USE OF BIOPRESERVATIVES AND PACKAGING ON THE SHELF LIFE AND QUALITY OF TOMATO

SANCHITA ROY

DEPARTMENT OF HORTICULTURE

SHER-E-BANGLA AGRICULTURAL UNIVERSITY

DHAKA-1207

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Approved by

Supervisor
Dr. Md. Nazrul Islam
Professor
Department of Horticulture
SAU, Dhaka

Co-Supervisor
Dr. Shormin Choudhury
Associate Professor
Department of Horticulture
SAU, Dhaka

Prof. Dr. Md. Jahedur Rahman
Chairman
Examination committee

Dedicated to My

Beloved Parents

Who has always helped me and believed that I could do it

DEPARTMENT OF HORTICULTURE

Sher-e-Bangla Agricultural University

Sher-e-Bangla Nagar, Dhaka-1207

Ref: Date:

CERTIFICATE

This is to certify that thesis entitled, "USE OF BIOPRESERVATIVES AND PACKAGING ON THE SHELF LIFE AND QUALITY OF TOMATO" submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE (MS) in HORTICULTURE, embodies the results of a piece of bona fide research work carried out by SANCHITA ROY, Registration No. 13-05298 under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that such help or source of information, as has been availed of during the course of this investigation has duly been acknowledged.

Dated: June, 2020
Dhaka, Bangladesh

Professor Dr. Md. Nazrul Islam
Supervisor
Department of Horticulture
Sher-e-Bangla Agricultural University
Dhaka-1207

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USE OF BIOPRESERVATIVES AND PACKAGING ON THE SHELF LIFE AND QUALITY OF TOMATO

ABSTRACT

BY

SANCHITA ROY

Tomato is a perishable fruit that has a relatively limited shelf life. Storage conditions affect the consistency and nutritional value of fresh products such as tomatoes. This work aimed at evaluating the effect of different packaging and biopreservatives on shelf life of tomato. Different biopreservatives i.e. P_0: Control, P_1: Aloe vera extract, P_2: Garlic extract, P_3: Neem leaf extract, P_4: Propolis and two postharvest packaging system i.e. non-packaging and perforated polyethylene packaging were used. The results revealed that biopreservatives treated tomatoes showed better shelf life and quality relative to untreated fruits. Among the biopreservatives, Propolis treated fruits showed highest values in titratable acidity, total soluble solid (%Brix), ascorbic acid content, beta carotene and lycopene and increased shelf life of tomato in polyethylene bags. Thus, propolis treatment appeared the most useful than other treatments for extending the shelf life and quality of tomato.

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LIST OF ACRONYMS

DAS	=	Days after storage
et al.	=	and others (at elli)
CRD	=	Completely Randomized Design
LSD	=	Least Significant Difference
pH	=	Hydrogen ion conc.
DMRT	=	Duncan's Multiple Range Test
BBS	=	Bangladesh Bureau of Statistics
DF	=	Degree of freedom
CV %	=	Percent of coefficient of variation
e.g.	=	Example
FAO	=	Food and Agriculture Organization
SAU	=	Sher-e-Bangla Agricultural University
SE	=	Standard Error
RH	=	Relative Humidity

CHAPTER I

INTRODUCTION

Tomato (*Lycopersicon esculentum* Mill.) being a climacteric fruit has a relatively short postharvest life, belong to solanaceae family (Smith, 2001). It is one of the most scientifically researched horticultural produce because of its commercial consideration (Correia et al., 2015). South American Andes (Naika *et al*., 2005) which is in present day Peru was believed as origin of tomatoes where it was growing in the wild at the foot of hills. Probably it was originated in South America over millions of years ago but it was brought in Asia in early 19th century. In global scale of production, tomato produced in 4.8 million hectares with an estimated production of 162 million tonnes (FAOSTAT, 2014).

Tomato has become not only an important cash and industrial crop in many parts of the world but also essential due to its nutritional value to human diet and subsequent importance in human health (Ayandiji *et al*., 2011; Willcox*et al*., 2003). It is a rich source of energy, carotenoids, flavonoids, phenolics, mineral nutrients, vitamin C and dietary fibers which are necessary and serve as protective ingredients for human health (Beecher, 1997; Wold *et al*., 2004).) and it is available around the year. Tomato contains larger amounts of lycopene, a type of carotenoid with anti-oxidant properties (Arab and Steck, 2000) which is helpful in reducing the incidence of some chronic diseases (Basu and Imrhan, 2007) like cancer and many other cardiovascular disorders (Burton-Freeman and Reimers, 2010). Lycopene is produced by phyto-chemical synthesis in plants and microorganisms whereas animals do not produce its (Sahasrabuddhe, 2011).

Tomatoes spend so much time on shelves and refrigerators that an estimated 20% are lost due to spoilage. From the jiffy produce is harvested the clock starts ticking and decaying starts. (Jaiswal*et al*., 2018). Postharvest loss has been defined as a measurable quantitative and qualitative loss of a given product at any moment along the postharvest chain (De Lucia and Assennato, 1994). Bio preservation is a novel food preservation method defined for extension of shelf life and enhanced food safety by the use of natural or controlled micro biota and or anti-microbial compounds (Baldwin *et al*., 1996). Edible

film coatings on fresh tomatoes can provide a modified internal atmosphere for the product and thereby acts as an alternative for reducing the quality and quantity losses, and thus the major post-harvest loss of tomatoes can be reduced (Dhall, 2013).

Propolis a non-toxic resinous is a natural glue produced by honey bees with main constituents being resins (flavonoids, phenolicsand their esters), waxes, vitamins and essential oils (Juliano *et al*., 2007). One of the most important chemical compounds in propolis is flavonoids, which also have many useful properties including antibacterial activity, anti-inflammatory activity, antioxidant activity, and antitumor activity (Pujirahayu *et al*., 2014). Aloe vera gel has been one of the promising bio preservatives which have been identified as a novel edible film coating with good antimicrobial properties (Misir*et al*., 2014).Treatment with plant extracts as aloe vera, garlic, neem, onion etc. (Anjum *et al*., 2016) is popularly practiced abroad. Neem (leaves, flowers, seeds, fruits, roots, bark) found its utility since ancient days and have been used to treat infections, inflammation, fever, skin diseases and dental disorders (Helmy *et al*., 2007; Mosaddek and Rashid, 2008). Azadirachtin is regarded as the most active substance in neem which has growth regulating, fungicidal, and insecticidal properties (Schmutterer, 1990).

Other studies reported that extracts of garlic has fungicidal effects (Singh and Agrawal, 1988). Ajoene, a garlic-derived sulfur-containing compound, demonstrated antimicrobial activity against some gram-positive bacteria and gram-negative bacteria (Naganawa *et al*., 1996). Harden bergs was the pioneer in using plastic films for fresh produce. Many of the initial applications focused primarily on reducing moisture loss, providing protection from handling damage and improving produce appearance. Perforated and micro-perforated polymeric packages reduce off-odour formation (Izumi *et al*., 1996). This study sought the effect of the biopreservatives (garlic, Aloe vera, propolis, neem) and packaging (perforated and non-perforated) on the postharvest quality and shelf life of tomato during storage.

Nowadays, biopreservatives are extensively used entirely the world for its spanking quality. However, in Bangladesh, there is limited information and experience to use

biopreservatives as postharvest treatment to extend the shelf life of tomatoes. Therefore, the present study was commenced to fulfill the following objectives:

i) to investigate the effect of different packaging and biopreservatives on shelf life of tomato; and

ii) to evaluate the quality parameters of tomato fruits during postharvest period.

CHAPTER II

REVIEW OF LITERATURE

Tomato production rate is high during harvest time, but post-harvest processing and preservation techniques are inadequate. Due to shortage proper systems of preservation and processing fruits are spoiled very early (Ameyapoh et al., 2008). Edible coatings can be a protection for fresh products and can effect similarly as modified atmospheric storage condition (Park et.al., 1994 and Tharanathan, 2003).

2.1 Role of bio-coating materials for prolonging shelf life of tomato

Tomato being climacteric in nature causes rapid ripening and postharvest losses of 25 to 40% or even more than this (Verma and Joshi, 2000; Sankar *et al*., 2002; Pulamte, 2010). Between harvest and consumption both qualitative and quantitative losses occur in fruits. Now a day's concern for high quality chemical preservatives free and extended shelf life is increasing. Edible coating which is applied to the product surface to extend post harvest shelf life (Baldwin *et al*., 1995) can be used as an alternation of natural waxy coatings to create a barrier to moisture, oxygen and solute movement (Mchugh*et al*., 2000). The pertinent literature regarding the effect of such substances on storage quality of tomatoes and some other stuffs is reviewed under following headings:

2.1.1 Effect of Aloe vera

Aloe vera is a tropical and subtropical plant which is used in medicinal and therapeutic properties (Eshun and He, 2004). It is widely used as an antimicrobial agent and protective to quality losses during ambient storage (Ergun and Satici, 2012). Aloe vera coating was effective for prevention of loss of moisture and firmness. It also control respiration rate in fruits such as grapes (Castilo *et al*., 2010), sweet cherries (Martinez *et al*., 2006) and nectarines (Ahmed *et al*., 2009).

An experiment was conducted by Vahdat*et al.* (2010) and result was found that Aloe vera coating remarkably minimized weight loss as compared to the control fruits. The minimum weight loss was observed in fruits coated with 100% (v/v) and minimum firmness was noticed in control fruits at the end of storage. Aloe vera treated fruits have shown higher titratable acidity, sugar content and ascorbic acid than untreated fruits. Romero *et al.* (2006) found that Aloe vera coating was effective to decrease

weight loss and reduce respiration rate during postharvest storage quality of sweet cherry.

Garcia *et al*. (2014) conducted a research on the effects of Aloe vera coating on the postharvest quality of tomato traditionally used edible coatings as barriers during processing, handling and storage of tomatoes to enhance its quality. The Aloe vera coating controlled the weight loss and maintained delayed ripening. According to the findings Aloe vera gel may be useful to breeders and postharvest technologists, as well as distributors, importers and exporters, in handling and processing tomatoes.

Chandran and Mini (2018) conducted a research on Aloe vera gel as a bio preservative for shelf life extension of mature green tomato in Kerala, India. Some physiological parameters such as weight loss, respiration rate and membrane integrity were observed and found that untreated tomatoes showed higher weight loss and respiration rate than the treated fruits. Aloe vera treated fruits showed higher membrane integrity. The experiment found that Aloe vera gel could be used as bio-preservative to enhance shelf life of mature tomatoes. Jaiswal *et al*. (2018) studied the enhancement of the shelf life of tomato using Aloe vera and neem-based herbal coating in India. It was found that the coatings minimized weight loss, ripening process and decay to a large extent and extended shelf life and quality of tomatoes.

2.1.2 Effect of garlic

Garlic has been well known for its dietary and medicinal properties for centuries Ross *et al*. (2001). Martin and Ernest (2003) proved in their studies that garlic has antimicrobial effects. Pai and Platt (1995) found that garlic has inhibitory effect on growth of both gram positive and gram negative bacteria the similar as yeast and mold. According to Wszelaki and Miller (2005), early blight of tomato can be reduced by garlic extract. Karapynar (1989) found that garlic clove can inhibit mycelia growth of *A. flavus* and aflatoxin.

Olaniran *et al.* (2015) studied on Bio-preservative effect of ginger (*Zingiberofficinale)* and garlic powder (*Alliumsativum)* on tomato paste in Nigeria. The study investigated fresh tomato fruits (Roma VF variety) samples with each receiving different concentrations of ginger, ginger and garlic powder (2 and 4% w/w) and stored over a period of 8 weeks. The study concluded that combined garlic and ginger (2 and 4%)

suitably preserved tomato paste for 8 weeks without deterioration at refrigeration temperature (4±1°C).

