EFFECT OF POSTHARVEST STORAGE TEMPERATURES AND BIOPRESERVATIVES ON QUALITY OF MANGO

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DECEMBER, 2019

EFFECT OF POSTHARVEST STORAGE TEMPERATURES AND BIOPRESERVATIVES ON QUALITY OF MANGO

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REGISTRATION NO.13-05383

A Thesis

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

IN

HORTICULTURE

SEMESTER: JULY-DECEMBER, 2019

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Dedicated to

My Beloved Parents

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

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CERTIFICATE

This is to certify that thesis entitled, "EFFECT OF POSTHARtfEST STORAGE TEMPERATURES AND BIOPRESERtfATItfES ON QUALITY OF MANGO" 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 ROMANA CHOWDHURY, Registration No. 13-05383 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.

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ACKNOWLEDGEMENTS

All praises to the ***"Almighty Allah"*** *who allow me to complete a piece of research work and prepare this thesis for the degree of Master of Science (M.S.) in Horticulture.*

I feel much pleasure to express my gratefulness, sincere appreciation and heartfelt liability to my venerable research supervisor ***Prof. Dr. Md. Nazrul Islam****, Department of Horticulture, Sher-e-Bangla Agricultural University (SAU), Dhaka- 1207, Bangladesh for his scholastic guidance, support, encouragement and valuable suggestions and constructive criticism throughout the study period.*

I also express my gratitude, gratefulness and thankfulness to reverend co-supervisor, Associate Professor ***Dr. Shormin Choudhury,*** *Department of Horticulture, Sher-e-Bangla Agricultural University (SAU), Dhaka-1207 for her constant inspiration, valuable suggestions, cordial help, heartiest co-operation and supports throughout the study period.*

It is also a boundless pleasure for the author to express her cordial appreciation and thanks to all ***respected teachers*** *of the Department of Horticulture, Sher-e-Bangla Agricultural University, for their encouragement and assistance in various stages towards completion of this research work.*

The author deeply acknowledges the profound dedication to her beloved ***Father, Mother, Sisters and other family members*** *for their moral support, perseverance and continuous prayer in all phases of academic pursuit from the beginning to the completion of study successfully.*

Finally, the author is deeply indebted to her friends and well-wisher specially ***Alifa Yasmin****,* ***Nayema Islam*** *and* ***Kazi Nawrin Antora*** *for their kind help, constant inspiration, co-operation and moral support which can never be forgotten.*

The Author

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ABSTRACT

BY

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Mango is one of the most economically important fruit facing greater problems in storage because of its perishable nature and reduction of quality. Application of biopreservatives is a key step to improve its quality and shelf life. An experiment was conducted to study the effect of different biopreservatives on ripening behavior, physiological changes of mango cv. 'Amrapali' under different storage temperatures. This study was carried out at the Postharvest Laboratory of Department of Horticulture, Sher-e-Bangla Agricultural University, Dhaka. Five postharvest preservatives e.g. P_0: Control, P_1: Aloe vera extract, P_2: Beewax emulsion, P_3: Neem leaf extract, P_4: Chitosan solution and three different temperatures e.g. T_1: , T_2: , T_3: were used in this present study. The two factorial experiment was laid out in completely randomized design (CRD). It was revealed that application of aloe vera and beewax reduced percentage of weight loss, pH, shrinkage severity, disease severity, increased moisture content, titratable acidity, total soluble solid (% Brix), ascorbic acid content, beta carotene, and prolonged shelf life compared to untreated control fruits. In line with this, combined application of biopreservatives under different temperatures disclosed that shelf life of aloe vera treated fruits in was 45 days. Considering the appearance, quality as well as shelf life Aloe vera in (P_1T_1) being the most effective treatment on all the parameters tested.

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LIST OF ACRONYMS

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

CHAPTER I

INTRODUCTION

Mango (*Mangifera indica* L.) is known as the king of fruits, prominent flavour with durable aroma and contains high amount of vitamin A and C, beta-carotenoids and trace amount of minerals and vitamins (Chauhan *et al*., 2014). In addition, mangoes are also very tasty, mushy and refreshing. Bangladesh is the world‟s eighth largest mango producing country and it accounts for about 4% of the world total mango production (Rahman and Khatun, 2018). Large varieties of fruits are grown in Bangladesh, but Amrapali is the most popular among them because of their high yield, good flavor, attractive color and good aroma. It is an important tropical fruit however, it is susceptible to a number of biotic and abiotic stresses that leads to rapid deterioration and large postharvest losses, estimated to be over 45% in some developing countries. Fungal disease is one of the most important causes of postharvest damages (Khewkhom and Shangchote, 2009). Furthermore, because of their short shelf life, quite high percentage of fruit is wasted and spoiled. Therefore, it is necessary to explore the ways and means to prolong the shelf life of the fruit while keeping the quality high.

To prevent the postharvest losses, the susceptible fruits are treated with synthetic chemicals which have toxicity effect; some are carcinogenic and cause environmental pollution. Thus, to find an alternative way for synthetic chemicals, biological components are being used which are nontoxic, specific in their action and safe to environment and for the living beings (Abirami *et al*., 2013). To overcome this rapid loss of fruits or other foods, biopreservatives are novel food preserving compounds which help to enhance food safety (Ergun and Satici, 2012). The coating technology is one of the important and well known techniques used to prolong the shelf life and reduce their wastage. A number of edible coatings have been used and discussed by the scientists and efforts are still going on to find the best one. Therefore, the chitosan, carnauba wax, aloe vera gel, shellac, polysaccharid based coating materials have been used by the scientists, and their efficiency and the problems correlated with them have been highlighted (Zhu *et al*., 2008, Abbasi *et al*., 2011). These compounds do not have side effects and due to presence of antimicrobial compounds, increases the food quality and storage period (Ashwini and Desai, 2018). Bioactive products of plant are less persistent in environment and are safe for humans and other non-targeted organisms.

Aloe vera gel based biopreservatives has exhibited to prevent loss of moisture and firmness, control respiratory rate and maturation development, delay oxidative browning and reduce microorganism proliferation in fruits (Kumar and Bhatnagar, 2014). Beeswax contains triacontanol as the main constituent, which is antioxidant, antiperoxidative, anti-inflammatory, gastroprotective and ant colitis; hence, it has good potential for coating. Chitosan is a modified natural carbohydrate polymer procured from chitin which has been found in a wide range of natural sources such as crustaceans, fungi, insects and some algae (Tolaimate *et al*., 2000) and is used in industrial products as a bioactive material (Cho *et al*., 2008). It inhibits the growth of a wide variety of bacteria (Sudarshan *et al*., 1992) and fungi (Stössel and Leuba, 1984). Chitosan is well known natural coating material used in several fruits for prolonging their shelf life (Graham, 1990). Neem (*Azadirachta indica*) extract has been used for centuries in Asia as insecticides and fungicides (Chaturvedi *et al*., 2003). Azadirachtin is regarded as the most active substance in neem which has growth regulating, fungicidal, and insecticidal properties (Schmutterer, 1990) with minimal impact on non-target organisms and is compatible with other eco-friendly biocontrol agents (Srivastava, 2003). Temperature is an important factor that affects fruit quality, appearance and shelf life (Nunes *et al.*, 2007). Proper temperature management during handling and storage is essential to delay ripening, preserve the fresh-market quality and extend shelf life. Low temperature storage is the most commonly adopted method to extend the shelf life of mangoes, although postharvest losses due to chilling injury have been reported (Rodov, 1996).

Nowadays, biopreservatives are widely used throughout the world for its excellent quality. However, in Bangladesh, there is limited information and experience to use biopreservatives as postharvest treatment to extend the shelf life of mangoes. Therefore, the present study was undertaken to fulfill the following objectives:

i) To investigate the effect of different storage temperatures and biopreservatives on shelf life of mango

ii) To evaluate the quality parameters of mango fruits after storage

CHAPTER II

REVIEW OF LITERATURE:

Mango being a highly perishable fruit contains a very short shelf life and reach to respiration peak of ripening process on 3rd or 4th day after harvesting at ambient temperature (Narayana *et al*., 1996). The shelf life of mango varies among its varieties basing on storage conditions. It extends from four to eight days at room temperature and 2-3 weeks in cold storage at 13ºC (Carrillo *et al*., 2000). Normally after harvesting, the ripening process in mature green mango takes 9-12 days. The ripening process of mango fruit involves a series of biochemical reactions thus lead to ripening of fruit with softening of texture to admissible quality (Herianus *et al*., 2003). Consumers around the world demand for food of high quality, without chemical preservatives, and a prolonged shelf life. Therefore, during recent years, global concern for safety of the environment has led researchers to investigate the use of natural flora as one of the sources of treatments for crop protection and the present investigations is also an effort in this direction. The relevant literature pertaining to the effect of such substances on the storage quality of mangoes and some other stuffs is reviewed under the following headings:

Effect of biopreservatives on quality and performance of Mango:

Biopreservatives could be defined as compounds, from natural sources or formed in food, able to restrict or retard spoilage related with chemical or biological deterioration that prolong product shelf life. Edible coatings are thin layers of edible substances applied to the product surface in addition to or as a replacement for natural protective waxy coatings and provide a barrier to moisture, oxygen and solute movement for the food (Avena-Bustillos *et al*., 1997 and Mchugh and Senesi, 2000). They are used directly on the food surface by dipping, spraying or brushing (Mchugh and Senesi, 2000). Edible coatings are used to create a modified atmosphere and to decrease weight loss during transport and storage (Baldwin *et al*., 1995). In fact, the barrier features of gas exchange for films and coatings are the subjects of much recent interest (Sumnu and Bayindirli, 1994).

Effect of Aloe vera:

Aloe vera is a tropical and subtropical plant that has been used for centuries for its medicinal and therapeutic characteristics (Eshun *et al*., 2004). There are some reports on the antifungal activity of Aloe vera gel against various pathogenic fungi including *Botrytis cinerea* (Jasso de Rodriguez *et al*., 2005; saks and Barkai-Golan, 1995). There has been increasing interest in the use of Aloe vera gel in the food industry as a functional component (Moore *et al*., 2005). Aloe vera based edible coatings have been shown to restrict loss of moisture and firmness, control respiration rate and maturation development, delay oxidative browning, and reduce microorganism proliferation in fruits such as sweet cherry, table grapes and recterones (Matinez-Romero *et al*., 2005). Aloe vera possesses phenol, saponin, anthraquinones components, have anti-bacterial, antiviral and antifungal properties. Aloe vera leaves are affluent in bioactive compounds some of which are antioxidants those are broadly used in food engineering as preservative such as mannans, antrachinon, cglycoside, antron, antrakuinon and lectine (King *et al*., 1995; Eshun and He, 2004). Aloe vera has shown antibacterial characteristics against gram positive and gram negative pathogens (Adetunji, 2008). *Aspergillus, Fusarium* and *Penicillium* are fungal kinds which are responsible for oxidation and spoilage of food (Babaei *et al*., 2013).

Recently, researchers from Spain have developed a gel based on Aloe vera that augments the conservation of fresh fruits (Tripathi and Dubey,2004).This natural product is a safe and environmentally amicable alternative to synthetic preservatives such as sulfur dioxide.

Aloe vera extracts were stated to be useful for “Kensington Pride” mangoes (Dang *et al*., 2008) and “Artic Snow” nectarines (Ahmed *et al*., 2009) for retaining quality losses after harvest.

Maintaining sweet cherry quality using Aloe Vera Coating was effective to decrease weight loss and lower respiration rate during postharvest storage (Romero *et al*., 2006).

Vahdat *et al*. (2010) found that coating fruits with Aloe vera remarkably reduced weight loss as compared to the control. The minimum weight loss was moticed in fruits coated with 100% (v/v) and minimum firmness was achieved with the control at the end of storage. Also treated fruits have displayed higher titratable acidity, sugar content and ascorbic acid than untreated fruits.

