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Impact of Natural Polysaccharides Based Edible Coatings on Postharvest Physiology and Bioburden of Lycopersicon Esculentum

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ABSTRACT: *Edible coatings are a promising postharvest quality management treatment for shelf-life enhancement and marketability of the agriculture products. This study aims to evaluate the effects of polysaccharide based edible coatings on the mean life of un-ripened tomatoes (Lycopersicon esculentum). Fruits were treated with three different edible coatings Aloe vera gel (T1), tragacanth gum (T2) and Aloe vera gel-tragacanth gum (T3) for postharvest preservation and the mean life was analyzed at 20°C. They were then examined for various physiological, physiochemical and microbiological changes. The results revealed that all the applied formulations could retard the metabolic processes involved in tomato ripening. However, the combined effects of Aloe vera gel and tragacanth gum (T3) were most successful in positively maintaining the tomatoes since the fruits in this category recorded comparatively superior results for most of the analyzed parameters. They reached full maturity in 45 days compared to the control set which ripened in 14 days. This treatment led to a substantial postponement in weight loss, disease incidence, microbial growth and ripening index of tomatoes. A significant difference was observed in rate of color change among treated and untreated samples ($P < 0.05$). For decay assessment, Uncoated samples decayed after 4 weeks while coated samples (T-3 tomatoes) remained healthy for 60 days. For weight loss assessment, the samples receiving treatments T1, T2, and T3 showed 16.23%, 14.45%, and 10.23 % weight loss at 20°C respectively. Thus, storage life and safety of tomatoes can be extended using Aloe vera-tragacanth gum based edible coating which has the potential to be used as natural preservative.*

Keywords: *Tomatoes; Aloe vera; Tragacanth gum; Shelf Life; Edible coating; Postharvest*

INTRODUCTION

Tomatoes are quite rich in minerals, vitamins, vital amino acids, sugars and dietary fibers. They harbor ample amount of lycopene (71.6%), vitamin C (12.0%), pro vitamin A carotenoids (14.6%) and vitamin E (6.0%) (García-Closas et al., 2004). Lycopene, primarily accountable for the distinctive deep red color, holds strong antioxidant properties hence able to reduce the risk of certain human cancers related to prostate, lung and stomach and chronic ailments such as cardiovascular disease (Heber, 2004). As per World Health Organization (WHO) estimates, lower consumption of fruits and vegetables is among the highest risk factors contributing to the mortality rates of 2.7 million deaths each year (Hartley et al., 2013). Preservation of vegetables and fruits is a great challenge for the world. According to rough estimates about 30-40% of vegetables/fruits are wasted due to improper and deficient processing facilities (Tiwari and Cummins, 2013). Being a climacteric fruit, respiration and transpiration primarily determine its storage life and quality of tomatoes. However, effects of these processes can be curtailed using a physical barrier for oxygen diffusion and moisture migration. Storage at low temperatures is the traditional strategy used to postpone and/or decrease ethylene synthesis, yet this approach may result in chilling harm (Singh and Shalini, 2016). In order to lower pathogen levels, pesticides and sanitizers are frequently used; however, their application can leave residues that could beyond the maximum permissible limits, which could be extremely harmful to human health (Singh and Shalini, 2016).

Recent studies have concentrated on investigating better and more effective postharvest processing and preservation methods to overcome these issues. Applying edible coatings has been a popular substitute among these methods for prolonging the postharvest shelf life of tomato fruit (Yaashikaa et al., 2023).