Rayman and Baysal (2017) studied on the antimicrobial effects of thyme, Garlic and Basil Oleoresins against *Bacillus Coagulans* in Tomato Sauce. It was investigated that the antimicrobial activities of thyme (*Thymus serpyllum*), basil (*Ocimum basilicum*) and garlic (*Allium sativum*) oleoresins against Bacillus coagulans in a tomato based sauce. The inhibitory and bactericidal effects of oleoresins against *B. coagulans* ATCC 7050 were tested in the sauce. These findings demonstrated that basil and garlic have the same inhibitory effects against bacteria and were more effective than thyme in the tomato based sauce.

Tijjani *et al*. (2014) conducted an experiment on effect of some selected plant extracts on *Aspergillus flavus*, a causal agent of fruit rot disease of tomato (*Solanum lycopersicum*) in Bauchi State in 2007. Different concentrations of some selected plant extracts (neem seed, moringa seed, garlic bulb and emulsified neem seed oil each with five concentrations were used and found that garlic bulb, emulsified neem seed oil and aqueous moringa seed extracts exhibited the highest control of the pathogen than aqueous neem extract.

2.1.3 Effect of neem

Neem is the mostly used plant with several biological activities in its leaves, bark, fruits and seeds. According to Wang *et al*. (2010), neem extracts can reduce the growth of pathogen in plum and pear fruits. Fungicidal effect of neem was observed in pineapple (Ribeiro et al., 2011), tomato (Lima *et al*., 2011; Oladimej *et al*., 2013), apple (Leite *et al*., 2009) and orange (Al-samarrai *et al*., 2013).

Hosea et al. (2017) conducted a research on Postharvest shelf life and quality of tomato fruits and found that neem leaf powder can extend the shelf life of tomatoes. Neem (leaves, flowers, seeds etc.) have been used traditionally against many diseases. Banna *et al*. (2014) studied on growth inhibitory effect of ethanolic neem leaves extract on Klebsiella, Salmonella and Staphylococcus aureus and found its growth inhibitory effect on Klebsiella and Salmonella but failed to inhibit Staphylococcus aureus. Siddiqua *et al*. (2018) conducted a research on effects of neem leaf extract and hot water treatments on shelf life and quality of banana in Bangladesh and

observed 40% neem leaf extract showed longest shelf life, minimum color change, minimum disease incidence and higher titrable acidity.

Gulhane *et al*. (2018) conducted a research on to evaluate the efficacy of natural plant extracts (edible coating solution) like Neem oil, Guar gum, Aloe vera gel and Marigold extract as potential fruit preservatives against 5 different types of fruits such as Apple, Banana, Chikoo, Papaya and Tomato. It was found that except Apple and Chikoo other selected fruits lost their weight even after treatment with the edible coating solution. The study revealed that Tomato was good; papaya and chikoo were slightly dull whereas banana was < 50% brownish in colour. Pulp colour analysis showed that Apple and Tomato were 100% good, papaya 75% good, chikoo 50% Good while Banana 25% Good.

2.1.4 Effect of propolis

According to Lima *et al*. (1998) Propolis has inhibitory effect on postharvest pathogens, *Botrytis cinerea* and *Penicillum expansum* .Ordónez et al. (2010) conducted a study on potential application of Northern Argentine propolis to control some phytopathogenic bacteria. Strong antibacterial activity was detected against *Pseudomonas syringae* p var tomato CECT 126, *Pseudomonas corrugata* CECT 124 and *Xanthomonas campestris* pvarvesicatoria CECT 792. It was concluded that application of the Argentine propolis extracts diluted with water may be promising for the management of postharvest diseases of fruits.

Putra *et al*. (2017) studied on tomatoes Stored at room temperature where as tomatoes were grouped into 5 group, namely control group (no coating application), ethanol group (tomato wash with ethanol), and application group (coated with 5%, 10%, and 15% propolis). Variables observed during study were weight change, fruit firmness, total soluble solute, vitamin C, and lycopene. All tomatoes were kept in room temperature for 14 days and observation conducted every 2 days. The application of 10% propolis as bio-coating minimized rate of weight loss and controlled fruit firmness than the untreated fruits. Yusuf *et al.* (2005) conducted a research with 10 different concentrations (10, 7, 5, 3, 1, 0.1, 0.07, 0.05, 0.03 and 0.01 μg mL^{-1}) of propolis methanol extract (PME) on *Phytophthora infestans, P. capsici* and *P. parasitica in vitro*. Among them four of PME concentrations (10, 7, 5 and 3 μg mL^{-1})

tested completely inhibited the mycelia growth of the three *Phytophthora* species,it was found that PME had compound with fungicidal effects on *Phytophthora* species.

Abdel-Kader *et al*. (2019) studied on black spot disease infecting Guava fruits under artificial infestation and found that no disease symptoms was observed at treatments of propolis dissolved in acetic acid, chitosan dissolved in acetic acid, beeswax dissolved in water or ethanol and combination between propolis plus chitosan dissolved in acetic acid. The research showed the efficiency of natural bio-agent to control and minimize guava fruit black spot caused by *Guignardiam angiferae*.

Propolis (bee glue) is a natural matter made up of resinous collected by bees from buds and trees to reinforce honeycombs and cover cracks in the hive. Agüero *et al*. (2010) found that propolis has different chemical compositions according to the botanical origin. Propolis was found to have many biological properties, including anti-inflammatory, antioxidant, antiviral, cytostatic, antiproliferative, cytotoxic, hepato-protective and antimicrobial activities. Some Argentine propolis shows antibacterial activity against antibiotic resistant human pathogenic bacteria (Isla *et al*., 2005).

2.2 Effect of packaging

In postharvest stage desiccation, wilting, shriveling and mechanical injury occurs in agricultural crops due to their continuous respiration (Rahman *et al*., 2010).The problem can be maintained by adapting scientifically packaging and storage methods.

Haile and Safawo (2018) used different packaging materials for increasing shelf life and quality of tomato and found that decay loss of tomato fruits at the end of storage was higher on non-perforated polythene bag than the perforated and carton. On perforated polythene bag color and overall acceptability was maximum. In the conclusion perforated polythene bags can extend the storage life of tomato fruit. Basel *et al*. (2002) found that packaging influence physiological weight loss (PWL) in tomato fruits. The lowest PWL was recorded from non-perforated polythene bag than perforated bag. This might be due to polythene protects the fruits from mechanical damage, reducing loss of moisture, providing modified atmosphere.

Mir and Beaudry (2000) revealed that, packaging material protects the product from external environment and minimize exposure to pathogens and contaminants to extend the shelf life of the produce. Thompson (2001) showed that mold growth may

be due to condensation of water vapor, in non- perforated polybag lower permeability of the film can cause water vapor. Badshah *et al*. (1997) described that, polythene packaging (perforated) helps the color retention of fruits. Ben-Yenonshuna (1985) reported that low density polythene bags delay ripening and softening, and hence improves marketability of climacteric fruits.

González *et al*. (2003) delay ripening and softening, and improvement of marketability of climacteric fruits can be determined by modified atmospheric condition created due to packaging. Paull (1993) found that, at ambient condition termination of shelf life of tomato was determined by shriveling, over ripening, discoloration and mold growth. On control fruits shriveling and mold growth were higher than packaged fruits. In non-packaged fruits shriveling and senescence can occur due to faster transpiration rate. Kader and Rolle (2004) reported that modified gas atmosphere inside package could be the cause to reduce respiration and ethylene production rates, lower ethylene action, delayed ripening and senescence, inhibiting decay causing pathogens and insects.

Nasrin *et al.* (2008) conducted an experiment on effect of postharvest treatments on shelf life and quality of tomato. The effect of chlorine, packaging and storage conditions on quality and shelf life of tomato was observed. Treated fruits kept in perforated polythene bag and ambient condition showed reduction of weight loss and decay. The treatment combination delayed compositional changes such as TSS, reducing sugar-carotene etc. It was concluded that packaging can be helpful to maintain the shelf life and quality of tomato. Sammi and Masud (2007) studied on different packaging systems on storage life and quality of tomato (*Lycopersicon esculentum* var. Rio Grande) during different ripening stages in rawalpindi, Pakistan. Different packaging systems were evaluated to extend storage life of tomato fruits. The results showed that within each ripening stage, the treated fruits remained better than that of control fruits.

CHAPTER III

MATERIALS AND METHODS

This chapter is composed of a concise representation about experimental period, storage room, its controlled condition, treatments used in this experiment, experimental design and layout, data collection and statistical analysis.

3.1 Experimental site

The postharvest experiment was conducted at the postharvest Laboratory of Department of Horticulture, Sher-e-Bangla Agricultural University, Dhaka during the period from April to May 2019.The average temperature and humidity was recorded 26±2°C and 60±2% respectively. Temperature and humidity was recorded with digital temp-humidity recorder (Thermogermany).

3.2 Experimental materials

Properly matured red colored tomatoes of same variety, size, and shape were used for this experiment. The tomatoes were collected in the early morning and transferred carefully to avoid injuries and placed in the postharvest Laboratory of SAU.

3.3 Treatments of the experiment

The experiment comprised of two factors:

Factor A: Postharvest Biopreservatives

i. Control (P_0)
ii. Aloe vera (P_1)
iii. Garlic extract (P_2)
iv. Neem extract (P_3)
v. Propolis (P_4)

Factor B: Postharvest Packaging

i. No packaging (B_1)
ii. Packaging with perforated polybag (B_2)

3.4 Experimental design and treatment application

The two factors experiment was placed in a completely randomized design (CRD) with three replications. Postharvest biopreservatives and packaging conditions were attributed randomly in each replication. Under each replication, five fruits were collected for physical and chemical analysis. A total number of 10×3×5 = 150 matured, uniform sized, fresh healthy fruits were selected. Then the fruits were washed, surface disinfected with ozonized water for 20 minutes and subjected to different treatments. For coating purposes, the fruit was dipped once in the coating material and restrain in it for less than 1 min to have a uniform thin layer of the material over the surface of the fruit. The coated and uncoated (control) fruits were stored in different packaging condition at ambient temperature.

3.5 Preparation of Biopreservatives

Different raw materials essential for the study were collected from karwan bazar, Dhaka. In the laboratory, ripen tomatoes were selected to obtain homogeneous batches based on colour, size, and absence of injuries and healthy. Then the fruits were washed, surface sanitized with ozonized water for 20 minutes. The fruits were subjected to different bio-preservatives as treatment (Plate 1).

3.5.1 Aloe vera gel preparation (P_1)

Extraction of aloe vera gel was completed according to the traditional hand filleted method described by Ramachandra and Rao (2008). Good quality fresh, mature Aloe vera leaves were culled from Krishi market, Dhaka. The fresh gel was prepared from collected aloe vera leaves. 100% aloe vera gel was made and for this at first they were cleaned with tap water and then with distilled water to remove dust. Then the sharp spines located on the leaf margins were removed by a sharp knife, the aloe gel was scooped out of the leaves and this mucilage hydro parenchymal layer was homogenized in a blender machine. No additional water was used here. The gel was then filtered by sieve to remove all unwanted lump and to get 100 percent fresh aloe gel. As the gel is sensitive to enzymatic degradation so the extract was kept in a glass jar in refrigerator.

3.5.2 Garlic extracts preparation (P_2)

Stock garlic solution (300g garlic and 300ml water) was prepared using fresh garlic bulbs which were cleaned, peeled and crushed in blender and then cheesed.10% garlic solution was made with the stock solution. It is then filled up into airtight container and stored in refrigerator.