Ergun and Satici (2012) used aloe vera gel as biopreservative to study effects of Aloe vera gel (0, 1, 5 and 10% w/v) coating on green-coloured “Granny Smith” and red-coloured “Red Chief” apples those were kept at 2 °C for 6 months. Aloe vera gel treatments substantially suppressed the rise in weight loss for “Granny Smith” apples but did not affect weight loss for “Red Chief” apples. Apples from both cultivars softened at certain rates over time, and these rates were not affected from any of the gel treatments. Aloe vera gel treatment restricted the green colour loss for “Granny Smith” but remained unaffected for “Red Chief” apples. Soluble solids content and percentage of titratable acidity was noted higher for “Granny Smith” apple fruit treated with Aloe vera gel (5 and 10%) during most of the storage period while no Aloe vera gel effects on colour for “Red Chief” apples was noted. The pH values for “Granny Smith” fruit slightly reduced while slightly increased for “Red Chief” fruit over time, yet values for both cultivars remained unaffected by Aloe vera gel treatments. The results exhibited that Aloe vera gel treatment may be used as bio preservative on “Granny Smith” apples for retarding quality losses.

An investigation was conducted by Munira *et al*. (2016) with postharvest mangoes (cv. Amrapali) treated with different level of aloe vera and chitosan under room temperature in respect of size of fruits. The treatment concentration had remarkable variation. Maximum shelf life (7.00 days) found with Aloe vera gel treatment compared with Chitosan and no coating Control treatment. With the concentration of Aloe vera and Chitosan, shelf life and disease-pest incidence of fruits vary remarkably. 1.5% Aloe vera gel treatment was best pursued by 1.5% Chitosan. Disease and pest incidence also alters accordingly. Among the other treatments, 1.0% Aloe vera and 1.0% Chitosan was statistically same. Semperfresh, aloe vera gel (50 and 100%) and mango carnauba-based coatings effectively slow down mango ripening. However firmness, soluble solids and total carotenoids were not maintained by the coatings.

Ochiki *et al*. (2014) reported that mango is a highly perishable fruit and high postharvest losses occur in Africa. In order to address this problem, 4 concentrations of Aloe vera gel (AG) (0, 25, 50 and 75%) and chitosan (1%) were tested at two temperature levels (room temperature 15-22°C and 13°C) to determine their impact on the postharvest life of mango (var. „Ngowe"). The experimental design was a 5 by 2 factorial experiment attached in a complete randomized design with three replications. It was found that at both temperatures 50 and 75% Aloe vera concentrations remarkably increased the shelf life and decrease in titrable acidity. Fruit color and ascorbic acid were also maintained for prolonged periods in these treatments. Findings of this study demonstrate the potential of using Aloe vera gel at 50% as a coating for improved postharvest shelf life and maintaining quality of mango fruits hence decreased postharvest losses.

Effect of Beewax:

Waxes and edible coatings can be regarded as varieties of modified atmosphere. Biopreservatives are used to develop the fruit‟s external appearance and to alter gas permeability, decrease weight loss and delay ripening (Banks *et al*., 1993). The surface coating is semipermeable, reducing gas diffusion and respiration (Banks *et al*., 1993; Dhall, 2013). Thus, fruit coating slows down ripening by lowering O_2 and increasing CO_2 concentration and hence, prolongs the storage life of mango fruit. In most cases, edible coatings are environmentally amicable and are used as an alternative to film packaging. Fruit coating is also less expensive than controlled or modified atmosphere technology (Baldwin, 2005).

Wax coatings are usually emulsions of synthetic polyethylene or natural carnauba wax, beeswax and others. The integration of both polysaccharide and protein based substances enhances the functionality of the coating (Gol *et al*., 2013). Surface coatings possessing synthetic waxes, natural waxes (carnauba and beeswax) and resins (shellac) limit water loss better than those containing polysaccharides (Amarante and Banks, 2001). Productions based on shellac result in a shinier appearance than those based on carnauba wax or polysaccharides (Hoa and Ducamp, 2008). Coating „Tommy Atkins" mango with carnauba wax and beeswax decreased water loss and chlorophyll loss, CI and decay after cold storage (Feygenberg *et al*., 2005). However, improper fruit waxing can lead to unwanted effects on fruit quality, including anaerobic respiration and development of off-flavours (Amarante and Banks, 2001).

Abonesh *et al*. (2019) carried out an experiment on the effect of beewax and chitosan on the quality and shelf life of mango cv. „Tommy Atkins" and „Apple." They used beewax and chitosan at various concentrations (0.5%, 1.5% and 2%), and two mango varieties (Apple and Tommy Atkins). Application of beewax and chitosan at (2%), significantly decreased physiological weight loss (%), Total Soluble Solidity (Brix), Titratable Acidity (%), pH, disease incidence (%), disease index (%), maintained Firmness (N) and extended shelf life of fruits compared with untreated control. It was decided that edible coatings used in the present study have a good potential in maintaining the fruit quality and beewax at 2% being the most e□ ective treatment on all parameters tested. After beewax, chitosan displayed better result.

However, improper fruit waxing can lead to unwanted effects on fruit quality, including anaerobic respiration and development of off-flavours (Amarante and Banks, 2001).

Hossain *et al*. (2001) studied the physio-chemical composition of three types of mango. The best fruit weight was lowest (221.33 gram) in Amarpali but the types of Bishawanath had the maximum fresh weight (256.0 gram) and keeping quality (8.75 days). The most keeping quality was in Amarpali (12.5 days) mango fruit. The TSS (23.50 percent), total sugar (26.85 percent) and pH of pulp (6. 0) were maximum in Amarpali, whereas Bishawanath indicated maximum Vitamin C (14.20 mg /100g) and acidity (titrable) (0.87 %). Amarpali fruit was better in respect of all properties as compared to other varieties.

Molla *et al*. (2011) studied the postharvest changes in mango and reported that color and quality of mango was very better in treated fruits compared to non-treated fruits.

Effect of Neem:

Shrestha *et al*. (2018) studied on effects of various plant leaf extracts on postharvest life and quality of mango (*Mangifera indica* L.).Freshly harvested mature green mangoes cv. 'Calcuttia maldah' of identical size and weight were dipped in 50% concentration of different plant leaf extracts and stored in ambient condition (32±2ºC and 65±5 % RH). The treatments were leaf extracts from five various plants viz. neem (Azadirachta indica), chinaberry (*Melia azadirach*), lantana (*Lantana camara*), ashok

(*Polyalthea longifolia*) and cinnamomum (*Cinnamomum zeylanicum*) while control was the other treatment. Additionally, carbendazim (fungicide) was also kept as a benchmark treatment. Each treatment comprised of 5 mangoes and replicated thrice. For each replication destructive specimen was also kept. The treatment with neem leaf extract showed the most promising result as there was minimum physiological weight loss, maximum ascorbic acid content, maximum acidity and minimum pH. Similarly, shelf life, total soluble solids, freshness and firmness were maximum in neem leaf extract treated fruits next to the carbendazim treated fruits. Control was the most void of all the treatments regarding all the parameters.

Shindem *et al*. (2009) studied the influence of different plant extract treatments to increase the shelf life and to minimize the postharvest losses in mango. Among the fruits treated with various plant extracts and wrapping materials, 10 per cent neem oil has been proved to be most effective in slow increase of TSS and slow decrease of ascorbic acid and acidity during storage.

Effect of Chitosan:

Chitosan has been successfully used recently as a food wrap due to its film-forming properties (No *et al*., 2007). Chitosan is a high molecular weight cationic polysaccharide achieved from alkaline deacetylation of chitin, a homopolymer of β-(1-4)N-acetyl-D-glucosamine, which is commercially extracted from shrimp and crab shells. Chitosan coatings are particularly promising due to their biocompatibility, biodegradability, non-toxicity and antimicrobial properties (No *et al*., 2007; Toan, 2009).

Chitosan is capable of inactivating or inhibiting various enzymes which cause deterioration in fruits and vegetables (BhaskarReddy *et al*., 2000; Bautista-Baños *et al*., 2006; Gonzalez-Aguilar *et al*., 2008). Modified atmosphere packaging and refrigeration are not sufficient to completely avoid tissue browning. Chitosan coatings were able to reduce browning in fruits by decreasing polyphenol oxidase and peroxidase activities (Zhang and Quantick, 1998). This effect was directly related to the modification of the internal atmosphere in the fruit, with reduced levels of O_2 and increased levels of CO_2.

Tassadit *et al*. (2010) carried out a study to observe the effect of chitosan on quality of mango (*Mangifera indica* cv. „Tommy Atkins"). Mango fruits were coated with chitosan solution (0.25% w/v) dissolved in 0.5% (w/v) citric acid, and stored for 9 days at 20°C under surrounding atmosphere. This study showed that chitosan coating, either alone or mixed with hot water, did not affect the taste and the flavour of mangoes. The chitosan coating mixed with hot water dipping or not inhibited the microbial growth for nine days at 20°C.

Sharmin *et al*. (2018) studied the impact of plant extract on shelf life of mango cv. „Amrapali" where neem leaf extract was used. Freshly harvested mango was treated with various concentrations (20% and 40%) of neem leaf and banana pulp extract alone or in combination. All treated and untreated mangoes were stored at room condition. Among the treatments, neem leaf extract at 20% and neem leaf extract 40% + banana pulp extract 40% treatments displayed longer shelf life (9.92 and 10.25 days, respectively), slower changes in color (score 2.77 and 2.93, respectively) and firmness (score 2.67 and 2.77, respectively); less disease severity (score 2.93 and 3.57, respectively), disease incidence (46.67% and 60.00%) and lower loss in weight (38.04% and 35.17%, respectively) at 9 DAT (Days after treatment).

Effect of Different Temperature:

A temperature range of 7-13ºC, basing on mango cultivar, growing conditions, stage of maturity and postharvest handling techniques have been suggested for mango fruit storage (Kalra *et al*., 1995). The lowest safe temperature for mango storage has been stated to be 7.2ºC (Sundaraj *et al*., 1972). However, Julie and Ceylon mango cultivars are stated to store well at 7ºC for 3-4 weeks and normal fruit ripening occurs (Wardlaw and Leonard,1936). Unripe physiologically mature (green) mangoes do not ripen uniformly when kept at 10ºC for 3 weeks, while ripe and partially ripe fruits kept well up to 3 and 6 weeks, respectively (RamaAyyer and Joshi,1929). It is also stated that mango fruits stored for 3 weeks at 5ºC or one week at 1ºC did not ripen well (Chaplin *et al*.,1991). Ripe mango fruit can be kept at 7.2ºC while unripe fruit should not be kept at temperatures below 10ºC (Akamine,1963).

Cecilia *et al*. (2006) carried out a study on quality curves for mango fruit (cv. Tommy Atkins and Palmer) stored at chilling and non-chilling temperature. In this study they kept the harvested fruits at 2 C, 5 C, 2 C, 5 C and 20 C for 7-20 days. Chilling injury and enhanced fruit softness were the limiting quality factors for mango stored at 2 and 5 C. Softening of the fruit, changes in color and improvement of decay were the limiting quality factors for mango stored at 2, 5 and 20 C. But from overall quality characteristics 20 C give the best result as the weight loss, color, firmness, vitamin C content, beta carotene, moisture content level was good for 14 days of storage.

Effect of Postharvest Treatments on Physico-Chemical Changes

Weight loss is an essential index of ripening. With the advancement of ripening and conversion of starch to sugar weight reduces (Popy *et al*., 2013) showed the behavior of mango (*Mangifera indica*) cv. Amropali and stated that the treatment of neem and garlic extract showed the minimum physiological weight loss when compared to other treatments and controls. Brishti *et al*. (2013) carried out an experiment on effect of bio preservatives on storage life of papaya fruit where Aloe vera gel (100%) had been used to preserve papaya fruit at room temperature 25°C-29°C and 82-84% relative humidity. All specimens demonstrated a gradual loss of weight during storage. Throughout storage, the weight loss of uncoated fruit (sample) was remarkably greater than that of Aloe gel coated fruit. At the end of the storage, uncoated papaya displayed 22.5 % loss in weight, but the weight losses of samples coated with Aloe vera gel was 7.93%. Tripathi and Dubey (2004) conducted an experiment to maintain quality and safety of table grapes by coating with Aloe vera gel in cold storage (1°C, 95%). Weight loss enhanced during cold storage and it was remarkably greater in control (uncoated fruits) than in Aloe-coated grapes. At the end of cold storage, control fruits lost 15.51 ± 0.32%, but the loss of weight in Aloe-treated grapes was 8.13 ± 0.59%. Carrillo *et al*. (2000) who found that coated or uncoated Haden mango in Mexico had an increasing trend of weight loss with the passage of storage time. Togrul and Arslan (2004) stated that the coating helps to reduce moisture loss and gaseous exchange from the fruits due to formation of a film on the top of the skin acting as an additional barrier. Similar results were reported by Thai *et al*. (2002) who showed that wax coating reduced the rate of respiration and transpiration and resulted in reduced weight loss, shriveling and increased shelf-life.