Edible coatings are a promising postharvest quality management treatment for marketability of the agriculture produce. Minimization of post-harvest losses would reduce the production cost, trade and distribution, increasing the farmer’s income and lowering the price for the consumers (Kiaya, 2014). Moreover, , two fungi that can colonize damaged fruits during harvesting and handling and quickly spread to neighboring fruits, causing large losses, are capable of attacking tomatoes (Tahmasebi et al., 2020*)*. When tainted tomatoes are consumed, bacterial illnesses, such as *Escherichia coli*, can also be harmful to human health. Currently, edible coatings are becoming a popular treatment when it comes to keeping intact the quality factors of fruits (Galgano, 2015). They provide a semi-permeable barricade for carbon dioxide and oxygen transfer, moisture and the movement of solute, thereby plummeting respiration, water loss and oxidation. This supports in regulating the quality control of the fruits (Mahfoudhi and Hamdi, 2015). The leeway in postharvest life of the fruits and vegetables and their quality critically depends upon reduction in desiccation, decline in physiological processes of maturation and senescence and slower rates of microbial growth (Rico et al., 2007). In addition, another important benefit is the decreased synthetic packaging waste since the coatings are made up of biodegradable raw material. The potential of *Aloe vera* and tragacanth based edible coatings in food preservation has been highlighted by recent studies. et al. Tarangini (Hadi et al., 2022, Mohebbi et al., 2012, Mohebbi et al., 2014).

In this study, three different edible coating viz., *Aloe gel*, tragacanth gum and *Aloe Vera* gel/tragacanth gum-based formulations have been used with the aim to enhance the shelf life of tomatoes with preserving the organoleptic quality.

MATERIALS AND METHODS

Sample Collection

Fresh tomato fruits (*Lycopersicum esculentum*) harvested at mature green stage were taken from a commercial market in Lahore, Pakistan. The fruits were selected based upon their size, weight, color, and physical appearance to maintain the uniformity in the sampling.

Preparation of the natural extracts

Aloe vera (*Aloe barbadensis*) leaves were washed with chlorine water (25%) and colorless hydro parenchyma was taken out. The excised gel matrix was grinded, and the solution was filtered to remove fibers. The extract was pasteurized and stored at ambient temperature. The tragacanth gum

(*Astragalus gummifer*) crystals were pulverized and sieved to get fine powder. 0.5% gum solution was prepared in distilled water and heated at 40°C for 10 mins and kept overnight under ambient conditions to fully hydrate. Next day, it was pasteurized and stored in an amber bottle.

Edible coating formulations and treatment

After extracts preparation, three different types of coating solutions were formulated as T1 (20 ml of *Aloe vera* gel extract, 0.5 g of CaCl2, 5 ml of lemon juice), T2 (20 ml of Tragacanth gum extract, 0.5 g of CaCl2, 5 ml of lemon juice) and T3 (10 ml of *Aloe vera* gel extract, 10 ml of tragacanth gum extract, 0.5 g of CaCl2, 5 ml of lemon juice). 1% agar was added to each of the above-mentioned coating formulations which were then heated slightly to dissolve it.

The experimental group of tomatoes were first dipped in the neem water followed by immersion in each edible coating formulation for 5 mins, air dried, packaged into zip locked polyethylene bags and stored at 20°C.

Parameters and Analysis

Evaluation was carried out of microbiological, physiochemical and sensory attributes. Tomatoes were observed for 3 days interval for their organoleptic qualities and 14 days interval for microbial load till 8 weeks.

Physiological studies

Physiological factors such as fruit color, firmness, weight loss and decay percentages were explored as an index of fruit quality during storage post treatments. The variations in the skin color were logged by comparing with a standard color chart (Royal Horticultural Society color chart).

Weight loss was determined as the percentage loss of the initial fresh weight using the method of Athmaselvi et al. (2013). Firmness was estimated through the reduction in girth size of coated and uncoated tomatoes. A measuring tape was used to measure the same at weekly intervals (Goyal et al., 2017). The decay incidence was recorded based on visual inspection of each fruit for infection and their percent decay using (Abdullah et al., 2017).