Fresh garlic extract was prepared using blender by following the method described by Singh and Majumdar (2001). Garlic bulbs were cleaned, peeled, washed and crushedin blender. The sample solution 1:1 (300g garlic and 300ml water) was prepared using fresh garlic bulbs. 10% garlic solution was made with the stock solution.

3.5.3 Neem extracts preparation (P_3)

Fresh green leaves of neem (about 250g) were collected from Sher-e-Bangla Agricultural University campus and blended with 500 ml of distilled water. Then 20% or 40% neem leaf extract solution was prepared by taking 60 ml or 120 ml raw neem leaf extract in 500 ml beaker with the addition of 240 ml or 180 ml distilled water separately to make a final volume of 300 ml (Mia, 2003). Finally the extracts solutions were filtered before use and stored in refrigerator at 5°C.

3.5.4 Propolis extraction preparation (P_4)

The propolis was collected from entomology department of Sher-e-Bangla Agricultural University. For EEP solution (ethanol extracted propolis), 10g ground propolis was added to 150ml (70% ethanol).Then heated it. It was kept overnight for proper extraction in freeze, then filtered it. The volume was made to 100ml with distilled water. Then the propolis extract was kept in refrigerator.

a **b** **c** **d**

Plate 1: Preparation of biopreservatives (a. Aloe vera, b. Garlic. c. Neem, d. Propolis) in the postharvest laboratory.

3.5.5 Methods of applying the bio preservatives

The fresh harvested sound ripe red tomatoes were disinfected by ozone water. The cleaned intense red colored tomatoes were coated. A treated by dipping them in each bio preservatives solution separately for 5 minutes. 10 tomato fruits per treatments were weighted by sensitive balance. The weighted tomato fruits were kept in plastic bag and without plastic bag with three replications at ambient condition.

3.6 Observation

During the entire postharvest storage period the experimental fruits were profoundly observed every day to observe any special change. Physical observations (weight loss, shrinkage severity and shelf life) were recorded on 15 days of storage. For determining chemical analysis total soluble solids (TSS), titratable acidity (TA), β-carotene, lycopene, ascorbic acid and pH of each samples were portrayed on 15 days of storage.

3.7 Methods of studying physico-chemical parameters

3.7.1 Physical parameters

3.7.1.1 Estimation of weight loss

Physiological loss in weight was calculated using the following formula and expressed as percentage (Enab, 2013).

Physiological weight loss (%) = (IW- FW/IW) x100

Where IW- Initial Weight, FW- Final Weight

3.7.1.2 Shelf life estimation

Shelf life of tomato fruits as influenced by different postharvest storage treatments and packaging system was calculated by counting the days required to decay 25% as to retaining and eating qualities. The fruit peel degree of blackish spot or decayed was graded.

3.7.1.3 Visual scoring of tomato skin

Visual scoring of tomato skin was assessed by estimating the percentage of shrinkage condition of tomato during storage. The percentage of shrinkage was graded as

following: 0= no shrinkage, 1=1-10% shrinkage, 2=>10-20% shrinkage, 3=>20-30% shrinkage, 4=>30-40% shrinkage, 5=>40% shrinkage.

3.7.2 Chemical parameters

3.7.2.1 pH

pH was estimated using a phs-25 pH meter. An electrolytic cell consists of two electrodes (calomel electrode and glass electrode) was standardized with buffer solution of pH 4. Any known pH value may be used as buffer solution. Then the electrodes were suspended into the test sample. A voltage corresponding resembling to the pH of the solution was determined by the instrument. For preparing sample solution of fruits, tomatoes were chopped into small pieces and ground into a fine paste by mortar and pestle. The tomato juice was transferred into a test tube and the pH of the paste was determined by inserting the electrodes into the juice and stagnated readings were recorded.

3.7.2.2 Total soluble solid (TSS)

Total soluble solids content of tomato was evaluated by using hand refractometer. Two drop of tomato juice squeezed from the fruit pulp on the prism of the refractometer. Percent TSS was gained from direct reading of the instrument.

3.7.2.3 Titratable acidity (TA)

Titratable acidity was calculated by chemical analysis process using tomato pulp. The titratable acidity of tomato fruit was determined by method of Ranganna (2004). From tomato fruit small piece of 5 gram was chopped, blended by mortar and pestle. Then, the juice was filtered by sieve in a beaker. The volume was made up to 100 ml by adding distilled water. 2 drops phenolphthalein indicator was added. From this solution 10 ml was taken in a conical flask and titrated against 0.1N NAOH. 0.1N NaOH was added drop wise and the solution shaken thoroughly until a pink color was gained. It was repeated 3 times. The acid content of the tomato sample was calculated using the formula below:

$$\text{TA\%} = \frac{(titrate \times Normality\ of\ alkali \times Volume\ made\ up \times Equivalent\ wt.of\ acid \times 100)}{(Volume\ of\ sample\ taken\ for\ estimation \times wt.of\ sample\ taken \times 1000)}$$

0.1N solution preparation:

To make 0.1N solution, 4.0 g of sodium hydroxide was mixed in water to make 1 liter volume.

Phenolphthalein indicator preparation:

To prepare phenolphthalein indicator 0.5g phenolphthalein was weighted. 50% ethanol was prepared by compiling 50 ml ethanol and 50 ml distilled water. Then 0.5g phenolphthalein was liquefied in 50% ethyl alcohol solution.

3.7.2.4 Ascorbic acid

Ascorbic acid content (ascorbic acid) was evaluated by using 2, 6-Dichlorophenol indophenols (DCPIP) visual titration method(Rangana,2004). 5gm tomato fruit sample was blended, juice was filtered by sieve. Volume was made up to 100 ml by adding oxalic acid.10 ml from solution was taken in conical flask and titrated against DCPIP (Standard dye) to a pink end point which should retain for at least 15 seconds. Ascorbic acid content in terms of mg/100g pulp weight was estimated using the following formula:

Ascorbic acid (mg/100g): =

$$\frac{\text{Titra} \times \text{dye factor} \times \text{volume made up}}{\text{Aliquot of extract taken for estimation} \times \text{wt. or vol. of sample taken for estimation}} \times 100$$

5% oxalic acid solution preparation

It was made by liquefying 50g oxalic acid powder in 1000 ml distilled water.

Dye solution preparation:

It was prepared by dispersing 260 mg of the sodium salt of 2, 6-dichlorophenol indophenol in approximately 1000 ml of hot distilled water containing 210 mg of sodium bicarbonate.

Standardization of dye solution

Ten milliliters (10 ml) of standard ascorbic acid solution was taken in a conical flask and 5 ml of oxalic acid was added to it. A micro burette was placed with the dye solution. The quantity of the conical flask was titrated with dye solution. The content of conical flask was titrated with dye till the pink colored end point arrived. The milliliters of dye solution requisite to complete the titration were recorded. Dye factor was estimated using the following formula:

Dye factor = 0.5/ titrate value

3.7.2.5 β-carotene content:

B-carotene in tomato was determined according to the method of (Nagata and Yamashita, 1992).One gram of pulp was mixed with 10ml of acetone: hexane mixture (4:6) and vortex for 5 minutes. The mixture was filtered and absorbance was measured at 453nm, 505nm and 663nm wave length. The estimation was done by following formula:

B-carotene (mg/100gm) = $0.216\ A_{663}$-$0.304\ A_{505}$+$0.452\ A_{453}$

3.7.2.6 Lycopene determination

Absorption determination for lycopene content was estimated following the method of Alda et al. (2009) by using T60 UV-Visible Spectrophotometer.1g tomato from each treatment was weighed into a conical flask. Lycopene in the tomato was extracted using hexane:ethanol: acetone (2:1:1) (v/v).One ml juice of each sample were homogenized with 25ml hexane: 12.5ml ethanol: 12.5ml acetone which were then placed on electric shaker for 30 minutes. Then 10ml distilled water was added and agitated continuously for 2 min. The solution was then left to separate into two distinct polar and non -polar layers. The absorbance was measured at 472nm, using hexane as a blank in spectrophotometer. The lycopene concentration was calculated using its extinction coefficient (E 1%, 1cm) of 3450 in hexane at 472nm.The lycopene concentration was expressed as mg/100g product.

$$\text{At } \lambda = 472 \text{ nm: lycopene content (mg/100g)} = \frac{E}{3.45} \cdot \frac{20}{m}$$

3.8 Statistical analysis

The collected data were statistically analyzed by STATISTIX 10 software. The mean of different parameters was compared by DMRT (Duncans Multiple Range Test). The significance of difference between the pairs of means was compared by least significant difference (LSD) test at the 1% level of probability (Gomez and Gomez, 1984).

CHAPTER IV

RESULTS AND DISCUSSION

This chapter portrayal the results acquired from the present study. The effect of different treatments in respect of physico-chemical changes and shelf life of tomato are represented and discussed in this chapter. These results are briefly mentioned under the following headings:

4.1 Physiological Weight loss:

In postharvest storage life of tomato, weight loss is considered as one of the main quality parameters. Different biopreservatives, their concentration, environmental condition manifested more conspicuous effect on total weight loss of tomato during storage. The weight loss percent calculating for each bio-preservative and packaging exhibited significant variation (Table 1, Appendix I).

It was recorded that maximum (3.48%, 5.60%, 7.87%, 10.24% and 13.94% at 3^{rd}, 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) percentage of weight loss of tomato under postharvest biopreservative treatment in P_0 (Controlled fruit) followed by P_2 (Garlic treated fruit), P_3 (Neem treated fruit) and minimum (1.79%, 3.59%, 4.87%, 6.47% and 7.51% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS) was in P_4 (Propolis treated tomato). Aloe vera treated fruits showed statistically the second (2.50%, 3.90%, 5.91%, 7.77% and 9.43% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS) lowest weight loss of storage (Figure 1). The weight loss percentage regardless of all biopreservatives was increased with storage time and the highest weight loss percentage was noticed at the end of storage day which was similar with the findings of Krishnamurthy and Babu (1993).

It was discovered that highest (3.68%, 6.29%, 8.49%, 10.75% and 12.59% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS) weight loss was observed in no packaging condition and lowest (1.67%, 2.91%, 4.54%, 6.37% and 8.13% at 3^{rd}, 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) weight loss was recorded in packaging condition (Figure 2). This result could be due to the maintenance of high humidity in micro atmosphere within packages and due to low water vapor transmission rates of packaging material (Moneruzzaman *et al*., 2009).