The works by Chien *et al*. (2007) and Zhu *et al*. (2008) reveal that applying a chitosan coating e□ ectively enhances the quality attributes and extends the shelf life of mango fruit. Baldwin *et al*. (1999) also observed that the carnauba wax coating significantly decreased water loss compared to uncoated and polysaccharide-coating treatments of mango fruits. Srinu *et al*. (2017) carried out an experiment on effect of postharvest treatment on quality and shelf life of papaya. They observed minimum physiological weight loss PLW (14.97%) at 15 days of storage was noted in fruit treated with 5% wax treatment at normal ambient temperature. The wax coating lowered down the rate of respiration, transpiration, decay and reduced the enzymatic activates responsible for disorganization of cellular structure, thus, delay senescence and thereby, reduce weight loss. Reddy and Raju (1988) conducted an experiment on mango fruit cv. Alphanose. They stated an average 3.96% weight loss in "Alphonso" mango stored at ambient temperature for 5 days compared with 3.9 and 3.7% weight loss in "TommyAtkins" or "Palmer" mangoes from the study conducted by Cecilia *et al*., stored at 20 C for 5 days.

Singh *et al*. (2000) stated that dipping of mango fruit cv. Langra in neem leaf extract (10%) significantly reduced the moisture loss as compared to control, where the moisture loss was higher. Kader *et al*. (1989) studied the effect of GA_3 and plant extracts (Neem leaf extract, castor oil and neem oil) on storage behaviour of mango (*Mangifera indica*) cv. Langra and stated that treatments of neem leaf extract (100%) showed minimum physiological loss in weight (7.77%) as compared to other treatments and controls in which maximum physiological loss in weight (17.28%) on the 12^{th} day of storage was stated. Neem leaf extract was also found better in decreasing fruit spoilage.

Bhardwaj and Sen (2003) studied the effect of neem leaf extract (10% and 20%) on mandarin (*Citrus reticulate Blanco*) cv. Nagpur Santra and stated that fruits treated with 20 per cent neem leaf extract significantly reduced the physiological weight loss (9.43%) and reduction in diameter of fruits (11.54%) whereas, under control fruits maximum reduction in fruit diameter (25.35%) and higher PLW (24.17%) was recorded after 42 days of storage. Sindhan *et al*. (1999) stated the beneficial effects of neem, eucalyptus, tulsi, datura, bougainvillea and ginger on the bio-chemical and physical quality characteristics of citrus and mango fruits, as these extracts significantly reduced moisture loss and retained higher soluble solid content over the uncoated fruits. Singh *et al*. (2000) stated that dipping of mango fruit cv. Langra in neem leaf extract (10%) significantly reduced the moisture loss as compared to control, where the moisture loss was high. Togrul and Arslan (2004) stated that the coating helps to reduce moisture loss and gaseous exchange from the fruits due to formation of a film on the top of the skin acting as an additional barrier. Srivastava (1967) stated that the green mango contained higher percentage of moisture as compared to ripe mangoes. Shahajahan (1994) stated that the moisture content of pulp of mature hard 'Fazli' mango was 79.95% but found it as 91% and in ripe mango 78-86%. Salunkhe and Desai (1984) observed that mango pulp possesses 81% moisture. Absar *et al*. (1993) stated that moisture content at the early stage of development varied from 87.4% to 90.1%, gradually decreased as the maturity advanced and at ripening stage it varied from 71.22 to 79.4%. They also observed that the reducing tendency of moisture content with the advancement of maturity of varieties Gopalbagh (82.13 to 79.23%), Khirsapat (82.1 to 79.25%), Langra (81.75 to 78.29%) and Fazli (82.30 to 79.95%). Mollah and Siddique (1973) carried out an experiment with 12 types of mango and found that moisture content of the pulp of all the varieties of mango ranged from 81.03 to 87.12%. They also studied the fruits of ten types of mango. The moisture percentage was the maximum (87.55) in Ranibhog whereas it was the lowest (78.96%) in Misribhog. This trait for the various varieties under consideration ranged from 78.96 to 87.55%.

Fruits are important for the proper maintenance of human health. Fruits are foods affluent in vitamins, minerals and supply arrays of colors, flavor, texture and bulkiness to the pleasure of eating. Tripathi and Dubey (2004) stated that Aloe vera led to a lower rise in TSS (Total Soluble Solid) and greater TA content (Titrable Acidity) retention of coated berries, which indicated that control (uncoated fruits) fruits presented a more pronounced maturation development than coated berries during storage periods (1°C, 95% RH+ 4 days at 20°C, 90% RH). In case of Aloe coated and uncoated oranges (12ºC, 9698% RH), there were no significant variations in TSS and TA content of fruits during storage periods. The value of ascorbic acid content for coated oranges was found to be more than that of uncoated fruits (Arowora *et al*., 2013). Chauhan and Joshi (1990) stated the efficacy of phytoextracts neem on the storage quality of mango cv. Ratna and found them remarkably better in retaining total soluble solids and sugar contents and in reducing reduction in the possible incidence of anthracnose pathogen in comparison to untreated fruits where lower soluble solid and sugar content and higher incidence of anthracnose pathogen was reported. The rise in TSS and sugar content may be due to the hydrolysis of insoluble polysaccharides into simple sugars. Such changes are expected to be slower and more gradual when the metabolism of the commodity is retarded by the application of various coating treatments. Sarmin *et al*. (2018) studied the effect of plant extract on shelf life of mango cv. „Amrapali" where total soluble solid was maximum in neem leaf extract 40% treated fruits with 18.73% more Brix in comparison to control and other treatments. Yonemoto *et al*. (2002) who explained that lower levels of total soluble solids in fruits coated with chitosan may be due to protective oxygen barrier that decreases oxygen supply to the fruit surface which in turn inhibited respiration. Sharafat *et al*. (1990) also found that as storage is prolonged, the rate of respiration, transpiration and other metabolic changes are increased in control fruits in comparison with edible coated mango fruits. Kittur *et al*. (2001) also found similar trends with the present study and stated Chitosan-based coatings were much superior in prolonging the shelf life and quality of banana and mango than polysaccharide-based composite coating formulations. In the case of Cat Hoa Loc mango fruit, the TSS at ripening stage should be minimum 20% to be accepted by the consumer (Hoa and Hien, 2001; Ba, 2007). Tai (2008) had stated that the TSS difference depends on the days after fruit set or density and temperature storage. The rise in TSS was the outcome of conversion of carbohydrates into simple sugars through a complex mechanism during storage, and the conversion rate enhanced with the increase in temperature. The rise in TSS might be due to the alteration in cell wall structure and breakdown in storage. Kittur *et al*. (2001) considered that this conversion was also one of the important indexes of the ripening process in mango and other climacteric fruit. Manzano *et al*. (2001) also found that temperature of storage affects the TSS. Ahmad *et al*. (2001) stated that bananas kept at higher temperatures showed greater TSS than those at lower temperatures.

Xing *et al*. (2012) found that fruits coated with chitosan showed the lowest decrease (about 33%) of TA and in case of control fruits loss of TA is highest.(about 45%). The same effect of chitosan on TA was previously found in pears (Xu *et al*., 2013). The high TA content can be assigned to slower ripening and respiration rates in coated than in uncoated fruits. Chitosan coating controls the availability of O_2 and

CO_2, playing a vital role in inducing the slower ripening rate of pear samples (Perdones *et al*., 2012). Moreover, as organic acids, including citric acid and malic acid, are used as substrates for respiration, a reduction on respiration rate implies higher TA values (Bico *et al*., 2009). High acidity in ripened mango fruit at low temperature has been stated by O'Hare (1995) and Baloch *et al*. (2012).

Fruits are natural sources of ascorbic acid and it is known that their levels reduce during the ripening process. In general, a gradual fall in ascorbic acid was observed during storage in both treated and untreated mango fruit. It has been observed that edible coatings had no remarkable effect on ascorbic acid of mango fruit during storage (Hoa and Ducamp, 2008). Contrary to other organic acids, ascorbic acid is quite volatile. This instability is mainly for the activity of ascorbate oxidase enzyme and the reaction with oxygen in the presence of heavy metal ions and light (Bode *et al*., 1990). Therefore, these dipping treatments might be insufficient to suppress losses of ascorbic acid in mango fruit at this storage temperature. Losses of ascorbic acid are common in various fresh fruits during storage. A sharp reduction in ascorbic acid was observed in fresh-cut and whole „Ataulfo" mango during storage for 5 days (Robles-Sánchez *et al*., 2009). Similarly, in „Brokin", „Julie" and „Peter" mango varieties, ascorbic acid continuously reduced during storage at ambient temperature stored for 12 days (Faasema *et al*., 2014). Zhu *et al*. (2008) stated that ascorbic acid was declined in control fruits and chitosan treated mango throughout the storage period. Brishti *et al*. (2013) observed that ascorbic acid content was higher in Aloe coated papaya fruits (86.55 mg) than the control fruits (61.10 mg) during the storage period at temperatures 25°C-29°C and 82-84% RH. Ahmed *et al*. (2009) found that a similar result was found in Aloe gel coated nectarines. This was because of low oxygen permeability of coating which delayed the deteriorative oxidation reaction of ascorbic acid content. Srinu *et al*. (2012) stated that coating reduces respiration of the fruits and retains the ascorbic acid in the fruits. Singh *et al*. (2000) also stated the effect of various extracts such as neem leaf extract, castor oil and neem oil on mango fruits and showed that among these extracts neem was best in retaining most of biochemical characteristics such as TSS (16.01° B), acidity (0.38%), pectin (0.98%) and ascorbic acid content (20.56 mg/100 ml juice) as compared to control fruits in which the values for these parameters were 12.03° B, 0.23%, 0.55% and 15.68 mg/100 ml juice, respectively after 12 days of storage. Bhardwaj and Sen (2003) studied the effect of various concentrations of neem leaf extracts on the storage quality of mandarin (Citrus reticulata) cv. Nagpur Santra and stated that among various treatments used neem leaf extract (20%) was significantly better in retaining higher ascorbic acid content (27.17 mg/100 ml. of juice) as compared to control fruits where it was only after storage.

Decay percentage was used to observe the effectiveness of coated substance on fruit in retarding fruit disease. Aloe vera gel was successful in decreasing microorganism proliferation in table grape, the effect being higher for yeast and molds than for mesophillic aerobics (Tripathi and Dubey, 2004). Interestingly, the Aloe vera gel coating was effective in controlling microbial growth of „Starking" cherry and „Crimson" table grape without incorporating other antimicrobial compounds such as garlic oil, potassium sorbate and nisin to enhance the activity (Pranoto *et al*., 2005). Brishti *et al*. (2013) found that in case of Aloe vera coated papaya fruits, no disease signs were observed until 1 week after the beginning of the storage period. At the end of the storage period, 100% disease incidence was found in uncoated fruits, whereas for Aloe gel coated fruits disease incidence was only 27%. This was due to the anti-microbial potentiality of coated substances which has been discussed earlier. Regarding microbial growth, chitosan coatings displayed a poor inhibitor effect on the growth of psychrophilic bacteria. Even with the higher chitosan concentrations, the final counts in pear slices were raised. However, coatings with 1.5 and 2 g L-1 chitosan showed a remarkable logarithmic reduction when compared with the control slices. These results suggest that chitosan concentrations higher than 2 g L-1 could be more efficient in decreasing bacterial growth. The inhibitory effect of chitosan coatings on the growth of mesophilic bacteria had been elaborately described (Chien *et al*., 2007; Gonzalez-Aguilar *et al*., 2008; Simões *et al*., 2009; Xu *et al*., 2013), however, only few information is available for psychrophilic bacteria. Campaniello *et al*. (2008) stated an important inhibitor effect on psychrotrophic microflora of minimally processed strawberries. This inhibition led to an appreciable prolongation of lag phase, a lower cell load and, consequently, an augment of the stability of the product. Ketsa *et al*. (2000) stated a rapid increase of disease in Thai mangoes upon removal from cold storage at 4 C for 3 weeks. Signs of decay in "Tommy Atkins" and "Palmer" mangoes became clear after 2 days at 2 C, after 4-5 days at 5 C and after 3 to 4 days at 20 C. Improvement of disease reached the maximum acceptable rate after approximately 14–18 days at 2 C, after 9 days at 5 C and after 7–8 days at 20 C in “Tommy Atkins” and “Palmer,” respectively. Ketsa *et al*. (2000) also stated that decay in the Thai mangoes stored at 25 C began to become visible after 4 days, and after 6 days, the fruits showed almost 25% of the surface area affected by decay. Hasabris and D‟souza (987) stated beneficial effects of natural plant products for the control of storage rots in Alphonso mango. They further stated that these treatments also checked the disease incidence in banana. Sarvamangla (1993) studied the effect of plant extracts on biochemical characteristics and control of fungal rot on mulberry fruit and stated that these extracts retained higher sugar content and nullify fungal attack and distribution of fruits.

