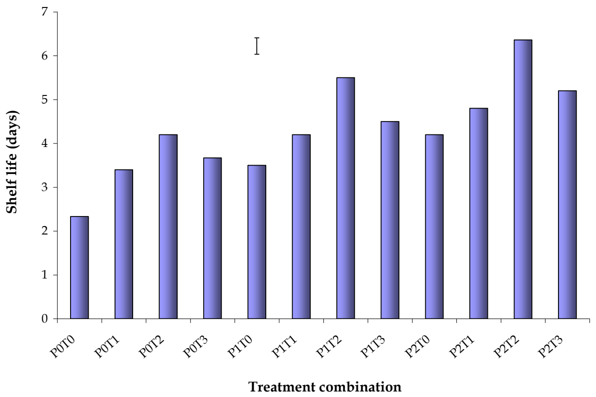

Figure 3. Combined effects of MAP and natural edible coatings on shelf life of strawberry fruits. The vertical bar represents LSD at 1% level of probability. P_0= Control (Without packaging), P_1= Low-density perforated polyethylene (LDPPE), P_1= Low-density perforated plastic box (LDPPB), T_0= Control (No treatment), T_1= Aloe vera @ 1%, T_2= Garlic @ 1:1, and T_3= Chitosan coating @ 0.2%.

Conclusion

From the present study it was found that strawberry is a highly perishable fruit, which cannot be stored for a longer period but different wrapping treatments along with natural edible coatings maintained the qualitative characteristics of stored fruits at ambient temperature condition. From the day, first onwards the loss in weight of fruits was observed but LDPPB packaging material along with garlic coating proved as the most effective one to control the weight loss. MAP conditions along with edible coating prevented decaying of strawberry fruits up to single day under the ambient condition. The fruit firmness, pH, titratable acidity and ascorbic acid contents ratings decreased in the stored fruit at ambient temperature and spoiled completely after 4th day of storage under without MAP and uncoated fruits. Therefore, it may be concluded that the LDPPB plus garlic extract was found to be the best treatment, which significantly reduce postharvest fungal infection, extend shelf life and retain quality of strawberry fruits.

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