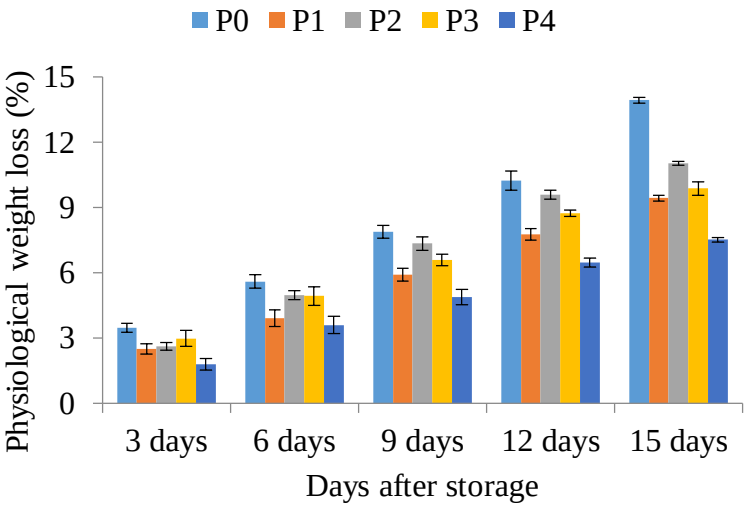

Figure 1: Effect of different biopreservatives on physiological weight loss (%) of tomato fruits at at the end of shelf life

P_0: Control, P_1: Aloe vera, P_2 : Garlic extract, P_3 : Neem extract, P_4 : Propolis

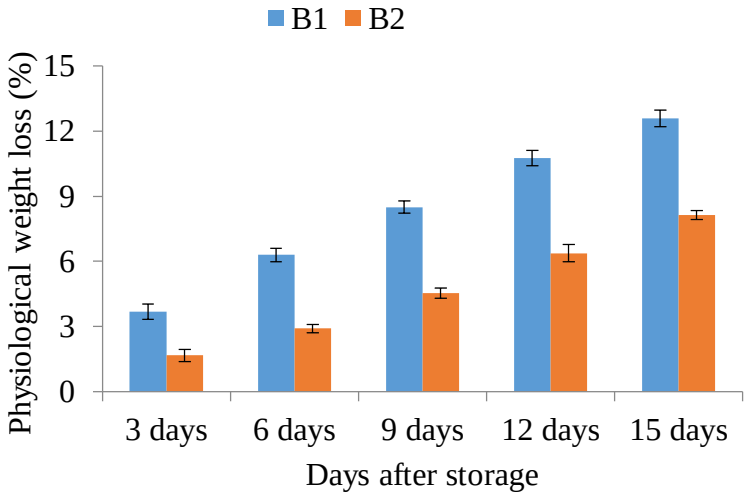

Figure 2: Effect of different packaging on physiological weight loss (%) of tomato fruits at the end of shelf life

B_1: No packaging, B_2: Packaging

The data revealed that the combined effect between the postharvest biopreservatives and different packaging were found statistically significant at 3rd, 6 th, 9th, 12th and 15th days after storage. The maximum (4.82%, 7.72%, 9.86%, 12.53% and 16.86% at 3rd, 6 th, 9th, 12th and 15th DAS) weight loss was showed in P_0B_1 (Controlled fruits in no packaging condition) combination and minimum (1.07%, 2.23%, 3.87%, 5.09% and 6.18% at 3rd,

6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) rate was recorded in P_4B_2 (Propolis treated fruits in packaging condition (Table 1). By considering all above findings, it was found that weight loss in controlled fruits was the highest due to higher respiration rate, transpiration or evaporation of moisture (Sani *et al*., 1997). Edible bio-coating (eg: propolis, Aloe vera, garlic, neem) is helpful to extend the shelf life of fruit by preventing the rate of respiration and moisture loss. A similar result is also reported by Thirupathi *et al*. (2006). Therefore, among the treatments propolis appeared to be the best bio-preservative in packaging and no packaging condition. The application of 10% propolis as bio-coating minimized rate of weight loss and controlled fruit firmness than the untreated fruits (Putra *et al*., 2017).

Table 1. Combined effect of different biopreservatives and packaging on physiological weight loss (%) of tomato at different days after storage (DAS)

Treatments	Physiological weight loss (%)				
	3 DAS	6 DAS	9 DAS	12 DAS	15 DAS
P_0B_1	**4.82 a**	**7.72 a**	**9.86 a**	**12.53 a**	**16.86 a**
P_0B_2	2.13 de	3.48 d	5.89 c	7.96 c	11.04 c
P_1B_1	3.74 b	5.14 c	7.89 b	9.93 b	11.07 c
P_1B_2	1.27 f	2.67 de	3.94 d	5.61 d	7.79 de
P_2B_1	3.13 c	6.52 b	9.62 a	11.81 a	13.31 b
P_2B_2	2.12 de	3.45 d	5.07 c	7.39 c	8.75 d
P_3B_1	4.2 b	7.13 ab	9.24 a	11.64 a	12.86 b
P_3B_2	1.76 e	2.73 de	3.94 d	5.84 d	6.91 e
P_4B_1	2.52 d	4.96 c	5.88 c	7.86 c	8.86 d
P_4B_2	**1.07 f**	**2.23 e**	**3.87 d**	**5.09 d**	**6.18 e**
LSD(0.01)	0.46	1.11	1.08	0.99	1.66
SE	0.16	0.38	0.37	0.34	0.58
CV (%)	7.35	10.21	7.02	4.91	6.82

P_0: Control, P_1: Aloe vera, P2: Garlic extract, P_3: Neem extract, P_4: Propolis, B_1: Non Packaging, B_2: Packaging; Means with different letters significantly differ at LSD's test at $P \leq 0.01$; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference.

Togrul and Arslan (2004) found that coating is helpful for reducing moisture loss and gaseous exchange which alter internal carbon dioxide, oxygen, ethylene level and slowdown ripening process and maintain good fruit shape. The film created on the skin of fruit performs as an additional barrier. Aloe vera coating was also effective to reduce

weight loss during postharvest storage quality of tomato (Romero *et al*., 2006). The packaging condition is also helpful for reduction of weight loss which is similar to the findings of Nasrin *et al*. (2008). The use of different natural plant extracts, prevent physical and chemical changes like weight loss than control untreated fruits. It is supported by the findings of Shindem *et al*. (2009).

4.2 Shelf life

The shelf life starts from the time of harvesting and ends with the beginning of fruit spoilage (Mondal, 2000).In this study shelf life was determined by visual observation. A significant variation was noticed in respect of shelf life of tomato due to the effect of different postharvest biopreservatives and packaging condition (Table 2, Appendix II).It could be mentioned that maximum (14.16 days) shelf life of tomato fruits was belong to propolis extract (P_4) followed by Aloe vera (12.83 days) treated fruits and minimum (6.5 days) shelf life was recorded in controlled fruits (P_0) (Figure 3).

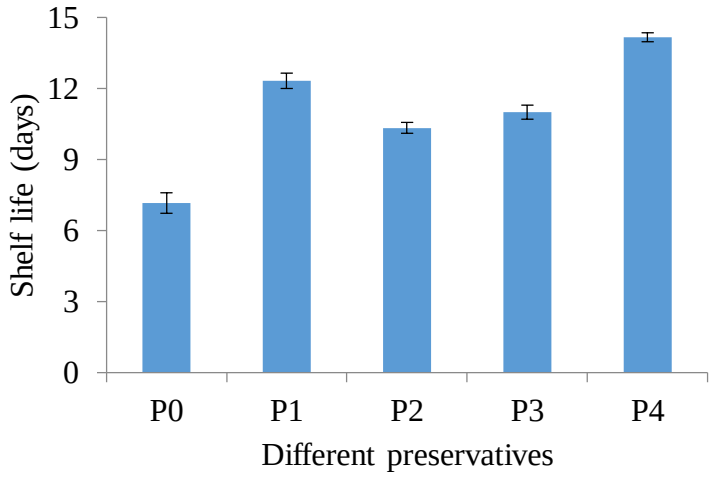

Figure 3: Effect of different biopreservatives on shelf life (days) of tomato fruits at the end of shelf life

P_0: Control, P_1: Aloe vera, P_2: Garlic extract, P_3 : Neem extract, P_4 : Propolis

Coating materials are generally used to enhance the shelf life and fruit quality during storage and actions of these coatings deal with the reduction of moisture loss and enhancement of natural appearance and products quality during storage by Olivas and Barbosa-Canoras. Baldwin *et al*. (1995) also stated that edible coating could be used to

enhance postharvest shelf life and to create an inhibitor of moisture, oxygen and solute movement (Mchughet *et al*., 2000).

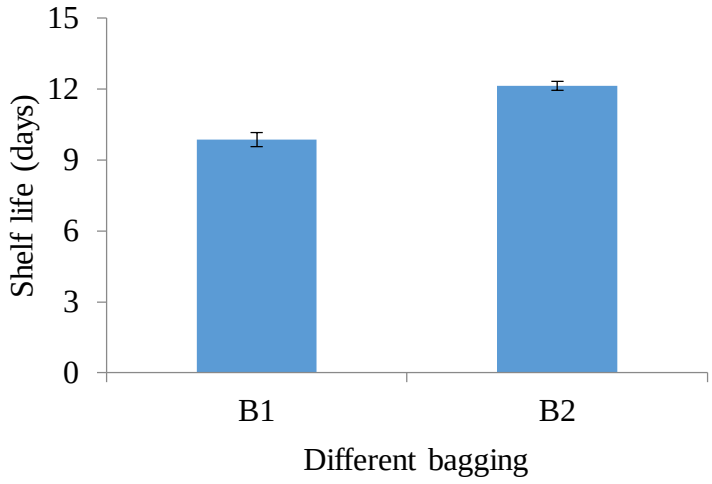

Figure 4: Effect of different packaging on shelf life (days) of tomato fruits after storage
B_1: No packaging, B_2: Packaging

The highest (11.5 days) shelf life was noted down in packaging condition and lowest (10.4 days) shelf life was found in non packaging condition (Figure 4). The fruits packed in bags and kept in ambient condition maintained a better aroma and texture. This is also supported by the studies of Nirupama *et al*. (2010) and Speirs *et al*. (1998) in tomatoes. Haile (2018) stated that packaging can extend the storage life and overall acceptability of tomato fruit. The fresh fruits and vegetables stored in plastic films inhibit the transmission of respiratory gases for the accumulation of carbon dioxide and oxygen depletion around the crop thus may enhance their shelf life (Kader *et al*., 1989).

The combination effect also showed significant differences among biopreservatives and packaging. P_4B_2 (Propolis treated fruits in packaging condition) combination showed 15 days shelf life followed by P_1B_2 (Aloe vera treated fruits in packaging condition) combination showed 13.66 days shelf life and lowest (5.33 days) shelf life was found in P_0B_1 (controlled fruits in no packaging condition) combination (Table 2).

Table 2. Combined effect of different biopreservatives and packaging on shelf life (days) of tomato after storage

Treatments	Shelf life (Days)
P_0B_1	**5.33 g**
P_0B_2	9.00 f
P_1B_1	11cd
P_1B_2	13.66 b
P_2B_1	9.33 ef
P_2B_2	11.33cd
P_3B_1	10.33 de
P_3B_2	11.667 c
P_4B_1	13.33 b
P_4B_2	**15.00 a**
LSD(0.01)	1.08
SE	0.37
CV (%)	4.21

P_0: Control, P_1: Aloe vera, P2: Garlic extract, P_3: Neem extract, P_4: Propolis, B_1: Non Packaging, B_2: Packaging; Means with different letters significantly differ at LSD's test at $P \leq 0.01$; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference

4.3 Severity on the basis of shrinkage

Shrinkage is one of the indexes of dilapidation, degenerating the quality and degrading the quantity. In the present study shrinkage occurred in all sample postharvest biopreservatives treated as well as untreated tomatoes. Postharvest biopreservatives and different packaging had significant effect on shrinkage severity in tomato skin (Table 3, Appendix III).

The maximum (39.385% at 15th DAS) shrinkage severity was recorded in P_0 (Controlled fruits) and minimum (11.25% at 15th DAS) value was recorded in P_4 (Propolis treated fruits). Aloe vera (P_1) treated fruits also showed the second lowest (24.08% at 15th DAS) shrinkage value (Plate 2; Figure 5). It might be due to the rejuvenation action of natural coatings which had a restrictive effect on ethylene biosynthesis and delay the activity of enzymes accountable for ripening, cell degradation was obstructed which gradually reduce moisture loss and shrinkage percentage. It is also supported by Gharezi *et al.* (2012).