Wongmetha and Ke (2013) stated that chitosan combined with 1-methylcyclopropene treatment extended the storage life of „Irwin‟ mango at 0°C up to 32 days. Gum arabic along with calcium chloride decreased decay incidence of mango fruit during storage (Khaliq *et al*., 2015). Similarly, polysaccharide based treatment and carnauba wax coating decreased decay in „Tommy Atkins" mango fruit (Baldwin *et al*., 1999). The reduction of decay incidence with GA 10% and CH 1% may be due to its film-forming characteristics, which acted as a fence and thus reduced microbial activity. A similar effect of chitosan on preserving color was found in other fresh-cut fruits such as mango (Chien *et al*., 2007), strawberries (Campaniello *et al*., 2008), papaya (Gonzalez-Aguilar *et al*., 2008), peach, pear and kiwifruit (Du *et al*., 1997). Feygenberg *et al*. (2005) carried out an experiment on use of organic coating for maintaining quality of mango cv. „Tommy Atkins‟. One coating is a colloidal solution based on beeswax (Bee Coat); whereas the other is based on carnauba wax. Coating mango effectively decreased the water loss, shrinkage, chlorophyll breakdown, chilling injury symptoms and decay development in the fruits, thereby extending their shelf life. In mango „Tommy Atkins‟, coating with the beewax based organic wax, „Bee coat‟ reduced the rates of weight loss, fruit softening, color development and acid breakdown, thus ensuring a longer shelf life. Moreover, after 3 weeks at 12°C following O days at 20°C, „Tommy Atkins" coated with Bee Coat displayed only a low level of the red spots which are symptomatic of chilling injury. Additionally, the coated fruits did not develop anaerobic metabolites or off-flavors, and were preferred by the taste panelists.

Marpudi *et al*. (2011) conducted an experiment to evaluate the ability of Aloe gel based antimicrobial coatings to reduce the loss of post-harvest fruit quality in mango and to compare the effects with a natural polysaccharide chitosan, an established coating material with antifungal activity. Freshly harvested mango fruits were coated with aloe gel (50%, mango leaf extract included aloe gel (1:1) and 2.5 % chitosan. The coated and uncoated (control) fruits were kept at 30 C and 42-55 % relative humidity for 15 days. The coated fruits survived the storage period of 15 days, whereas all the uncoated controls decayed within 10 days. The uncoated control fruits exhibited remarkably greater changes in all the parameters tested. The coatings controlled the physiological weight loss, ripening process (chemical changes, color development and softening of fruit tissue) and decay to a greater extent and they enhanced the shelf life quality of fruits. On the basis of the overall physiological changes aloe gel based antimicrobial coating has been identified as a suitable method to prolong the shelf life of mango fruits. Singh *et al*. (2011) studied the effect of various concentration of Aloe vera gel coating on refrigerated strawberry quality and shelf life with the aim to extend the shelf life of strawberries without hampering the sensory attributes under cold storage uncoated fruits showed increase in weight loss, colour changes, loss of firmness and quality deterioration during the storage (16 days). However, strawberries treated with Aloe vera gel (1:3 ratio) significantly decreased weight loss (9.95 ± 2.1 %) compared to 13.79 ± 0.13 % in control), maintained colour, firmness, quality characterstics (TSS of 8.4°B compared to 7.0° B in control, acidity of 1.37 % compared to 0.83% in control and ascorbic acid of 45 ± 0.4 mg/100g compared to 30± 0.5 mg/100g) in control and ultimately extend storability upto 16 days when stored at 5° C and RH 95%. Aloe vera gel probably had some effects on the decrease of cell wall degrading enzymes responsible for pineapple softening. These results show beneficial effects of the Aloe vera coating on enhancing the pineapple shelf life, since it has been postulated that fruit softening and texture changes during pineapple storage determine fruit storability and shelf life, as well as reduced incidence of decay and less susceptibility to mechanical damage (Batisse *et al*., 1996; Vidrih *et al*., 1998). Wang *et al*., (2007) carried out an experiment on quality and shelf life of Mango (*Mangifera Indica* L. cv. „Tainong‟) coated by using Chitosan and Polyphenols. They observed chitosan-based coatings were used to slow down ripening and prolong shelf life of mango fruit stored at 5- 8 C and 85–90% RH for 35 days. Mango fruits were mixed with 2% chitosan solution or with 2%

chitosan containing 1% tea polyphenols (TP–chitosan). Specimens were taken at regular intervals for analysis. Results indicated that chitosan coating alone could reduce the decay incidence and weight loss, and delay the change in color, pH and titratable acidity of mango fruit during storage. Firmness of the control fruit collapsed rapidly to 8.6N after 5 days of storage at 5 C, which was 22.8% or 71.5% lower than that of the fruit treated with chitosan or TP–chitosan, respectively. Chitosan treated fruit oppressed the growth of a wide variety of bacteria and fungi as compared to the control treatments. The fruit-spoiling fungi (*Colletotrichum gleosporioides*) were found in untreated control fruits after 2 weeks and in irradiated chitosan coated fruits after 5 weeks of storage. The control fruits were affected 13.3%, after 14 days of storage whereas irradiated chitosan coated fruits were affected only 6.9%. At the end of storage control fruits were fully destroyed. El-Ghaouth *et al*. (1991) suggested that chitosan induces chitinase, a defense enzyme (Mauch *et al*., 1984), which catalyzes the hydrolysis of chitin, a common element of fungal cell walls (Hou *et al*., 1998), thus preventing the growth of fungi on the fruit. The results suggest that irradiated chitosan coating is effective on the conservancy of fresh fruits. It can enhance the shelf life (Eissa, 2007), limit the growth of fungi, and decrease the spoilage without affecting on ripening characteristics of fruit (Lam and Diep, 2003). Aina (1990) stated that some physical and chemical measurements were applied to mature green African mango fruits (*Irvingia gabonensis Baill*) during a 7- day storage ripening period at tropical surrounding conditions (27-30°C and 68- 70% relative humidity). Changes in fruit weight, texture and color reflected the most remarkable chemical changes in the fruit such as starch degradation, formation of sugars and increase in total carotenoids. The postharvest ripening changes found are discussed and compared with similar changes in other mango varieties.

From the above reviews, it is clear that quite large volumes of works have been done in various parts of the world. Various issues related to the physiochemical changes, shelf life extension, and diseases have been cited above. Similar statements are scanty in Bangladesh. Very little information is present in Bangladesh regarding the use of biopreservativesas a postharvest treatment on physiochemical changes, shelf life and diseases during storage and ripening.

CHAPTER III

MATERIALS AND METHODS

This chapter is comprised of a brief description about experimental period, storage room, its controlled condition, planting material, treatments used in this experiment, experimental design and layout, data collection and statistical analysis.

Experimental location:

This experiment was conducted from June to August 2019 in the postharvest Laboratory of Horticulture Department at Sher-e-Bangla Agricultural University, Dhaka-1207, Bangladesh.

Experimental materials:

Mature, green mangoes (cv. Amrapali) were obtained from Rajshahi district, Bangladesh. Uniform sized, undamaged, healthy fruits were selected and transferred to the central Laboratory, Sher-e-Bangla Agricultural University as early as possible with careful handling to avoid injury.

Treatments of the experiment:

The experiment consisted of two factors:

Factor A: Postharvest Biopreservatives

i. Control (P_0)
ii. Aloe vera (P_1)
iii. Beewax (P_2)
iv. Neem (P_3)
v. Chitosan (P_4)

Factor B: Postharvest Temperatures

i. 10 (± 1) C (T_1)
ii. 20 (± 2) C (T_2)
iii. 30 (± 2) C (T_3)

Experimental design and treatment application:

The two factor experiment was laid out in a completely randomized design (CRD) with three replications. The postharvest biopreservatives and temperatures were assigned randomly in each replication. Under each replication, five fruits were collected for physical and destructive analysis. A total number of 15×3×5= 225 matured, uniform sized, undamaged healthy fruits were selected. Then the fruits were washed, surface sanitized with ozonized water for 20 minutes and subjected to various treatments. For coating purposes, the fruit was dipped once in the coating material and retained 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 temperatures at 10 (± 1) C, 20 (± 2) C and 30 (± 2) C.

Preparation of Biopreservatives:

Aloe vera extract preparation (P_1)

Extraction of aloe vera gel was done according to the traditional hand filleted method narrated by Ramachandra and Rao, (2008). Twenty aloe vera leaves were obtained from local town hall market, Dhaka. All were fully extended and mature enough. They were completely free from any defects. The fresh gel was made from collected aloe vera leaves. 100% aloe vera gel was prepared and for this at first they were cleaned with tap water and then with distilled water to free from dust. Then each of one side of skin was peeled off, scoop out the gel of the leave, this colorless hydro parenchyma was homogenized in a blender machine. No water was added here. The gel was then filtered by sieve to remove all unwanted lump and to get 100 percent fresh aloe gel (Plate 1a). As the gel is susceptible to enzymatic degradation so the extract was kept in a glass jar in refrigerator.

Beewax emulsion preparation (P_2)

Beewax was obtained from beehives and it was then collected from Department of Entomology, Sher-e-Bangla Agricultural University. In this experiment 6% Beewax emulsion was used. The method that is used here was described by Purwoko and Fitradesi, (2000). It was prepared by heating 60g of beewax to melt at 70 C. It was heated continuously to achieve a temperature of 80- 90 C (Plate 1b). 160 ml oleic acid was slowly mixed in this melted wax with constant stirring. In line with this, 840 ml of distill water (pre heated at same temperature of 80- 90 C) was added slowly with continued stirring for 5 minutes. The prepared emulsion was cooled and stored in a container for future use. Before use the emulsion was heated.

Neem extract preparation (P_3)

Neem solution was prepared by neem leaves. Fresh neem leaves were collected from Horticulture Farm of Sher-e-Bangla Agricultural University. 250 g neem leaves were removed from the twig and cleaned with distilled water (Plate 1c). Then they were blended by adding 500 ml of distilled water. After that the juice was collected through sieve. Only clean, pure juice of neem was collected. Then 40% neem leaf extract solution was prepared by taking 120 ml raw neem leaf extract in 500 ml beaker with the addition of 180 ml distilled water to make a final volume of 300 ml (Mia, 2003). Finally, the solutions were stored in refrigerator at 0 C.

Chitosan solution preparation (P_4)

Chitosan was collected from Hatkhola Road, Tikatoli, Dhaka-1203. It is a chemical of analytical grade. Preparation of chitosan solution was made following the methods indicated in Wongmetha and ke, (2012). Briefly, 5 gram of chitosan powder was dispersed in 850ml of distill water to which 50 ml glacial acetic acid was added to dissolve the chitosan. Chitosan solution was diluted at 2% concentration and it was done by adding 2 ml solution in 100 ml water (Plate 1d). As it is highly viscous glacial acetic acid was used to dissolve it. Then the solution was stored in air tight bottle in ambient temperature.

a b c d

Plate 1: Preparation of biopreservatives (a. Aloe vera, b. Beewax. C. Neem and d. Chitosan) in the postharvest laboratory

Observation:

During the entire postharvest storage period the experimental fruits were keenly observed every day to observe any special change. Physical observations (weight loss, shrinkage %, browning or black spot %, disease severity and shelf life) and moisture content % were recorded on 15 days of storage. For estimating chemical analysis total soluble solids (TSS), titratable acidity (TA), β-carotene, ascorbic acid and pH of each samples were drawn on 15 days of storage.