Physiochemical studies

Physiochemical changes in pH, titratable acidity and reducing sugars were examined to study the progression of ripening. The pH of the juice was measured after every 7 days via pH meter. Titratable acidity was estimated by titrating 5 ml of tomato juice in 10 ml distilled water against 0.1 N NaOH (Abebe et al., 2017). The amount of reducing sugar present in tomatoes was determined by DNS (3,5 Dinitrosalicylic acid) method (Miller, 1959).

Microbiological Studies

A serial dilution method was used for microbiological studies. The total viable microbial count was observed on Nutrient agar, MacConkey agar and Potato Dextrose agar.

Statistical analysis

The results were validated by Costat 6.4 using a completely randomized block design. The means were compared using ANOVA and Duncan's New Multiple Range test at $p \leq 0.05$ with three replicates.

RESULTS AND DISCUSSION

Physiological Parameters

Color and physical transformations of tomatoes during storage

Color is an important index of fruit ripening and consumer acceptability. The tomato fruit undergoes color transitions due to chlorophyll degradation and formation of carotenoids (Abebe et al., 2017). In this study, mature green tomatoes turned tarnished yellow to red in color indicating the progression of ripening (Fig. 1a). A significant difference was observed in rate of color change among treated and untreated samples ($P < 0.05$). Control group changed color more rapidly than the experimental group and reached the 6th stage of maturity within 20 days at 20°C. *Aloe vera* gel (T1) coated fruits ripened within 35 days in storage while tragacanth gel (T2) coated samples turned red (6th stage) after 40 days. However, the combination of both gel formulations (T3) retarded color and physiological changes at maximum since the fruits receiving this treatment ripened in 45 days. Investigations carried out by Bhatnagar (2018) also indicated retardation in color development of tomatoes treated with *Aloe vera* gel based coatings as per the slow rate of respiration and decreased ethylene production. The altered atmosphere in the fruit formed by the edible coatings delimited the respiration rate deferring color variation. Preeminent CO_2 levels (>1%) in fruit tissues attained by coating materials may also slow down fruit ripening by constraining ethylene production (Zapata et al., 2008).

Fruit firmness also dictates the postharvest life and fruit quality. The degradation of cell wall components may render the fruit susceptible to different postharvest handling factors. The loss in width diameter was taken as an index of decrease in fruit firmness during ripening (Fig. 1b).

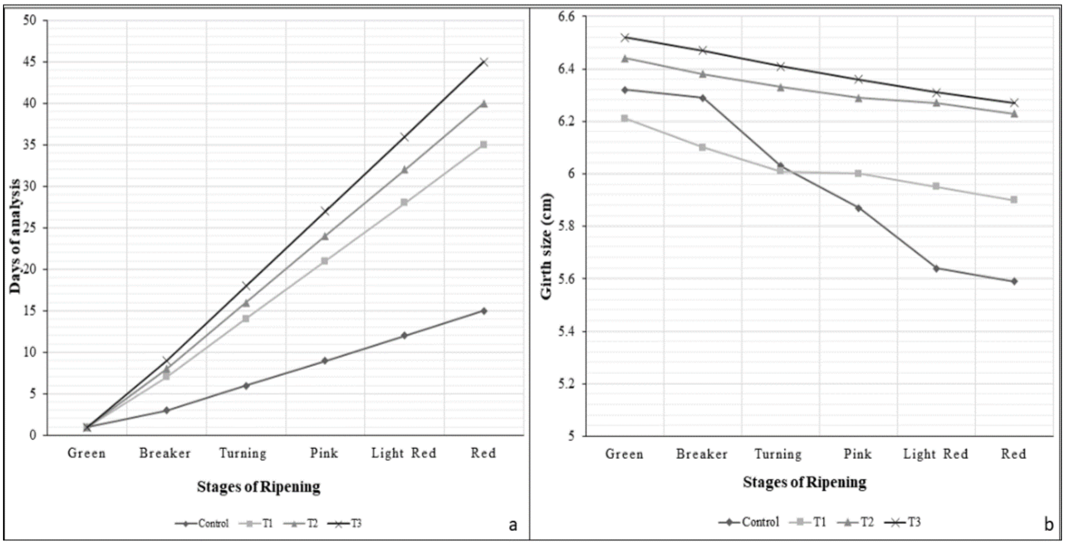

Fig. 1. Comparative analysis of ripening behavior and fruit firmness of Tomatoes following edible coating treatments (a) Progression of ripening in treated and control tomatoes (b) Changes in the girth size of treated and control tomatoes