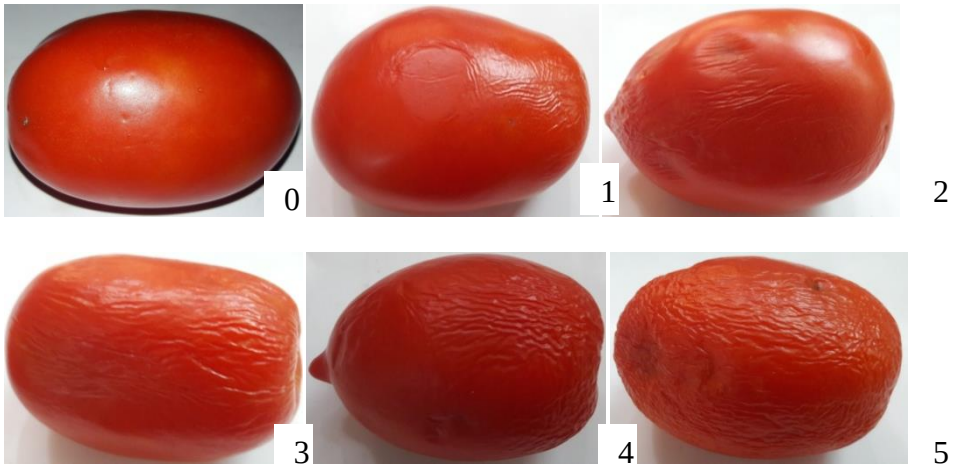

Plate 2: Visual scoring of tomato skin according to shrinkage severity before end of shelf life; 0= no shrinkage, 1=1-10% shrinkage, 2=>10-20% shrinkage, 3=>20-30% shrinkage, 4=>30-40% shrinkage, 5=>40% shrinkage

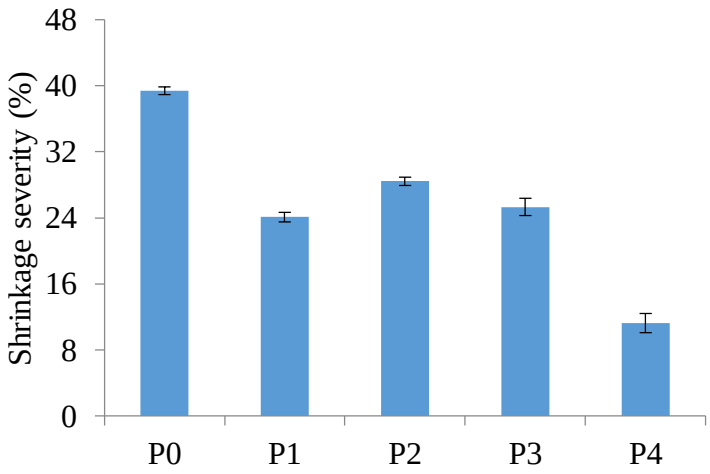

Figure 5: Effect of different biopreservatives on shrinkage severity (%) of tomato at the end of shelf life

P_0: Control, P_1: Aloe vera, P_2 : Garlic extract, P_3 : Neem extract, P_4 : Propolis

Highest (28.36% at 15th DAS) shrinkage occurred in B_1 (no packaging) condition and lowest (23.01% at 15th day) value was recorded in B_2 (Figure 6). This could be assigned to the compliance of high humidity in the micro atmosphere within the packages by the respiring fruits and due to ignoble water vapour transmission rates of packaging material (Moneruzzaman *et al*., 2009).

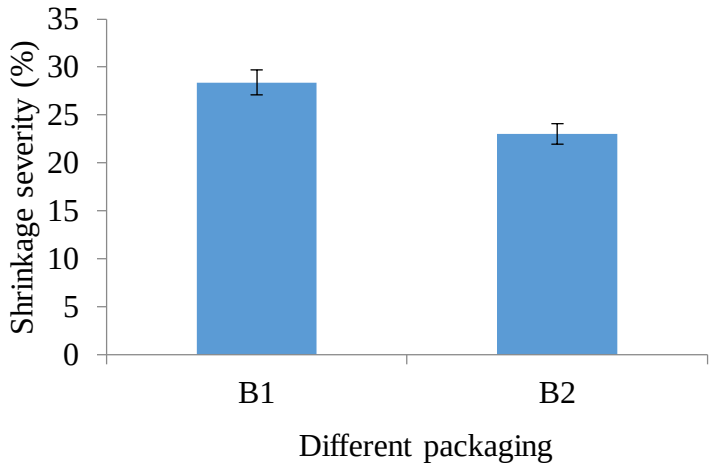

Figure 6: Effect of different packaging on shrinkage severity (%) of tomato at the end of shelf life

B_1: No packaging, B_2: Packaging

Table 3. Combined effect of different biopreservatives and packaging on shrinkage severity (%) of tomato at the end of shelf life

Treatments	Shrinkage severity (%)
P_0B_1	**45.13 a**
P_0B_2	33.64 b
P_1B_1	26.23 de
P_1B_2	21.93 f
P_2B_1	29.70 c
P_2B_2	27.17 d
P_3B_1	26.50 d
P_3B_2	24.10 ef
P_4B_1	14.27 g
P_4B_2	**8.25 h**
LSD(0.01)	2.39
SE	0.83
CV (%)	3.95

P_0: Control, P_1: Aloe vera, P2: Garlic extract, P_3: Neem extract, P_4: Propolis, B_1: Non Packaging, B_2: Packaging; Means with different letters significantly differ at LSD's test at $P \leq 0.01$; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference

Significant variation was observed in combined effect of postharvest biopreservatives and packaging on shrinkage severity of tomato. The highest (45.13% at 15^{th} DAS) value was observed in P_0B_1 combination (Controlled fruits in no packaging condition), and lowest

value (8.25% at 15th day) was observed in P_4B_2 (Propolis treated fruits in packaging condition) followed by P_1B_2 (aloe vera treated fruits in packaging condition) combinations (Table 3).

4.4 pH

Significant variations in pH were recorded in different postharvest treated tomatoes before end of shelf life (Table 4, Appendix IV). The highest (5.28) pH value was recorded in P_0 (controlled or untreated tomatoes) followed by P_2 (Garlic coated tomatoes), P_3 or neem coating (5.03) and the lowest (4.5) value was found in P_4 or propolis coated tomatoes. Aloe vera treated tomatoes showed lower pH value and that was 4.8 (Figure 7).

The maximum (5.08) pH value was recorded in B_1 and minimum (4.82) pH value was observed in B_2. So, from the above explanation it was summarized that untreated fruits showed the topmost value and propolis coated fruits showed lowest value. Moreover, after propolis, Aloe vera recorded the second lowest value. By recking the packaging effect B_2 (Packaging) showed the lowest value compared to B1 (No packaging) (Figure 8). Due to general catabolization of organic acids and their conversion into sugar the ranges of pH value become larger and acidity get lower day by day . The result showed that propolis gave the best result in packaging and no packaging condition.The coating of propolis remarkably reduced the increase of tomato juice pH.

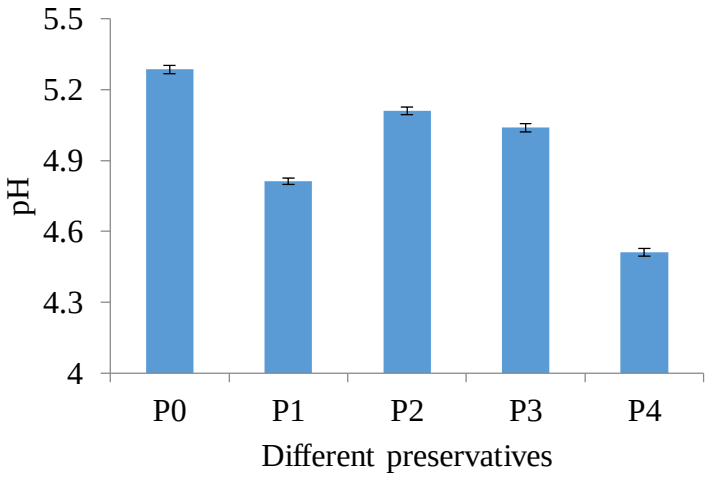

Figure 7: Effect of different packaging on pH of tomato at the end of shelf life

P_0: Control, P_1: Aloe vera, P_2 : Garlic extract, P_3 : Neem extract, P_4 : Propolis

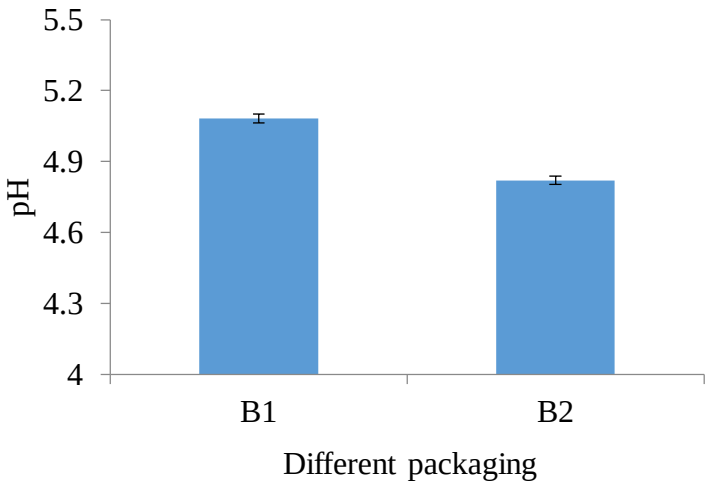

Figure 8: Effect of post-harvest treatments on pH of tomato at the end of shelf life

B_1: No packaging, B_2: Packaging

Table 4. Combined effect of different biopreservatives and postharvest packaging on pH of tomato at the end of shelf life

Treatments	PH
P_0B_1	**5.53 a**
P_0B_2	5.04 b
P_1B_1	4.98 b
P_1B_2	4.65 c
P_2B_1	5.25 ab
P_2B_2	4.97 b
P_3B_1	5.09 b
P_3B_2	4.99 b
P_4B_1	4.56 c
P_4B_2	**4.46 c**
LSD(0.01)	0.30
SE	0.10
CV (%)	2.54

P_0: Control, P_1: Aloe vera, P2: Garlic extract, P_3: Neem extract, P_4: Propolis, B_1: Non Packaging, B_2: Packaging; Means with different letters significantly differ at LSD's test at $P \leq 0.01$; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference.

The results are in agreement with the findings reported by Jiang and Li (2001). They observed coated tomato fruits reduced pH and reported that the higher acidity maintained in coated fruits might be due to reduction of respiration rate and limited availability of

oxyzen. Aloe vera treated fruits also kept lower pH and displayed better effect comparing with untreated fruits. Aloe vera treated tomatoes also reduced pH (Athmaselvi *et al*., 2013). Morever, untreated fruits accelerated the ripening process due to the scarcity of any coating. As a result they displayed higher pH value. The results are comparable with the findings reported by Wani *et al*. (2014).

The combination effect of biopreservatives and different packaging showed significant result. The maximum (5.53) pH value was recorded from P_0B_1 (controlled fruits in no packaging condition) combination and minimum (4.46) value was noticed in P_4B_2 (propolis coating in packaging condition) combination proceeded by P_1B_2 (Aloe vera coated fruits in packaging condition) combination where pH value was 4.65 very much near to P_2B_2 combination (Table 4).

4.5 Total Soluble Solid (TSS):

The TSS is a refractometric indicatory index of the proportion (%) of dissolved solids in a solution (Beckles, 2012).There was a significant difference in TSS content during postharvest storage due to different biopreservatives and packaging condition (Table 5, Appendix V).The propolis (P_4) coated tomatoes maintained the lowest TSS value (3.1%) followed by Aloe vera (3.15%), while uncoated control tomatoes maintained the highest TSS value (4.21%) (Figure 9).

Packaging and no packaging condition showed significant variation in TSS value as the topmost value B_1 (3.63%) was recorded in tomatoes kept in no packaging condition and lowest value B_2 (3.36%) was recorded in tomatoes kept in packaging condition (Figure 10).The range of TSS in ripe tomatoes is 3.5 to 6.0 °Brix, which revealed that packaging maintains the proper temperature conditions and maintains high fruit quality. It is similar with the findings of Filgueira (2004).