Methods of studying physico-chemical parameters:

Physical parameters

Estimation of weight loss:

Mango fruits were placed on a digital weighing balance and throughout the storage period each reading was recorded to calculate the weight loss during storage and then percentage of weight loss was calculated as:

Weight loss (%) = $\frac{(\ )\ (\ )}{(\ )}$

Estimation of moisture content:

One fruit was weighed in a porcelain crucible (which was previously cleaned, dried and weighed) from each treatment and replications. The crucible was placed in electric oven at 80°C for 72 hours until the weight became constant. It was then cooled in desiccators and weighed again. Percent moisture content was calculated by using the formula

Moisture content (%) =

Visual scoring of mango skin:

Visual scoring of mango skin was done on the basis of shrinkage severity, browning or black spots severity and disease severity. These parameters were taken by eye estimation. Mango fruits skin was scored from 0-5, whereas, 0= no shrinkage, 1=1-10% shrinkage, 2=>10-20% shrinkage, 3=>20-30% shrinkage, 4= >30-40% shrinkage, 5= >40% shrinkage. In browning or black spots 0= no browning/black spots, 1=1-10% browning/black spots, 2=>10-20% browning/ black spots, 3= >20-30% browning/ black spots, 4=>30-40% browning/black spots, 5= >40% browning/black spots. In case of disease severity 0= no disease, 1=1-10% disease, 2=>10-20% disease, 3=>20-30% disease, 4=>30-40% disease, 5=>40% disease.

Assessment of percentage of shrinkage, browning or black spots and disease severity:

The percentage of fruit skin shrinkage, browning or black spots and disease severity was recorded from 6th day of storage as visual symptom was visible. Fruits were stored till >30% fruit skin considered commercially unacceptable. All the infected fruits were selected to determine percent of fruit area infected.

Chemical parameters:

pH:

pH was measured using a phs-25 pH meter. An electrolytic cell comprise of two electrodes (calomel electrode and glass electrode) was standardized with buffer solution of pH 4. Buffer solution of any known pH value may be used here. Then the electrodes were dipped into the test sample. A voltage corresponding to the pH of the solution was identified by the instrument. For preparing sample solution of fruits, mangoes were chopped into small pieces and ground into a fine paste by mortar and pestle. The mango juice was transferred into a test tube and the pH of the paste was determined by inserting the electrodes into the paste and stabilized readings were recorded.

Total soluble solid (TSS):

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

Titratable acidity (TA):

Titratable acidity was estimated by chemical analysis process using mango pulp. Titratable acidity was declined slowly when stored in low temperature. The titratable acidity of mango pulp was determined by method of Ranganna, (1979). From mango 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 obtained. It was repeated 3 times. The acid content of the mango sample was calculated using the formula below:

$$TA\% = \frac{(}{)\ (\ )}$$

0.1N solution preparation:

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

Phenolphthalein indicator preparation:

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

Ascorbic acid

Ascorbic acid content (ascorbic acid) was estimated by using 2,6-Dichlorophenol indophenol (DCPIP) visual titration method (Rangana, 2004). 5gm mango 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 persist for at least 15 seconds. Ascorbic acid content in terms of mg/100 g pulp weight was calculated using the following formula:

Ascorbic acid (mg/100g):=

5% oxalic acid solution preparation:

It was prepared by dissolving 50g oxalic acid powder in 1000 ml distilled water.

Dye solution preparation:

It was prepared by dissolving 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 filed with the dye solution. The content of the conical flask was titrated with dye solution. The content of conical flask was titrated with dye till the pink colored end point appeared. The milliliters of dye solution required to complete the titration was recorded. Dye factor was calculated using the following formula:

Dye factor = 0.5/ titrate value

β-carotene content:

β-carotene in mango pulp was determined according to the method of (Nagata and Yamashita, 1992). One gram of pulp was mixed with 10 ml 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 calculation was done by following method:

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

Shelf life

Shelf life of mango fruits were influenced by different storage temperatures and biopreservatives. When 30% shrinkage severity, browning or black spots and disease severity occur it considered to be the end of shelf life.

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

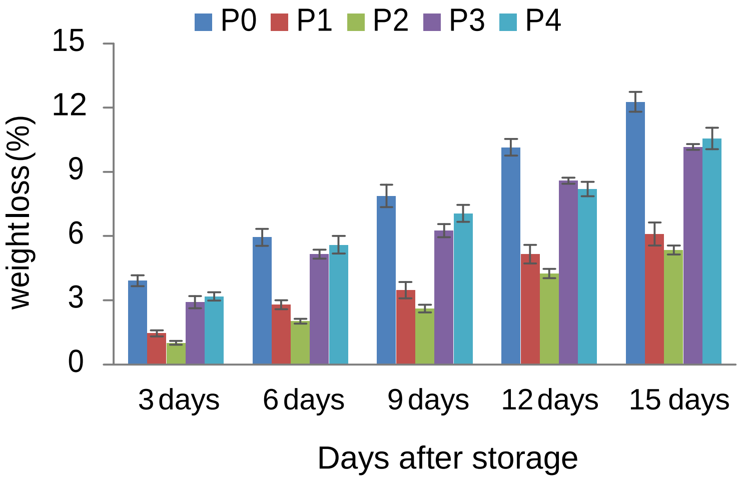
This chapter accounts for the presentation of the results acquired from the present study. The results of the study on physico-chemical changes during postharvest losses of “Amrapali” mango variety are represented and discussed from Table 1 to Table 11 and Figure 1 to Figure 22 in this chapter. These results are explained under the following headings:

Weight loss

In the postharvest life of fruits, weight loss is used as one of the main quality parameters during storage. The mango variety, different biopreservatives, their concentration, environmental condition like temperature, relative humidity exhibited more pronounced effect on total weight loss of mango during storage. The weight loss percent calculating for each biopreservative and temperatures showed significant variation (Table 1, Appendix I).

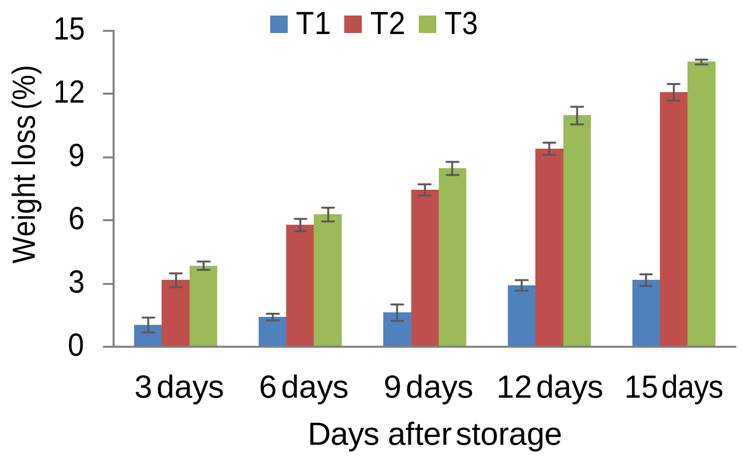
It was seen that the maximum (3.94%, 5.94%, 7.88%, 10.15% and 12.28% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS) percentage of weight loss of mango under postharvest biopreservative was found in P_0 (Controlled fruit) followed by P_4 (Chitosan treated fruit), P_3 (Neem treated fruit) and minimum (1.01%, 2.02%, 2.62%, 4.25% and 5.35% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS) was in P_2 (Beewax treated fruit). Aloe vera treated fruits showed statistically the second (1.46%, 2.79%, 3.47%, 5.15% and 6.09% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS) lowest weight loss of storage (Figure 1). The percentage of weight loss, regardless of all biopreservatives was increased with the advancement of storage time and it was highest at the end of the storage day. This result was compatible with Krishnamurthy and Babu (1993). They reported that the weight loss of Alphonso Mango after 19 days of storage could be as high as 18.13% depending on the storage condition.

It was revealed that highest (3.86%, 6.29%, 8.47%, 10.98%, and 13.51% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS) weight loss was occurred in 30 (2) C and lowest (1.05%, 1.43%, 1.64%, 2.93% and 3.17% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS) weight loss was occurred in 0 ( ) C (Figure 2). The result was very much similar to Gill *et al.* (2017). He did an experiment on mango cv. Dashehari. He kept the fruits on 20 C, 25 and room temperature. He observed that progression of ripening changes in fruit was found to be less in 20 C and 25 C than at room temperature. It occurs as high temperature promotes higher respiration rate so weight loss is high and opposite occurs in low temperature.



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 1: Effect of postharvest biopreservatives on weight loss (%) of mango at different days after storage (DAS)



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 2: Effect of postharvest temperatures on weight loss (%) of mango at different days after storage (DAS)

The data showed that the combined effect between the postharvest biopreservatives and temperatures were found statistically significant at 3rd, 6Th, 9th, 12th and 15th Days after storage. The maximum (5.69%, 8.2%, 10.83%, 13.95% and 17.07% at 3rd, 6Th, 9th, 12th and 15th DAS) rate of weight loss was observed in P_0T_3 [Controlled fruits in 30 (2) C] combination and minimum (0.22%, 0.54%, 0.54%,1.06% and 1.06% at 3rd, 6Th, 9th, 12th and 15th DAS) rate was recorded in P_2T_1 [Beewax treated fruits in 0 ( ) C] combination (Table 1).

Table 1. Combined effect of postharvest biopreservatives and temperatures on weight loss (%) of mango at different days after storage (DAS)

Treatments	**Weight loss (%)**				
	3 DAS	**6 DAS**	**9 DAS**	**12 DAS**	**15 DAS**
P_0T_1	1.54 f^z	1.87 fg	2.35 hi	3.75 h	4.29 h
P_0T_2	4.51 bc	7.75 ab	10.46 ab	12.75 b	15.48 b
P_0T_3	**5.69 a**	**8.2 a**	**10.83 a**	**13.95 a**	**17.07 a**
P_1T_1	1.16 f	1.14 gh	1.74 ij	2.91 h	3.58 h
P_1T_2	3.01 de	4.83 d	6.42 f	8.57 e	10.16 e
P_1T_3	3.21 de	5.41 cd	8.26 de	11.47 c	15.05 c
P_2T_1	**0.22 g**	**0.54 h**	**0.54 k**	**1.06 i**	**1.06 i**
P_2T_2	1.05 fg	2.25 f	3.48 gh	5.15 g	6.16 g
P_2T_3	1.77 f	3.29 e	3.83 g	6.54 f	8.83 f
P_3T_1	1.08 fg	1.66 fg	1.15 jk	3.32 h	2.88 h
P_3T_2	2.74 e	5.87 c	7.46 ef	9.97 d	12.45 d
P_3T_3	4.91 ab	7.93 ab	10.16 abc	12.47 bc	15.16 bc
P_4T_1	1.26 f	1.93 fg	2.43 hi	3.59 h	4.05 h
P_4T_2	3.84 cd	7.21 b	9.12 cd	9.17 de	13.49 de
P_4T_3	4.45 bc	7.64 ab	9.62 bc	11.84 bc	14.14 bc
LSD (0.01)	0.93	0.95	1.15	1.17	1.54
SE	0.34	0.35	0.42	0.43	0.56
CV (%)	15.37	9.43	8.71	6.71	7.16

P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan, T_1: 0 ( ) C, T_2: 20 (2) C,T_3: 30 (2) C; ^{z}Means with different letters significantly differ at LSD" s test at P ≤ 0.0 ; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference.

By considering all the above results, it was revealed that in controlled fruits weight loss were topmost due to higher rate of respiration, transpiration or evaporation of moisture (Sani *et al*., 1997). As there was no coating or barrier so vapor pressure difference between the fruits and the surrounding atmosphere was high. As a result weight loss was also high. Some other researcher indicated that due to rotting, dehydration high percentage of weight loss occurred in differently treated fruits other than fresh one (Tandon *et al*., 1985, Joshi and Roy, 1988). It can also be attributed to mishandling. Therefore, among the treatments beewax appeared to be the best biopreservative in all temperatures. This investigation was supported by Togrul and Arslan (2004). They described that the coating helps to reduce moisture loss and gaseous exchange, it make alteration to internal carbon di oxide, oxygen, ethylene level and delays ripening process and keep the fruits in good shape. The formation of a film on top of the skin performs as an addition barrier. The hydrophobic nature of beewax than chitosan and neem which acts as barrier for movement of water between inner and outer environment of fruits helps to show best result. Similar results were recorded by Thai *et al*. (2002) who showed that wax coating decrease the rate of respiration and transpiration by clogging up lenticel or stomata and resulted reduced weight loss.