The coated tomatoes were significantly firm for a longer period than the controls ($P < 0.05$) which indicated a 12% loss in width diameter till mature red stage. In contrast, the treated samples sustained high degree of firmness with only 4 to 5% decrease in width diameter until red stage. The decrease in firmness results from the breakdown of pectin and starch present in the cell wall by enzymes such as hydrolases, pectin esterase, and polygalacturonase produced during ripening (Jahanshahi et al., 2018). The above findings are in line with the previous studies in which *Aloe vera* gel and tragacanth gum based edible coatings were able to cause firmness retention in cherries, grapes and strawberries

(Emamifar and Bavaisi, 2017, Martínez-Romero et al., 2006).

Decay Assessment

Both coated and uncoated samples showed zero decay percentage up to 18 days (Table 1).

Table 1. Effect of edible coatings on the spoilage rate of tomatoes stored at 20^oC

Days of storage	Percentage of spoilage (%)			
	Control	T1	T2	T3
1	0	0	0	0
5	0	0	0	0
10	0	0	0	0
15	0	0	0	0
20	0	0	0	0
25	65.3	0	0	0
30	D	0	0	0
35	-	0	0	0
40	-	16.5	0	0
45	-	67.8	0	0
50	-	D	0	0
55	-	-	14.8	0
60	-	-	57.5	0

Uncoated samples decayed after 4 weeks while coated samples (T-3 tomatoes) remained healthy for 60 days. The antagonistic action of the edible coatings used against the decay causing microorganisms are primarily attributed to the presence of compounds such as polysaccharides, mannans, anthraquinones, and lectins (Monjezi et al., 2019, Thiruppathi et al., 2010). Mohebbi et al (2012) has also reported that the synergistic application of the *Aloe vera* and tragacanth gum based edible coating was most effective in positively maintaining the quality of button mushrooms.

Weight Loss

This quality parameter is crucial since every loss in weight can be translated into economic loss. Fig. 2 shows the percentage weight loss of the coated and uncoated samples.

The results revealed that weight loss occurred at a faster rate in case of controls. The samples receiving treatments T1, T2, and T3 showed 16.23%, 14.45%, and 10.23 % weight loss at 20°C respectively. This was significantly different compared to 23.61% weight loss in the control tomatoes at this temperature ($P < 0.05$).

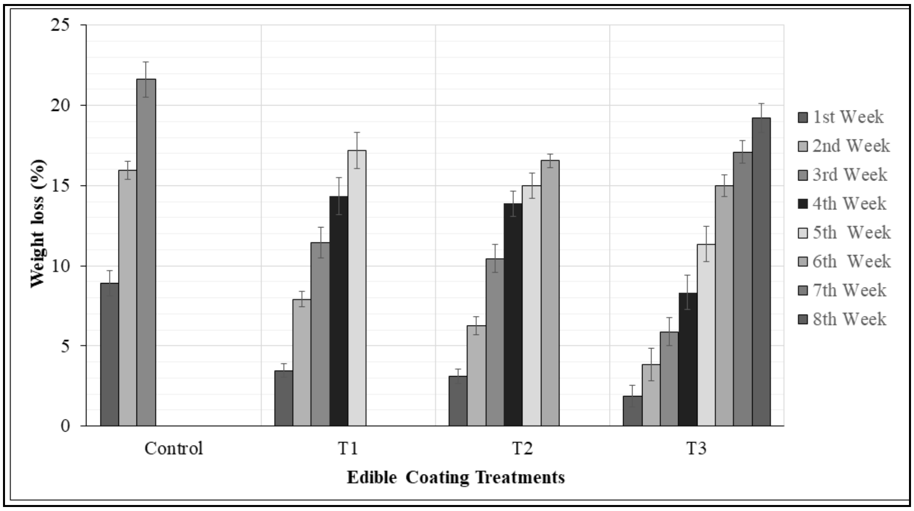

Fig. 2. Comparative analysis of % weight loss between coated and control tomatoes