The combined effect of biopreservatives and packaging condition in content of TSS were recorded to be significant. The maximum (4.40%) TSS value was observed in P_0B_1 (Controlled fruits in no packaging condition) combination and minimum (3.03%) TSS value was observed in P_4B_2 (Propolis treated fruits in packaging condition) combination (Table 5).

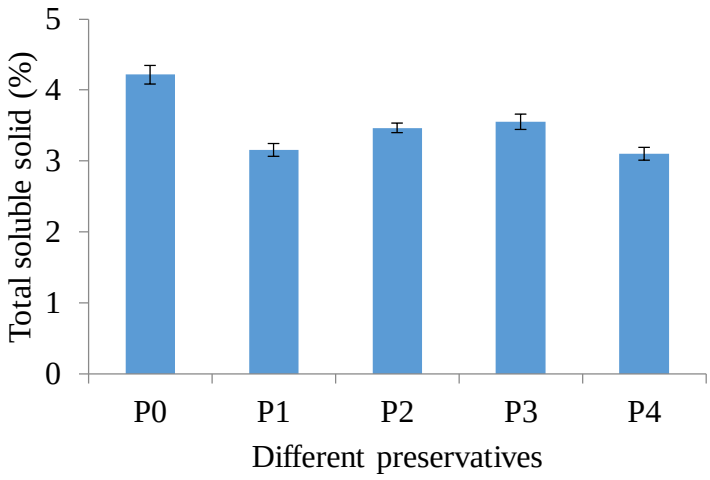

Figure 9: Effect of different biopreservatives on total soluble solid (%) of tomato at the end of shelf life

P_0: Control, P_1: Aloe vera, P_2 : Garlic extract, P_3 : Neem extract, P_4 : Propolis

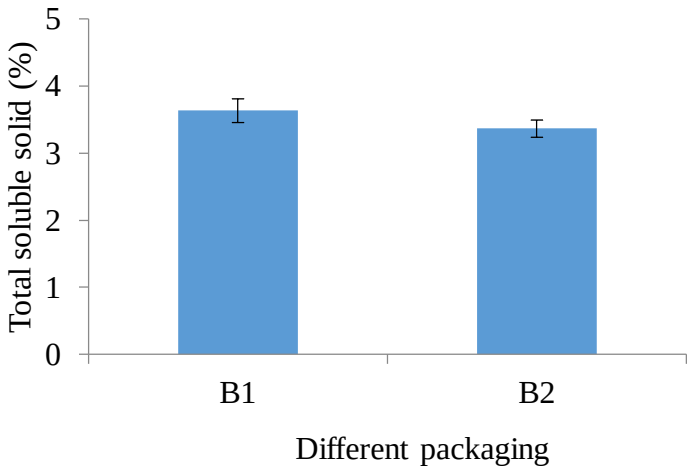

Figure 10: Effect of different packaging on total soluble solid (%) of tomato at the end of shelf life

B_1: No packaging, B_2: Packaging

Final TSS was 3.03 to 4.40% which is more or less similar with the findings of Salunkhe (1991) and changes of TSS were lower in packed fruits than the control fruits. The results revealed that the coating could be helpful to reduce oxygen or elevate carbon dioxide as a result reduce ethylene production .It also reduce respiration rate as a result lower TSS content is occurred in coated fruits. Similar result was found by Yaman and Bayoındırl (2002). Furthermore, the reduction of TSS during storage is generally due to sugar-acid

metabolism. Increase of TSS in uncoated control tomatoes could be due to excessive moisture loss which enhances hydrolysis of carbohydrates to soluble sugars (Waskar *et al*., 1999; Nath *et. al*., 2011).

Table 5. Combined effect of different biopreservatives and postharvest packaging on TSS of tomato at the end of shelf life

Treatments	Total Soluble Solid
P_0B_1	**4.40 a**
P_0B_2	4.03 ab
P_1B_1	3.22 de
P_1B_2	3.09 de
P_2B_1	3.76 bc
P_2B_2	3.17 de
P_3B_1	3.6 bcd
P_3B_2	3.5 cde
P_4B_1	3.17 de
P_4B_2	**3.03 e**
LSD(0.01)	0.51
SE	0.18
CV (%)	6.25

P_0: Control, P_1: Aloe vera, P2: Garlic extract, P_3: Neem extract, P_4: Propolis, B_1: Non Packaging, B_2: Packaging; Means with different letters significantly differ at LSD's test at $P \leq 0.01$; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference.

4.6Titratable Acidity (TA)

There was a significant variation TA (%) of tomatoes during storage due to various effective bio-coatings and packaging conditions (Table 6, Appendix V). The maximum value (0.67%) of titrable acidity for tomatoes was recorded for propolis (P_4) followed by Aloe vera (P_1), the value was (0.61%) and minimum value (0.38%) was recorded for untreated control fruits (P_0) (Figure 11). The acidity of tomato plays a main role and bestows taste to the fruit. Citric acid and malic acid are predominantly presented in ripen tomato. According to one author malic acid concentration decreases during ripening and citric acid increases up to turning period, whereas another observed that maleic acid increased unswervingly entirely maturation (Humle, 1971).

Fruit coating is helpful for reduction of respiration rate and minimize the utilization of respiratory products such as organic acid. As a result control fruits have less TA value

than coated fruits. It is also supported by the findings of Tefera *et al*. (2008). Higher fruit acidity is a precedence, as it causes a lower occurrence of fungal infection (Getinet et al., 2008). Reduction of TA generally occurred with proceeding maturity and preservation period in tomatoes. It is also supported by Getinet et al. (2008). The curtailment in acidity as the storage time proceeded might be due to the alternation of organic acid into sugar and their derivatives or their application in respiration (Rai et al.,2012). Kumar (1998) and Kumar & Dhawan (1995) also recorded the similar results in mango. However, packaging technique have different results in case of pear and apple fruits (Huang et.al. 2009).

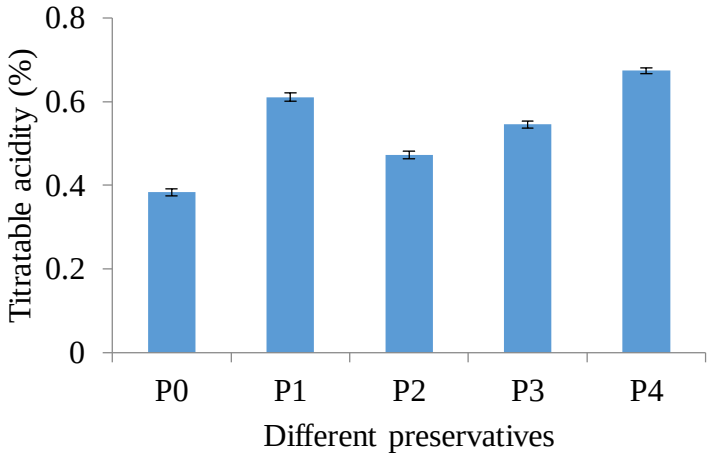

Figure 11: Effect different biopreservatives on Titratable acidity (%) of tomato at the end of shelf life

P_0: Control, P_1: Aloe vera, P_2 : Garlic extract, P_3 : Neem extract, P_4 : Propolis

The maximum value (0.56%) was recorded in tomatoes kept in packaging condition followed by (0.50%) in no packaging condition (Figure 12). Accumulation of carbon dioxide in the fruit tissue and formation of carbonic acid, causing acidiosis. It is satisfying the findings of Carrillo Lopez*et al*. (1995). The lower acidity contents at the end of storage period probably due to packaging condition as described by Badshah *et al*. (1997) and Ali and Thompson (1998).

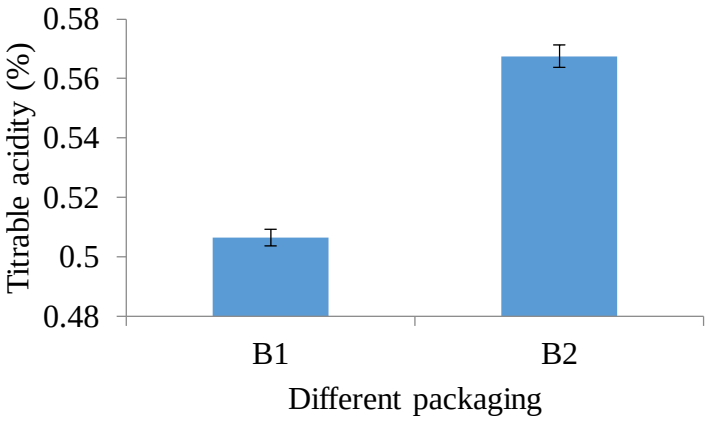

Figure 12: Effect of different packaging on Titratable acidity (%) of tomato at the end of shelf life

B_1: No packaging, B_2: Packaging

Table 6. **Combined effect of different biopreservatives and postharvest packaging on the titratable acidity (%) of tomato at the end of shelf life**

Treatments	Titratable acidity (%)
P_0B_1	**0.34 e**
P_0B_2	0.43 cde
P_1B_1	0.56 abcd
P_1B_2	0.66 ab
P_2B_1	0.41 de
P_2B_2	0.54 bcd
P_3B_1	0.58 abc
P_3B_2	0.51 bcd
P_4B_1	0.64 ab
P_4B_2	**0.71 a**
LSD(0.01)	0.16
SE	0.06
CV (%)	7.94

P_0: Control, P_1: Aloe vera, P2: Garlic extract, P_3: Neem extract, P_4: Propolis, B_1: Non Packaging, B_2: Packaging; Means with different letters significantly differ at LSD's test at $P \leq 0.01$; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference.

The combined effect of biopreservatives and packaging in respect of tritable acidity were observed to be significant. The top most (0.71%) value was recorded in P_4B_2 (Propolis coated fruits in packaging condition) combination followed by P_1B_2 (aloe vera treated fruits in packaging condition) combination where the value was (0.66%). On the other

hand, lower (0.34%) value was recorded in P_0B_1 (controlled fruits in no packaging condition) combination (Table 6).

4.7 Ascorbic acid content

Ascorbic acid naturally found in fresh fruits and loss of ascorbic acid is very common. As ascorbic acid is very responsive to oxidation it is difficult to preserve it (Mandal, 2016). During storage phenol oxidase and ascorbic acid oxidase enzymes activity is the cause of reduction of ascorbic acid (Salunkhe *et al*., 1991). The significant variation was noticed in biopreservatives and different packaging (Table 7, Appendix V).

Different biopreservatives showed significant differences at storage time. The maximum value (13.66 mg/ 100 g) was recorded for propolis (P_4) treated fruits followed by aloe vera (11.18 mg/ 100 g), neem (10.65mg/ 100 g) , garlic(10.43 mg/ 100 g) and lowest (9.10 mg/ 100 g) value was observed in controlled fruits (P_0) (Figure 13). Coating could serve as a protective layer to maintain the diffusion of oxygen and maintain respiration. Srinivasa *et al*. (2006) stated that high ascorbic acid found in coated fruit could have an association with slower respiration rate which is due to the coatings and reduction of oxygen diffusion.