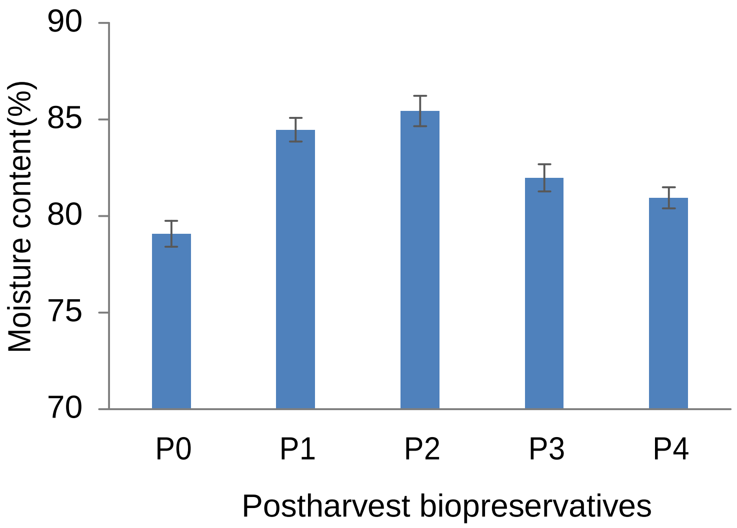
Sophia *et al*. (2015) revealed that the percentage of weight loss was high in all treatments but mango fruits that are treated with 75% aloe vera gel and stored at 13°C had the lowest weight loss followed by those treated with 75% aloe vera gel at room temperature. Aloe gel based edible coating act as barricade, therefore restricting water transfer and protecting fruit skin from mechanical injuries. This finding was supported by Tripathi and Dubey (2004). The fruits coated with different plant extracts, showed lower and slower rate in physical and chemical changes like weight loss than the uncoated fruits. Shindem *et al*. (2009) also observed more or less the similar results.

Moisture content of mango pulp

Various preservatives adopted in the study showed significant variation in relation to moisture content at 15 days after storage (Table 2, Appendix II). The maximum (85.437%) moisture content was noticed in P_2 (Beewax treated fruits) followed by P_1 (Aloe vera treated fruits) where moisture content was 84.462%. But minimum (79.076%) moisture content was found in P_0 (Controlled fruits) (Figure 3). In general the moisture content reduced with the increase in storage time under different postharvest biopreservatives and temperatures. The above outcome was in partial agreement with the findings of Joshi and Roy (1988). Srivastava (1987) described that green unripe mangoes contained higher percentage of moisture as compared to ripe mangoes. Bhatnargar and Subramanyam (1973) stated that 90% moisture content present in green ripe mango whereas pulp of ripe mango held 81% moisture. The reduction in percent moisture content was due to transpiration and starch hydrolysis. Total decrease was probably more than the increase in water due to osmotic withdrawal of water from peel to pulp and complete failure of starch to CO_2.

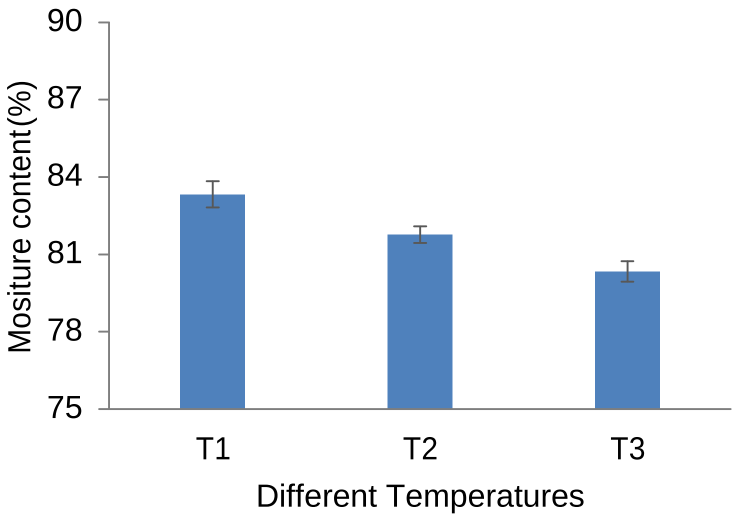
It was observed that highest (83.338%) moisture content was recorded in 0 ( ) C (T_1), followed by 20 (2) C (81.765%) and lowest (81.44%) moisture content was recorded in 30 (2) C (T_3) (Figure 4). The study indicated that the moisture content of mangoes of both temperatures 20 (2) C and 30 (2) C except those from refrigerator stored, low. This decrease in moisture content of matured mangoes may be attributed to lower loss of soluble solid due to respiration, lower rate of evaporation at prevailing weather conditions and thus moisture did not transfer from peel. The higher moisture content given by the refrigerated mangoes may be due to the fact that rate of soluble solid loss due to respiration is much lower than that at room temperature. However, the obtained moisture contents are within the range and it is reported by Jain (1961). The decrease rate was higher in control fruits and lower in treated fruits and this result was supported by Pathmanaban *et al*. (1995).

The combined effect of biopreservatives and temperatures in respect of moisture content were found to be significant. The maximum (86.413%) moisture content was observed in P_2T_1 [Beewax coated fruits in 0 ( ) C] combination followed by P_1T_1 [Aloe vera coated fruits in 0 ( ) C] combination where the value was 85.30%. On the other hand, minimum (76.14%) moisture content was observed in P_0T_3 [Controlled fruits in 30 (2) C] combination (Table 1).



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 3: Effect of postharvest biopreservatives on moisture content (%) of mango pulp at 15 days after storage



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 4: Effect of postharvest temperatures on moisture content (%) of mango pulp at 15 days after storage

Table 2. Combined effect of postharvest biopreservatives and temperatures on moisture content (%) of mango pulp at 15 days after storage

Treatments	Moisture content (%)
P_0T_1	80.643hi[z]
P_0T_2	77.443 k
P_0T_3	**76.140 l**
P_1T_1	85.300 b
P_1T_2	84.360 cd
P_1T_3	83.727 de
P_2T_1	**86.413 a**
P_2T_2	85.283 b
P_2T_3	84.527 c
P_3T_1	83.523 e
P_3T_2	82.830 f
P_3T_3	79.597 j
P_4T_1	81.920 g
P_4T_2	80.887 h
P_4T_3	80.047 ij
LSD (0.01)	0.7
SE	0.25
CV (%)	0.34

P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan, T_1: 10 (±) C, T_2: 20 (2) C,T_3: 30 (2) C; [z]Means with different letters significantly differ at LSD" s test at P ≤ 0.0 ; CV: Coefficient of Variation; SE: Standard Error, LSD: Least Significant Difference.

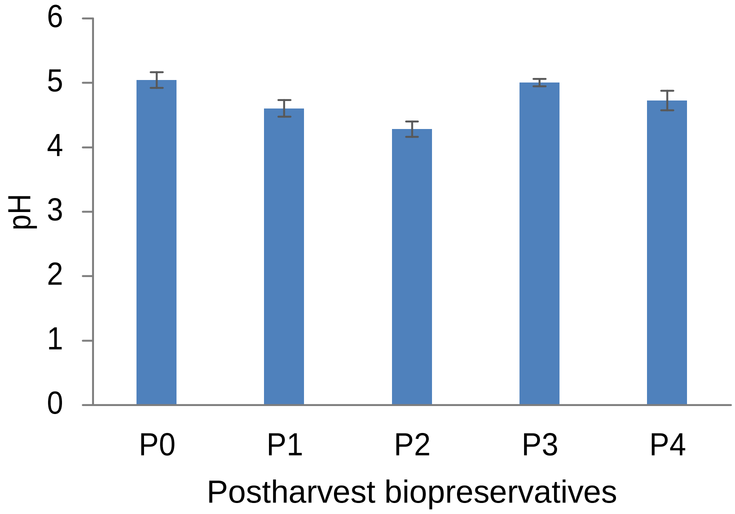
pH

Wide variations in pH of coated mangoes under different postharvest treatments were observed during successive days of storage (Table 3, Appendix II). The pH value of coated fruits showed significant differences. The highest (5.04) pH value was recorded in P_0 (controlled or untreated fruits) followed by P_3 or neem coating (5.00), P_4 or chitosan coating (4.73) and the lowest (4.28) value was observed in P_2 (Beewax coated fruits). Aloe vera treated fruits also showed lower pH value and that was 4.6 (Figure 5).

The maximum (5.41) pH value was observed in T_3 followed by T_2 (4.97) and minimum (3.82) pH value was recorded in T_1. So, from the above discussion it was concluded that untreated fruits showed highest value and beewax treated fruits showed lowest value. Moreover after beewax, aloe vera coating made its remark. By considering the temperature effect 0 ( ) C showed the lowest value compared to 20 (2) C and 30 (2) C (Figure 6).

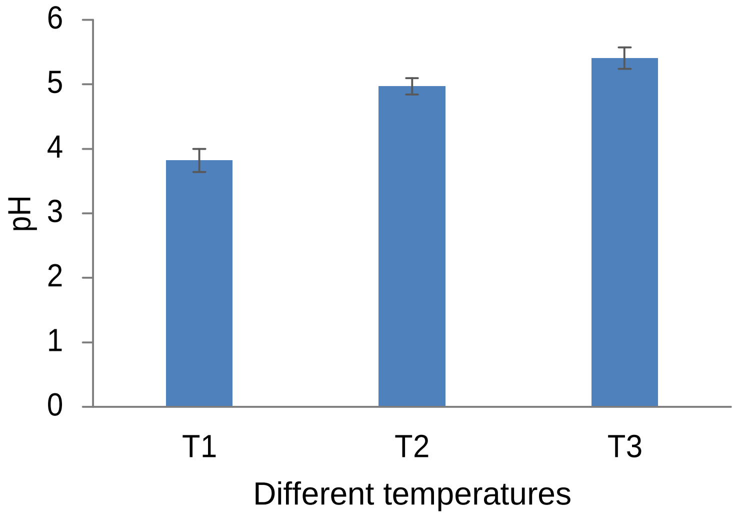
The pH value was affected by variety, waxing materials and their concentration, maturity stage of mango, their storage condition and so on. Ranges of pH value get larger continuously during the entire period of storage as acidity get lower day by day due to advancement of ripening. It was happened for the general catabolization of organic acids and their conversion into sugar. The result indicated that beewax showed the best result in all three temperatures .The coating of beewax significantly reduced the increase of fruit juice pH. The result also indicated that higher the temperature faster is the rate of ripening, so as the pH rate. On the other hand, lower the temperature, slower the rate of ripening so as the pH rate. However, untreated fruits were lack of any coating which triggered their ripening process. As a result they showed higher pH value. The results are in agreement with the findings reported by Wani *et al*. (2014). According to report as the storage period advances, total acidity could decrease and resulted in increase in fruit pH. Doreyappa and Huddar (2001) also reported similar pattern in different varieties of mangoes stored in 18-34 C.

The combined effect of biopreservatives and different temperatures also showed significant result. The maximum (5.83) pH value was noticed from P_0T_3 [controlled fruits in 30 (2) C] combination and minimum (3.64) value was recorded in P_2T_1 [Beewax coating in 0 ( ) C] combination proceeded by P_1T_1 [Aloe vera coated fruits in 0 ( ) C] combination where pH value was 3.65 very much near to P_2T_1 combination.