The post-harvest weight losses generally results from water loss through transpiration (Ergun and Satici, 2012). The guard cells and stomata present in the epidermal layers control the gaseous exchange and loss of moisture from the fruit surface. The covering formed by the edible coatings provides a semi permeable physical barrier to limit the moisture loss (Toğrul and Arslan, 2004). This may also interferes with the level of oxygen uptake by the fruit, retarding the rate of respiration and its associated weight loss (Abbasi et al., 2009). The previous investigation carried out by Emamifar and Bavaisi (2017) also supported the present study and described that *Aloe vera*-tragacanth gum treatment expressively reduced the weight loss levels in strawberries.

Physiochemical Parameters

Effect on pH, titratable acidity and reducing sugar content

Acidity constitutes a crucial quality aspect that greatly affects the fruit taste. The effect of different coating formulations on the pH of tomato fruits is presented in Fig. 3a. The pH value is mainly dependent upon the acid content of the fruit. The pH value showed a progressive increase with the advancement in maturity during the storage period. The control set had the highest pH value of 4.19 after 3rd week of storage. In contrast, all the treated fruits had lower pH values in that period. The fruits receiving the treatment T3 had the lowest pH value of 4.09 when they reached the red stage of maturity after the 8th week of storage. Low pH values related to higher acidity in coated fruits might be accounted by reduced respiration rates due to restricted oxygen availability (Jiang and Li, 2001). Athmaselvi et al (2013) and Abebe et al (2017) also stated that, aloe vera treated tomato fruits were superior in keeping pH and exhibited a healthier response in contrast with untreated fruit.