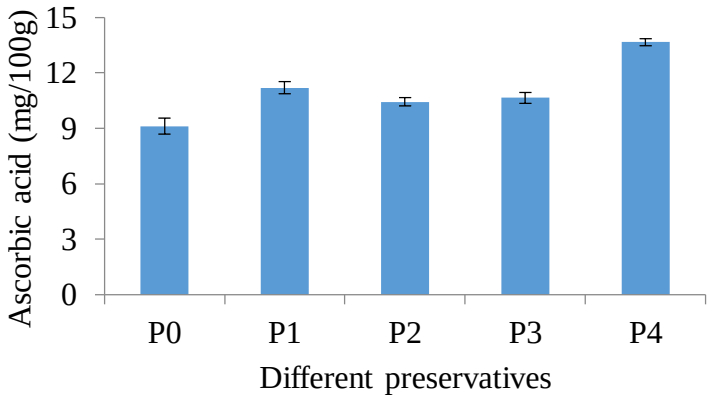

Figure 13: Effect of different biopreservatives on ascorbic acid (mg/100g) of tomato at at the end of shelf life

P_0: Control, P_1: Aloe vera, P_2 : Garlic extract, P_3 : Neem extract, P_4 : Propolis

Significant variation was reported in case of packaging like highest (11.91 mg/100g) value of ascorbic acid was noticed in no packaging condition lowest (10.11mg/100 g) value was observed (Figure 14). Incase of packed fruits vitamin C reduction rate was less

than the no packaging fruits and these chemical compositions and changing trend of its after storage are supported by the statement of Salunkhe (1991).

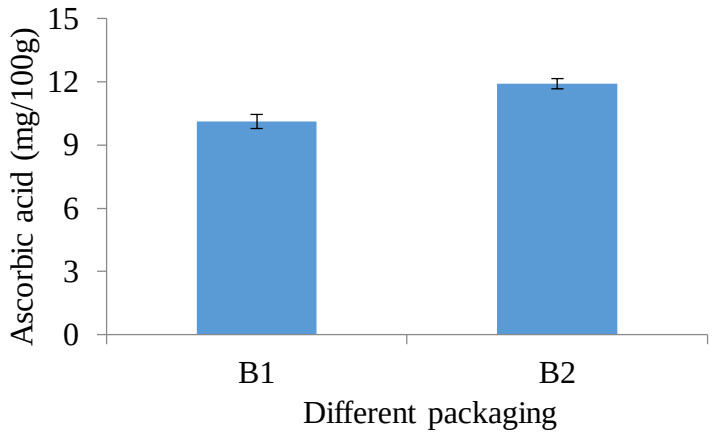

Figure 14: Effect of different packaging on ascorbic acid (mg/100g) of tomato at the end of shelf life
B_1: No packaging, B_2: Packaging

Table 7. Combined effect of different biopreservatives and postharvest packaging on ascorbic acid (mg/100g) content of tomato at the end of shelf life

Treatments	Ascorbic acid (mg/100g)
P_0B_1	**8.20 h**
P_0B_2	10.02 ef
P_1B_1	10.33 e
P_1B_2	12.05 c
P_2B_1	9.30 g
P_2B_2	11.57 d
P_3B_1	9.65 fg
P_3B_2	11.66 cd
P_4B_1	13.1 b
P_4B_2	**14.23 a**
LSD(0.01)	0.46
SE	0.16
CV (%)	1.77

P_0: Control, P_1: Aloe vera, P2: Garlic extract, P_3: Neem extract, P_4: Propolis, B_1: Non Packaging, B_2: Packaging; Means with different letters significantly differ at LSD's test at $P \leq 0.01$; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference.

The combined effect of biopreservatives and packaging in respect of Ascorbic acid were observed to be significant. The topmost (14.23 mg/100 g) value was recorded in P_4B_2

(Propolis coated fruits in packaging condition) combination followed by P_1B_2 (Aloe vera treated fruits in packaging condition) combination where the value was 12.05mg/100g. On the other hand, lower (8.20mg/100g) value was recorded in P_0B_1 (controlled fruits in no packaging condition) combination (Table 7).

4.8 β-carotene

Carotenoids are naturally occurring colored compounds which is responsible for the pleasing yellow, orange and red colour of fruits and vegetables (Dutta *et al*.,2005).β-carotene content of tomato showed significant variations in case of biopreservatives and packaging condition and their combined effects also appeared to be significant (Table 8, Appendix V).

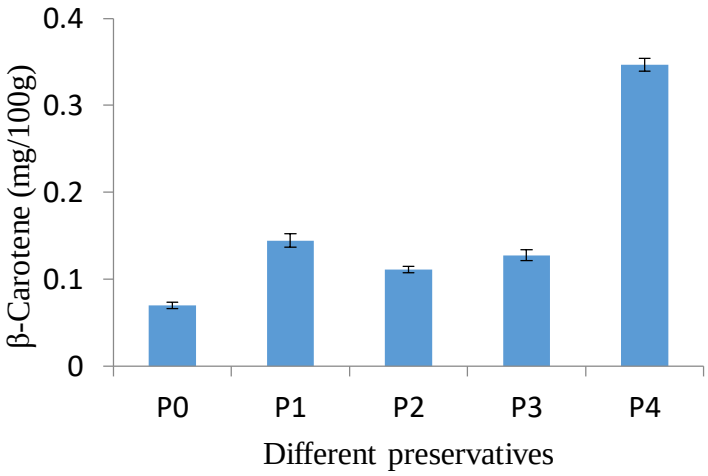

Figure 15: Effect of different biopreservatives on β-Carotene (mg/100g) of tomato at at the end of shelf life

P_0: Control, P_1: Aloe vera, P_2 : Garlic extract, P_3 : Neem extract, P_4 : Propolis

The maximum (0.346 mg/100 g) β-carotene content was recorded in propolis (P_4) treated fruits followed by aloe vera (0.14 mg/100 g), neem (0.13 mg/100 g), garlic (0.11 mg/100 g) and minimum (0.06 mg/100g) β-carotene content was recorded in controlled (P_0) fruits (Figure 15). Biopreservatives formed a protective layer on fruit by keeping the respiration rate lower and maintained the coated fruits quality in storage condition. In this study, the accumulation rate of the beta-carotene content in the control fruits was lower compared to the coating treated a fruit which is supported by the findings of Kator *et al*.(2018).β-carotene has preventive effect on oral, pharynx and larynx cancers (Mayne *et al*.,1993).

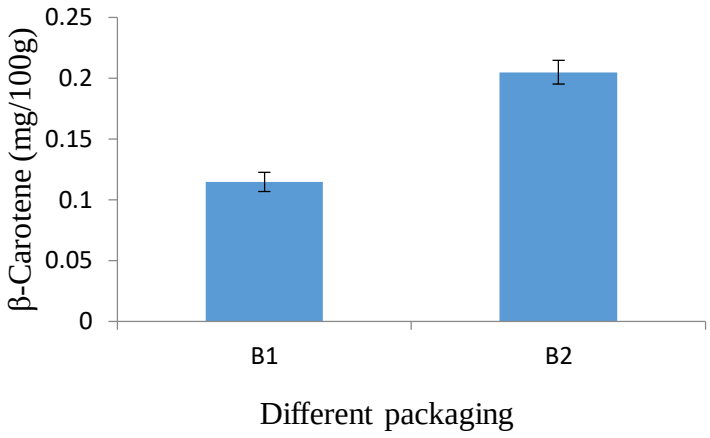

Figure 16: Effect of different packaging on β-Carotene (mg/100g) of tomato at the end of shelf life
B_1: No packaging, B_2: Packaging

Table 8. Combined effect of different biopreservatives and postharvest packaging on the β-carotene of tomato at the end of shelf life

Treatments	β-carotene (mg/100g)
P_0B_1	**0.05e**
P_0B_2	0.09 d
P_1B_1	0.13 c
P_1B_2	0.15 b
P_2B_1	0.09 d
P_2B_2	0.13 c
P_3B_1	0.13 c
P_3B_2	0.13 c
P_4B_1	0.17 b
P_4B_2	**0.53 a**
LSD(0.01)	0.02
SE	6.69
CV (%)	5.13

P_0: Control, P_1: Aloe vera, P2: Garlic extract, P_3: Neem extract, P_4: Propolis, B_1: Non Packaging, B_2: Packaging; Means with different letters significantly differ at LSD's test at $P \leq 0.01$; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference.

The highest (0.21 mg/100 g) β-carotene content was observed in packaging condition and lowest (0.11 mg/100 g) β-carotene content was noticed in no packaging condition (Figure 16). It is shown of that during storage, B-carotene was higher when tomato packed in polyethylene bag which is similar to the findings of Nasrin *et al*. (2008). Saltveit (1999)

studied that enhancement in carotenoid with fruit ripening is associated with the climacteric increase in respiration and ethylene production.

It was seemed that maximum (0.53 mg/100 g) β-carotene content was observed in P_4B_2 (Propolis fruits in packaging condition) combination and lowest (0.05 mg/100 g) value was recorded in P_0B_1 (Control fruits in no packaging condition) combination. Moreover, P_1B_2 (Aloe vera treated fruits in packaging condition) where the value was 0.15 mg/100 g also showed significantly lower β-carotene content (Table 8).

4.9 Lycopene

Lycopene is the major pigment which is responsible for the characteristic deep red colour of ripe tomato fruits. Lycopene exists as small globules, in the chromoplasts and distributed throughout the fruit. Shi and Maguer (2000), Opined that lycopene should be protected from unbearable heat and extreme pH conditions, expouser to light, oxygen and lipid deteriorative enzymes due to inhibit its oxidation and isomerization. Lycopene content of tomato showed significant variations in case of biopreservatives and packaging and their combined effects also appeared to be significant (Table 9, Appendix IV).

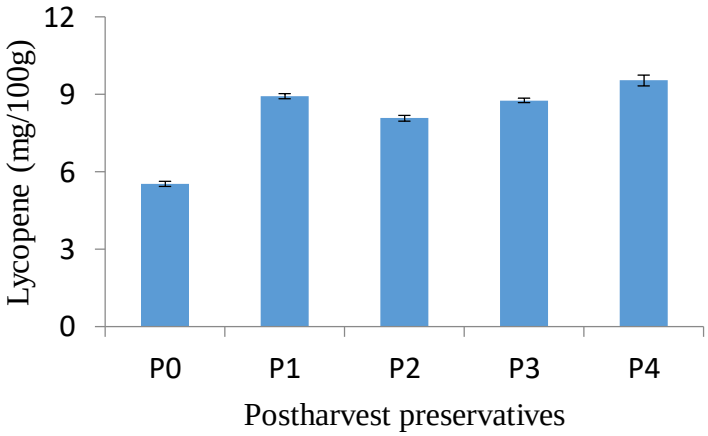

Figure 17: Effect of different biopreservatives on lycopene (mg/100g) of tomato at the end of shelf life

P_0: Control, P_1: Aloe vera, P_2 : Garlic extract, P_3 : Neem extract, P_4 : Propolis

The highest (9.54 mg/100 g) lycopene content was recorded in propolis (P_4) treated fruits followed by aloe vera (8.92 mg/100 g), neem (8.76mg/100 g), garlic (8.07mg/100 g) and lowest (5.53 mg/100g) lycopene content was recorded in controlled (P_0) fruits (Figure

17). Carotenoids are very potent natural antioxidants. Lycopene is considered as the mightiest agent against the risk of this type of tumor, in certain in its more lethal form (Giovannucci, 2002).

Further vindication supporting the above findings has been delivered by the recent meta-analysis of the experiments on the role of tomato products and lycopene in the prevention of prostate cancer (Etminan *et al.*, 2004). Mainly lycopene is the pigment responsible for the characteristic deep red colour of ripe tomato fruits. During ripening chlorophyll degradation and increased lycopene synthesis results in the characteristic colour development in tomatoes (Yadav *et al.*, 2009).