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 6: Effect of postharvest biopreservatives on pH of mango at 15 days after storage



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 6: Effect of postharvest temperatures on pH of mango at 15 days after storage

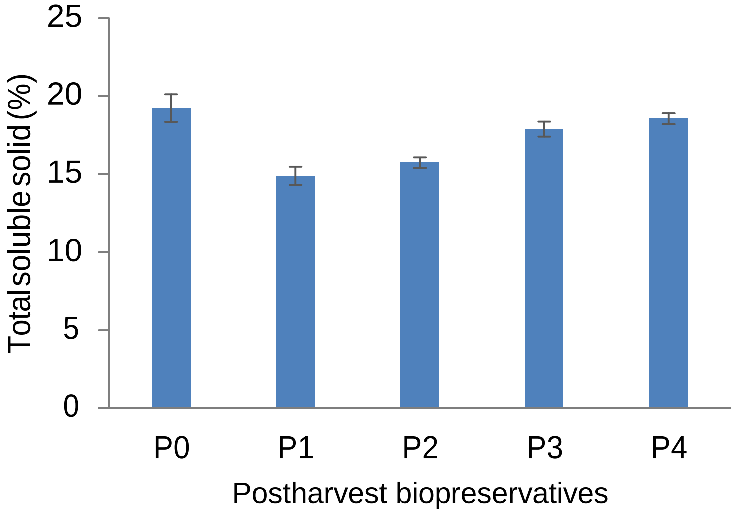
Table 3. Combined effect of postharvest biopreservatives and temperatures on the pH of mango at 15 days after storage

Treatments	pH
P_0T_1	4.02 b^z
P_0T_2	5.28 e
P_0T_3	**5.83 f**
P_1T_1	3.65 a
P_1T_2	4.86 d
P_1T_3	5.30 e
P_2T_1	**3.64 a**
P_2T_2	4.41 c
P_2T_3	4.80 d
P_3T_1	3.88 b
P_3T_2	5.43 e
P_3T_3	5.71 f
P_4T_1	3.91 b
P_4T_2	4.88 d
P_4T_3	5.39 e
LSD (0.01)	0.18
SE	0.07
CV (%)	1.73

P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan, T_1: 0 ( ) C, T_2: 20 (2) C,T_3: 30 (2) C; ^{z}Means with different letters significantly differ at LSD" s test at P ≤ 0.01; CV: Coefficient of Variation; SE: Standard Error, LSD: Least Significant Difference.

Total Soluble Solid (TSS)

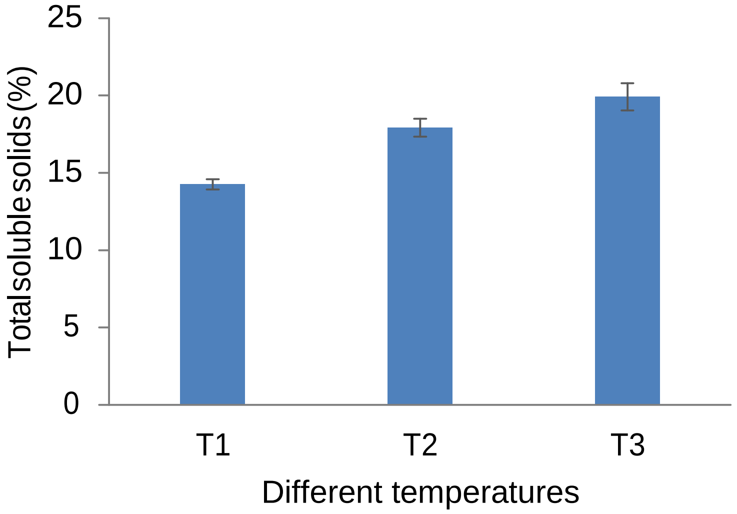
The total soluble solid content of mango was affected by the biopreservatives as the treatments showed various results on the basis of mango variety, environmental condition and waxing material. There was a significant variation in TSS during storage due to biopreservatives and temperatures. (Table 4, Appendix III). The fruits coated with aloe vera extract (P_1) maintained the lowest TSS value (14.88%) followed by beewax (15.73%), while untreated control fruits (P_0) maintained the highest TSS value (19.22%) (Figure 7).



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 7: Effect of postharvest biopreservatives on TSS (%) of mango pulp at 15 days after storage

Temperatures showed significant variation as the highest value (19.93%) was recorded in fruits kept in 30 (2) C (T_3) and lowest value was recorded in 0 ( ) C (T_1) where the TSS value was 14.27% (Figure 8).



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (± 2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 8: Effect of postharvest temperatures on TSS (%) of mango pulp at 15 days after storage

The combined effect of biopreservatives and temperatures in respect of TSS were found to be significant. The maximum (22.33%) TSS value was observed in P_0T_3 [Controlled fruits in 30 (2) C] combination and minimum (12.67%) TSS value was observed in P_1T_1 [Aloe vera treated fruits in 0 ( ) C] combination (Table 4).

Table 4. Combined effect of postharvest biopreservatives and temperatures on the TSS of mango pulp at 15 days after storage

Treatments	**TSS (%)**
P_0T_1	14.67 ef[z]
P_0T_2	20.67 b
P_0T_3	**22.33 a**
P_1T_1	**12.67 g**
P_1T_2	15.33 def
P_1T_3	16.67 d
P_2T_1	14.00 fg
P_2T_2	16.67 d
P_2T_3	18.33 c
P_3T_1	15.67 de
P_3T_2	18.67 c
P_3T_3	21.33 ab
P_4T_1	14.33 ef
P_4T_2	18.33 c
P_4T_3	21.00 ab
LSD (0.01)	1.37
SE	0.5
CV (%)	3.5

P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan, T_1: 0 ( ) C, T_2: 20 (2) C,T_3: 30 (2) C; [z]Means with different letters significantly differ at LSD" s test at P ≤ 0.0; CV: Coefficient of Variation; SE: Standard Error, LSD: Least Significant Difference.

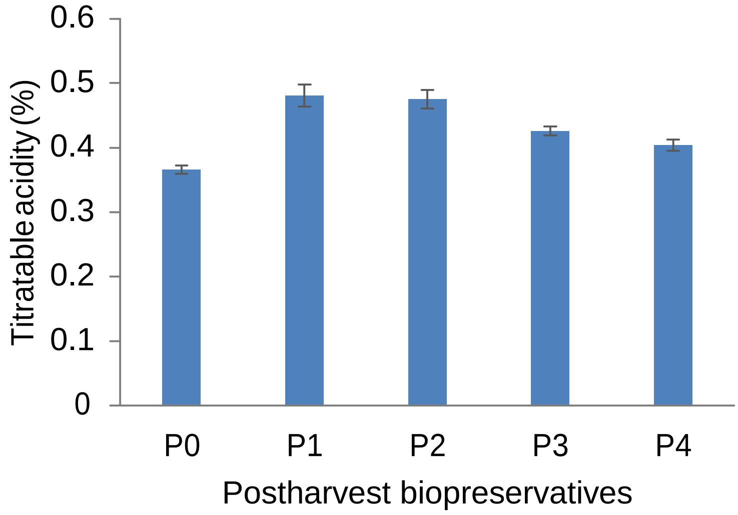
The delay in TSS content upon coating application could be related with the oxygen barrier property of edible coating and reduction of respiration. Similar observation was reported by Yonemoto *et al*. (2002) who explained that lower levels of total soluble solids in fruits coated with aloe vera may be due to protective oxygen barrier that reduces oxygen supply to the fruit surface which in turn inhibited respiration. Sharafat *et al*. (1990) also observed that as storage is prolonged, the rate of respiration, transpiration and other metabolic changes are increased in control fruits in comparison with edible coated mango fruits. Kittur *et al*. (2001) also observed similar trends with the present study and stated aloe vera-based coatings were much superior in prolonging the shelf life and quality of banana and mango than polysaccharide-based composite coating.

Titratable Acidity (TA)

There was a significant variation in TA (%) of mango fruits during storage due to effective coatings and temperatures. (Table 5, Appendix III). The maximum value (0.48%) of titratable acidity for mango fruits was recorded for aloe vera (P_1) followed by beewax (P_2), the value was 0.47% and the minimum (0.36%) value was recorded for control fruits (P_0) (Figure 9). Fruit coating at higher concentration slowed down fruit respiration and the utilization of respiratory product like organic acid was minimal. So coated fruits have higher TA value than control fruits. Tefera *et al*. (2008) found similar findings that fruit acidity is decreased because of postharvest treatments as they delay respiration and utilization rate of respiratory substrates such as organic acids.

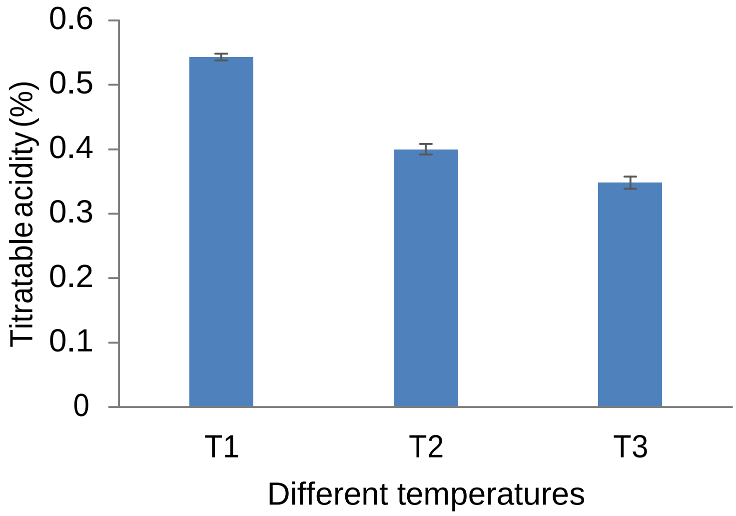
The maximum value (0.54%) was observed in 0 ( ) C (T_1) temperature followed by 0.40% in 20 (2) C (T_2) temperature whereas minimum value (0.35%) was recorded for 30 (2) C (T_3) temperature (Figure 10). Low temperature delay the ripening, so the storage period of mango fruits was longer. While the amount of TA in all mango fruits was brought down, the TSS in the fruits increased through the harvest period. This indicates that when the storage period of mango fruits was longer, ripe mangoes tasted sweeter and less sour. Joshi and Roy (1988) showed that acidity of fruits decreased continuously during storage at 00 C up to 32 days. The decrease in acidity can be attributed to metabolic reaction of acids during storage. The The pattern of changes in titratable acidity in all treated mangoes are almost similar. Doreyappa and Huddar (2001) stated that different varieties of mango fruits stored at temperature of 18-34 C showed similar pattern. General pattern is that it decreases with increase in storage period.

Combined effect of postharvest biopreservatives and different temperatures appeared significant differences. The above interaction table showed that highest (0.56%) TA value was recorded in P_1T_1 [Aloe vera treated fruits in 0 ( ) C] combination followed by P_2T_1 [Beewax coated fruits in 0 ( ) C] combination with TA value of 0.55% and the lowest (0.24%) value of TA was noticed in P_0T_3 [Controlled fruits in 30 (2) C] combination (Table 5).



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 9: Effect of postharvest biopreservatives on TA (%) of mango pulp at 15 days after storage



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 10: Effect of postharvest temperatures on TA (%) of mango pulp at 15 days after storage

Table 5. Combined effect of postharvest biopreservatives and temperatures on the TA (%) of mango pulp at 15 days after storage

Treatments	**TA (%)**
P_0T_1	0.53 c^z
P_0T_2	0.33 f
P_0T_3	**0.24 h**
P_1T_1	**0.56 a**
P_1T_2	0.45 d
P_1T_3	0.43 e
P_2T_1	0.55 ab
P_2T_2	0.45 de
P_2T_3	0.44 de
P_3T_1	0.54 bc
P_3T_2	0.44 de
P_3T_3	0.29 g
P_4T_1	0.54 bc
P_4T_2	0.33 f
P_4T_3	0.34 f
LSD (0.01)	0.02
SE	6.84
CV (%)	1.95

P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan, T_1: 0 ( ) C, T_2: 20 (2) C,T_3: 30 (2) C; ^{z}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

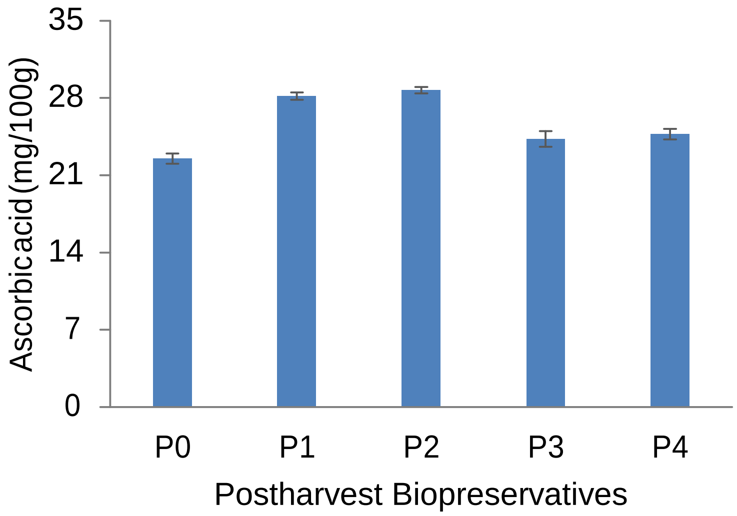
Ascorbic acid content:

Fruits are the natural source of ascorbic acid and loss of ascorbic acid is very much common in fresh fruits. It is very responsive to degradation due to its oxidation (Veltman *et. al*. 2000) compared to other nutrient during food processing, preservation and storage. As the fruits proceed towards ripening process, the level of acid gradually decreased. In general, a gradual decline was observed both treated and untreated controlled mango fruits. The significant variation was observed in biopreservatives and different temperatures (Table 6, Appendix III).