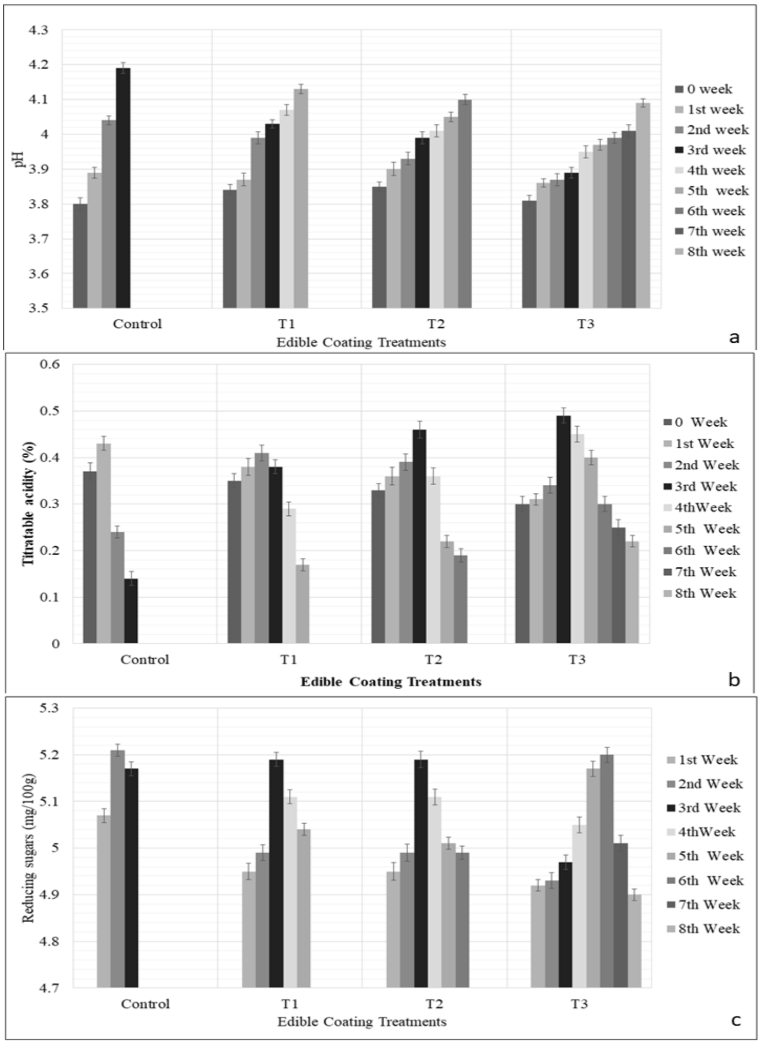

Fig. 3. Comparative analysis of (a) pH, (b) titratble acidity and (c) reducing sugar content between coated and control tomatoes

Tomatoes are highly susceptible to after harvest respiration activity because the organic acid, especially citric acid, undergoes metabolism to provide mediators to the tricarboxylic acid cycle with a rise in respiration. Tomato titratable acidity decreases during ripening. Coated tomatoes showed a statistically higher titratable acidity during the experiment (P = 0.05). Titratable acidity of the tomatoes decreases with storage time (Fig. 3b). The acid content peaks as the tomatoes undergo transition from mature green to turning stage but thereafter shows a decline with the onset of light red stage. The uncoated tomatoes experienced significantly ($p < .05$) greater reduction in acidity from initial day (0) to end day of storage (12) and the value drops from 1.01% to 0.22%. The coated tomatoes showed a slower decline in acidity. The slower decline in acidity values in samples that were coated indicated a decrease in the rate of transpiration due to a decreased accessibility of organic acids, that is important for acidity in fruits. The gradual change in the acid concentrations of the experimental groups compared with the control indicated that edible coatings were effective in delaying the metabolic changes occurring in the treated fruits. The uncoated fruits showed reduced TA values for the ripened stage (0.14%) because of the rapid conversion of organic acids into sugars. The degrees of titratable acidity in coated fruits were considerably maintained with T3 (0.22%) exhibiting the most positive results. It has been reported that tragacanth and *Aloe vera* gum worked well to maintain the titratable acidity of coated strawberries in comparison to the controls (Nasar et al., 2023). As the ripening progresses, the acids present in the fruit might be converted into sugars, used in respiration or other cellular activities. The edible coatings slow down the rate of acid metabolism. Since organic acids (malic or citric acid) can be utilized as primary substrates for respiration, a decrease in acidity is seen in terms of upsurge in respiration of cells of fruits.

During the storage, total reducing sugar rose markedly from mature green stage to the onset of yellow pigmentation with the propensity to decrease as the ripening progressed (red stage) (Fig. 3c). These changes were analogous to both control and experimental groups however the transition was slow with the treated tomatoes since the process of ripening had been retarded by the edible coatings. The tomatoes in all the three treatments T1, T2 and T3 showed the lowest reducing sugar content values i.e., 5.01, 4.95 and 4.93 mg/ml respectively than the control group (5.14 mg/ml) as they ripened completely.