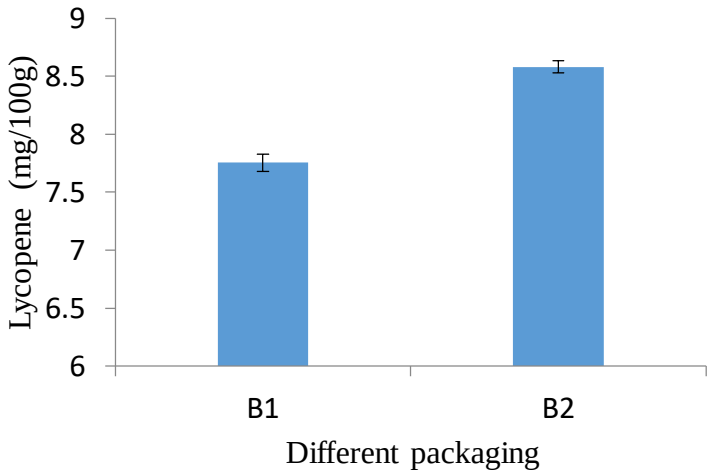

Figure 18: Effect of different packaging on lycopene (mg/100g) of tomato at the end of shelf life
B_1: No packaging, B_2: Packaging

Postharvest biopreservatives formed a protective layer on the fruit skin and kept the fruit temperature low which resulted lower respiration rate. So, the coated fruits were in good condition at the 15 days storage. On the contrary, as fruit temperature was high in control untreated fruits, they could not maintain lycopene content and get spoiled earlier. According to Ayala-Zavala (2008), herbal extracts could extend the shelf life of tomatoes and could maintain or enhance the lycopene, ascorbic acid and total phenolic compounds. The maximum (8.58 mg/100 g) lycopene content was observed in packaging conditioned fruits and minimum (7.75 mg/100 g) lycopene content was observed in no packaging

conditioned fruits (figure 18). Polythene packaging could be helpful to retain colour of preserved fruits. It is supported by Badshah *et al*. (1997).

It was recorded that highest (10.11 mg/100 g) lycopene content was observed in P_4B_2 (Propolis treated fruits in packaging) combination and lowest (4.39 mg/100 g) value was recorded in P_oB_1 (Controlled fruits in no packaging) combination (Table 9).

Table 9. Combined effect of different biopreservatives and postharvest packaging on the lycopene content (mg/100g) of tomato at the end of shelf life

Treatments	Lycopene content (mg/100g)
P_0B_1	**4.39 d**
P_0B_2	6.67 c
P_1B_1	8.76 ab
P_1B_2	9.09 ab
P_2B_1	8.03 bc
P_2B_2	8.12 bc
P_3B_1	8.61 ab
P_3B_2	8.92 ab
P_4B_1	8.98 ab
P_4B_2	**10.11 a**
LSD(0.01)	1.62
SE	0.57
CV (%)	8.47

P_0: Control, P_1: Aloe vera, P_2: Garlic extract, P_3: Neem extract, P_4: Propolis, B_1: Non Packaging, B_2: Packaging; Means with different letters significantly differ at LSD's test at $P \leq 0.01$; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference.

CHAPTER V

SUMMARY AND CONCLUSIONS

5.1 Summary

The experiment was carried out at the Postharvest Laboratory of Department of Horticulture, Sher-e-Bangla Agricultural University, Dhaka during the period from June to August, 2019. The objectives of the present study were to investigate the effect of different packaging and biopreservatives on shelf life of tomato cv. Roma and to evaluate the quality parameters of tomato fruits after storage. In this two factorial experiment preservatives were denoted as Factor A and different packaging was denoted as Factor B. Four different postharvest preservatives used in this study are: i) 100% Aloe vera extraction (P1), ii) Garlic extract solution (P2), iii) 40% Neem solution (P3), iv) 10% Propolis solution, untreated fruits marked as Control (P0) and two different packaging such as i) No packaging condition (B_1) ii) Packaging condition (B_2) were used in this experiment. The experiment was laid out in Completely Randomized Design (CRD).

In this experiment observations were made on external and internal fruit characteristics, physiochemical attributes such as total weight loss, pH, total soluble solid content, Ascorbic acid, Lycopene content, B-carotene content, Visual scoring of tomato skin on the basis of shrinkage severity and shelf life. In this research work tomato of each treatments were collected randomly at three, six, nine, twelve and fifteen days after harvest for physiochemical studies. The data were statistically analyzed and demonstrated. The results of the experiment evolved that almost all the parameters studied were significantly subjugated by the above factors.

Total ten postharvest treatments were used in this experiment with control. Among all those treatments the highest total weight loss (3.48%, 5.60%, 7.87%, 10.24% and 13.94% at 3^{rd}, 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) was observed in controlled fruits (P_0) and the lowest value (1.79%, 3.59%, 4.87%, 6.47% and 7.51% at 3^{rd}, 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) was noticed in propolis treated fruits (P_4). pH was found to be the highest (5.28) at the end of shelf life in untreated fruits (P_0) whereas propolis coating (P_4) represented the lowest

value (4.5). TSS value was mostly influenced by propolis (P_4) to keep its peak the lowest level (3.1%) and the highest value (4.21%) was obtained by untreated controlled fruits (P_0). TA value which was an important quality parameter of tomato showed the maximum value (0.67%) of titrable acidity for tomatoes was recorded for propolis (P_4) followed by Aloe vera (P_1), the value was (0.61%) and minimum value (0.38%)was recorded for untreated control fruits (P_0). Ascorbic acid content was recorded to be the maximum value (13.66 mg/100 g) was recorded for propolis (P_4) treated fruits at the end of shelf life where controlled treatment (P_0) represented the lowest ascorbic acid content (9.10 mg/100g). However, 10% propolis (P_4) treated fruits represented the highest β-carotene content (0.346mg/100 g) and controlled fruits represented lowest (0.069 mg/100g) β-carotene content. The maximum (39.385% at 15^{th} DAS) shrinkage severity was recorded in P_0 (Controlled fruits) and minimum (11.25% at 15^{th} DAS) value was noticed in P_4 (Propolis treated fruits). The highest (9.54 mg/100 g) lycopene content was recorded in propolis (P_4) treated fruits followed by aloe vera (8.92 mg/100 g), neem (8.76 mg/100 g), garlic (8.07mg/100 g) and the lowest (5.53 mg/100g) lycopene content was recorded in controlled (P_0) fruits. Among the treatments, the highest shelf life of tomato (15 days) was belonged to 10% propolis (P_4) and the lowest (5.33 days) shelf life was recorded in controlled fruits (P_0).

Total weight loss (3.68%, 6.29%, 8.49%, 10.75% and 12.59% at 3^{rd}, 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS), pH value (5.08), TSS (3.63%) and shrinkage severity (28.365% at 15^{th} DAS) was found to be the highest in B_1 (no packaging condition) and the lowest weight loss (1.67%, 2.91%, 4.54%, 6.37% and 8.13% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS) , pH value (3.82), TSS (3.36%) and shrinkage severity (23.018%) was recorded in B_2 (packaging condition). On the other hand, the highest value of TA (0.56%), Ascorbic acid (11.91 mg/100g), shelf life (11.467 days) was found in packaging condition (B_1) and lowest TA (0.50%), Ascorbic acid (10.11mg/100g), shelf life (10.4 days) was found in no packaging condition. However, (8.58mg/100g) lycopene content was observed in packaging conditioned fruits and minimum (7.75mg/100g) lycopene content was observed in no packaging conditioned fruits. In case of β-carotene content the highest (0.205 mg/100g) was observed in packaging condition and the lowest (0.114 mg/100 g) β-carotene content was noticed in no packaging condition fruits.

The combined effect between the postharvest biopreservatives and different packaging were found that maximum (4.82%, 7.72%, 9.86%, 12.53% and 16.86% at 3rd, 6th, 9th, 12th and 15th DAS) rate of weight loss, pH value (5.53) was observed in P_0B_1 (Controlled fruits in no packaging condition) combination and minimum weight loss (0.22%, 0.54%, 0.54%, 1.06% and 1.06% at 3rd, 6th, 9th, 12th and 15th DAS), pH value (4.46) was recorded in P_4B_2 (Controlled fruits in no packaging condition) combination. In case of β-carotene and lycopene content maximum (0.53 mg/100g) and (10.11 mg/100g) was observed in P_4B_2 and the minimum β-carotene and lycopene content (0.05 mg/100 g) and (4.39 mg/100 g) value was recorded in P_oB_1. Again the significant effect of treatments on TSS gave the maximum value (4.40%) in P_0B_1 and minimum (3.03%) in P_4B_2.

In the present study, the highest (0.71%) TA value was recorded in P_4B_2 and the lowest (0.34%) value was noticed in P_0B_1 combination. In case of interaction effect the maximum (14.23 mg/100 g) value of Ascorbic acid was observed in P_4B_2 and minimum (8.20 mg/100 g) in P_0B_1 combination. In case of shrinkage severity maximum value (45.13% at 15th DAS) was determined in P_0B_1 and the lowest value (8.25% at 15th day) was observed in P_4B_2. Combined effect revealed that the highest shelf life of tomato was recorded in all biopreservatives treated fruits kept in packaging condition and the lowest (5.33 days) was found in controlled unpacked fruits P_0B_1 combination.

5.2 Conclusion

In an endeavor to maintain the freshness and quality of tomato fruits, propolis supposed to be the grand bio-preservative. It successfully decreased weight loss, delayed ripening, locked up moisture, and checked the pH, TA, Ascorbic acid content of tomato. Moreover, no skin shrinkage was appeared. So, it can be concluded that propolis in packaging condition (P_4B_2) is the best for long time storage, home consumption and the possibility of use in processing industry.

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APPENDICES

Appendix I: Effect of postharvest biopreservatives and different packaging on weight loss (%) of tomato at different days after storage (DAS)

Sources of variation	Degrees of freedom	Mean square of weight loss at different days after harvest				
		3	6	9	12	15
Replication	2	0.022**	0.344**	0.111**	0.137**	0.473**
Factor A	4	2.317**	4.116**	8.36**	13.373**	33.735**
Factor B	1	30.410**	85.618**	117.456**	143.602**	148.945**
AB	4	0.8218**	1.173**	2.231**	1.737**	3.247**
Error	11	0.0387**	0.221**	0.210**	0.177**	0.499**

**Significant at 1% level of significance

Appendix II: Effect of postharvest biopreservatives and different packaging on shelf life

Sources of variation	Degrees of freedom	Mean square of Shelf life
Replication	2	0.4000**
Factor A	4	40.4167**
Factor B	1	38.5333**
AB	4	1.2833**
Error	11	0.2148**

**Significant at 1% level of significance

Appendix III: Effect of postharvest biopreservatives and different packaging on shrinkage severity (%) of tomato at the end of shelf life

Sources of variation	Degrees of freedom	Mean square of shrinkage severity%
Replication	2	1.468**
Factor A	4	609.136**
Factor B	1	214.455**
AB	4	20.959**
Error	11	1.030**

**Significant at 1% level of significance

Appendix IV: Effect of postharvest biopreservatives and different packaging on Lycopene content (mg/100g) and pH of tomato at the end of shelf life

Sources of variation	Degrees of freedom	Mean square at the end of shelf life	
		Lycopene	pH
Replication	2	0.2817**	0.00992**
Factor A	4	14.6808**	0.53534**
Factor B	1	5.1191**	.51745**
AB	4	1.2344**	0.04139**
Error	11	0.4788**	0.01578**

**Significant at 1% level of significance

Appendix V: Effect of postharvest biopreservatives and different packaging on TSS, TA, ascorbic acid and β-carotene of tomato at the end of shelf life

Sources of variation	Degrees of freedom	Mean square at the end of shelf life			
		TSS	TA	Ascorbic acid	β-carotene
Replication	2	0.01213**	0.01483**	0.0408**	0.00005**
Factor A	4	1.19262**	0.07820**	16.7299**	0.07021**
Factor B	1	0.53333**	0.02803**	23.9467**	0.06107**
AB	4	0.06917**	0.00897**	0.2708**	0.03380**
Error	11	0.04783**	0.00483**	0.0379**	0.00007**

**Significant at 1% level of significance

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