Microbiological Analysis

Aloe vera and Tragacanth gum have both received attention as a food preservative due to their antimicrobial activity against a wide range of bacteria, yeast and fungi. There may be synergistic benefits when using tragacanth gum and aloe vera. The direct antibacterial action of *Aloe vera* is complemented by the protective layer that tragacanth gum helps build for efficacy. The antagonistic action of *Aloe vera* gel against microorganisms has been credited to the presence of natural anthraquinones (Reynolds and Dweck, 1999). The geraniol and carvacrol of tragacanth also exhibit broad spectrum antimicrobial effects (Jahanshahi et al., 2018). The antimicrobial constituents of the edible *Aloe vera* and Tragacanth-based coatings retards the growth rate of microorganisms that directly affect the quality of tomatoes. The coatings serve to limit the gaseous exchange along with the moisture and nutrients availability which are the stakeholders of microbial development. Furthermore, protective coatings prevent the subsequent microbial contamination during storage (Rong-yu and Yao-wen, 2003). The significant effect of applied edible coatings on total viable bacterial and fungal counts of tomato fruits obtained on nutrient agar, MacConkey agar and Potato Dextrose agar is shown in fig. 4.

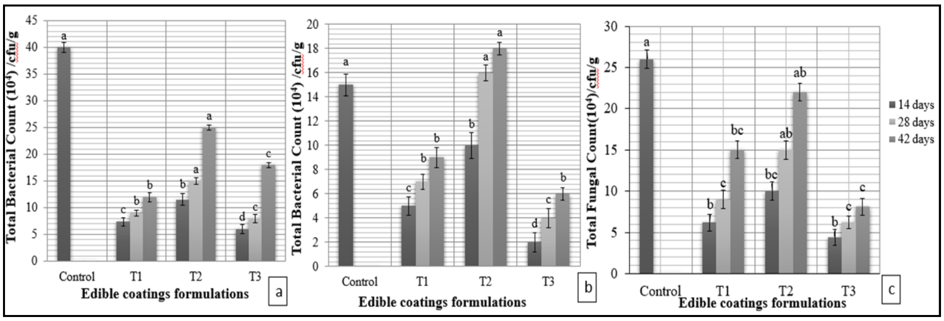

Fig. 4. Comparative analysis of bioburden exhibited by coated and control tomatoes (a) Total viable bacterial count observed on Nutrient agar (b) Total bacterial count observed on MacConkey agar (c) Total fungal count observed on Potato dextrose agar

All the applied coatings exhibited antagonist behavior against bacteria and fungi as observed by the lesser bioburden isolated from treated fruits in comparison to that of control fruits. The bacterial count increased gradually as the days progressed in the case of coated tomatoes but remained low in comparison to the control tomatoes. For instance, after six weeks of storage T3 registered the total viable bacterial count of 8.1×10^4cfu/g (nutrient agar), less than the one for the control tomatoes after 2 weeks. A significant growth inhibition of gram-negative bacteria was witnessed for T3 as seen on MacConkey agar. The effects of all the treatments on total viable bacterial count vary significantly from each other at $p \leq 0.05$. The ability of edible coatings to retard the fungal growth depended upon the nature of applied coatings. The data clearly showed that the fungal load of tomato fruits was considerably higher than that seen for the total bacterial load. A significant reduction was deployed by the T1 and T2. However, the most effective results were noted for the T3 since maximum fungal growth inhibition was registered by the T3 treatment. The effects of all the treatments on total viable fungal count vary significantly from each other at $p \leq 0.05$.

CONCLUSION

The comparative analysis of physical, physiochemical and microbiological parameters revealed that *Aloe vera*-Tragacanth coating (T3) was proven to be the most efficient natural treatment to extend the shelf life of tomatoes up to 8 weeks. *Aloe vera* and tragacanth gum are being investigated as preservatives in fruits, vegetables, meats, and other perishable foods because of their natural origins and proven antibacterial activity. Although these natural ingredients have a lot of potential, more study is required to determine their safety and effectiveness for a range of food preservation applications.

Edible coating offers a layer of defense that prevents food from spoiling in addition to fixing surface damage to the fruit. Natural polymers for bio-preservation of fruits appears to be a low cost, ecofriendly alternative to conventional physical and chemical treatments and would be reasonable for building up a biological process for large scale production.

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N/A

CONFLICT OF INTEREST

Authors declare no conflict of interest.

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