It was seen that the biopreservatives showed significant differences. The highest value (28.71 mg/ 100 g) was recorded for beewax (P_2) treated fruits followed by aloe vera

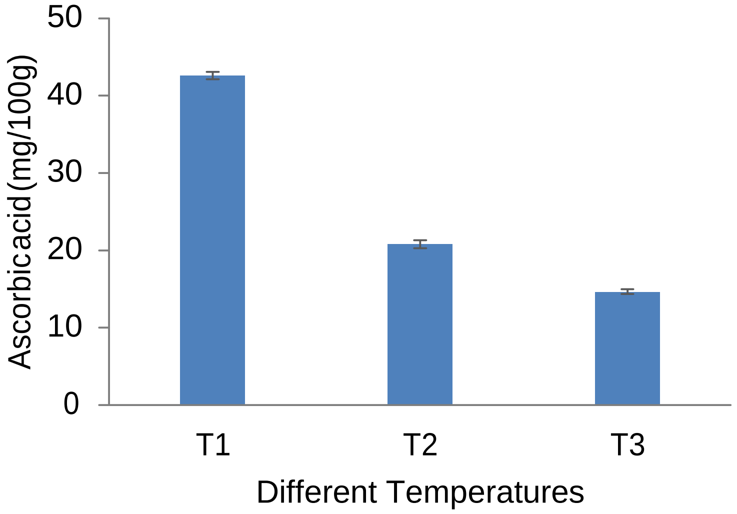
(28.17 mg/ 100 g), chitosan (24.73 mg/ 100 g), neem (24.29 mg/ 100 g) and lowest (22.51 mg/ 100 g) value was observed in controlled fruits (P_0) (Figure 11).

Significant variation was reported in case of temperatures like highest (42.59 mg/100g) value of ascorbic acid was noticed in 0 ( ) C followed by 20 (2) C (19.79 mg/100 g) and lowest (14.66 mg/100 g) value was observed in 30 (2) C (Figure 12). The results are in agreement with the research findings narrated by Bristi *et al*. (2013) that ascorbic acid content was higher in Aloe vera coated papaya fruits (86.55 mg) than the control fruits (61.10 mg) during the storage period at temperatures 25°C-29°C and 82-84% RH. This was because low oxygen permeability of coating delayed the deteriorative oxidation reaction of ascorbic acid content (Ayranci and Tunc 2003). Srinu *et al*. (2012) stated that coating reduces respiration of the fruits and retains the ascorbic acid in the fruits. Carrillo *et al*. (2000) who performed an experiment on Haden mangoes and examined a slower decreasing rate of ascorbic acid in Haden mangoes coated with different concentrations of semper fresh as compared to non- coated fruits at 3 C during 32 days of storage. These results are compatible with Dhaka *et al*. (2001) who described that retention of ascorbic acid to tapuri mango was highest (9.89%) when coated with 8.0% wax emulsion as compared to untreated fruits (8.27%) at ambient temperature.



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 11: Effect of postharvest biopreservatives on Ascorbic acid content of mango pulp at 15 days after storage



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 12: Effect of postharvest temperatures on Ascorbic acid content of mango pulp at 15 days after storage

The combined effect of biopreservatives and temperatures in respect of Ascorbic acid were found to be significant. The maximum (48.02 mg/100 g) value was observed in P_1T_1 [Aloe vera coated fruits in 0 ( ) C] combination followed by P_2T_1 [Beewax treated fruits in 0 ( ) C] combination where the value was 46.48 mg/100 g. On the other hand, minimum (12.31 mg/100 g) value was observed in P_0T_3 [controlled fruits in 30 (2) C] combination (Table 6).

Table 6. Combined effect of postharvest biopreservatives and temperatures on the Ascorbic acid content of mango pulp at 15 days after storage

Treatments	**Ascorbic acid (mg/100g)**
P_0T_1	38.05 d^z
P_0T_2	17.18 h
P_0T_3	**12.31 j**
P_1T_1	**48.02 a**
P_1T_2	21.36 e
P_1T_3	15.13 i
P_2T_1	46.48 b
P_2T_2	22.18 e

2T 3	17.48 gh
3Ti	39.19 d
3T	19.56 f
P3T 3	14.11 i
4T t	41.21 c
4T 2	18.7 fg
4T 3	14.29 i
LSD (0.01)	1.43
SE	0.51
CV (%)	2.48

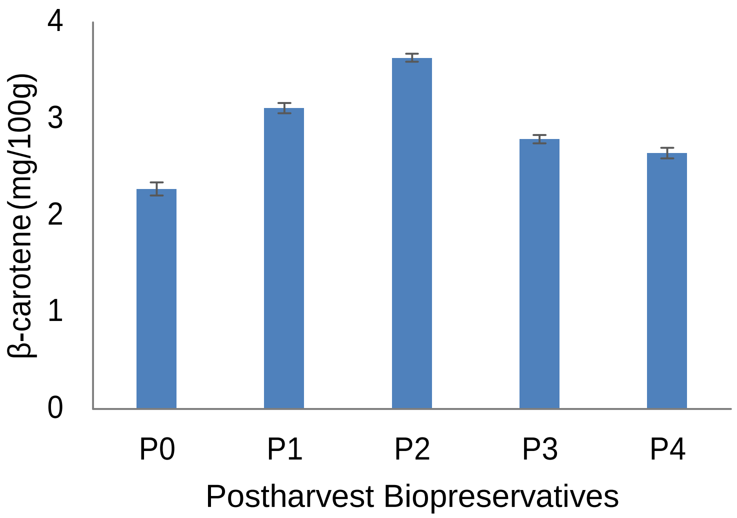
Ph: Control, P t : Aloe vera, 2: Beewax, P,: Neem, P4: Chitosan, T : 10 (A l) " C, T2• 20 (z 2) C, T : 30 (+ 2) C; ‘Means with different letters significantly differ at LSD’ s test at P < 0.01; CV: Coefficient of Variation; SE: Standard Error, LSD: Least Significant Difference.

b-carotene

§-carotene content of mango pulp showed significant variations in case of biopreservatives and temperatures and their combined effects also appeared to be significant (Table 7, Appendix III).

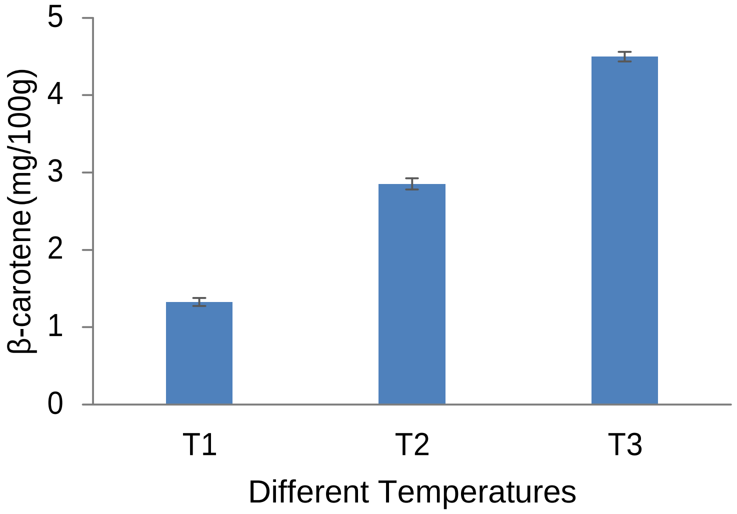
The highest (3.63 mg/100 g) b-carotene content was recorded in 6% beewax (P_2) treated fruits followed by aloe vera (3.11 mg/100 g), neem (2.79 mg/100 g), chitosan (2.65 mg/100 g) and lowest (2.28 mg/100g) §-carotene content was recorded in controlled (P_0) fruits (Figure 13). Biopreservatives formed a layer on the fruit; it kept the fruit temperature low as a result the respiration rate was slow. So, the coated fruits reached to their best edible stage in 15 days. On the contrary, as fruit temperature was high the respiration rate was high in control or untreated fruits, so they reached to ripen stage earlier than coated fruits and get spoiled before.

The highest (4.50 mg/100 g) §-carotene content was observed in 30 (+ 2) C (T_3) temperature followed by 20 (+ 2)" C (2.85 mg/100g) and lowest (1.33 mg/100 g) §-carotene content was noticed in 10 (+ 1) C (T_o) temperature (Figure 14). The §-carotene of mango significantly increased with the development of storage period. This occurs due to breakdown of chlorophyll and increase in carotenoid content by chlorophyllase enzyme during the storage period. Saltveit (1999) examined that increase in carotenoids with fruit ripening is associated with the climacteric increase in respiration and ethylene production. When temperature is low breakdown of chlorophyll is slow, so ripening process is slower, and formation of carotene is slower and lower also.



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 13: Effect of postharvest biopreservatives on β-carotene content of mango pulp at 15 days after storage



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 14: Effect of postharvest temperatures on β-carotene content of mango pulp at 15 days after storage

It was seemed that highest (5.67 mg/100 g) β-carotene content was observed in P_0T_3 [Controlled fruits in 30 (2) C] combination and lowest (0.90 mg/100 g) value was recorded in P_1T_1 [Aloe vera treated fruits in 0 ( ) C] combination. Moreover, P_2T_1 [Beewax treated fruits in 0 ( ) C] where the value was 0.97 mg/100 g also showed significantly lower β-carotene content (Table 7).

Table 7. Combined effect of postharvest biopreservatives and temperatures on the β-carotene content of mango pulp at 15 days after storage

Treatments	**β-carotene content (mg/100 g)**
P_0T_1	1.6 i[z]
P_0T_2	3.67 e
P_0T_3	**5.67 a**
P_1T_1	**0.90 k**
P_1T_2	2.53 h
P_1T_3	3.98 d
P_2T_1	0.97 k
P_2T_2	2.35 h
P_2T_3	3.42 f
P_3T_1	1.27 j
P_3T_2	2.94 g
P_3T_3	4.58 c
P_4T_1	1.56 i
P_4T_2	2.97 g
P_4T_3	4.99 b
LSD (0.01)	0.19
SE	0.07
CV (%)	2.84

P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan, T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; [z]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

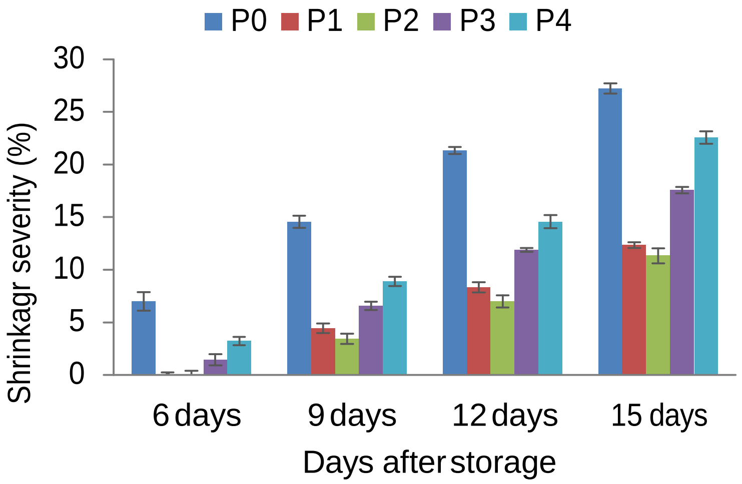
Visual scoring of mango skin

A significant change in the skin of mango was observed. Mango skin was scored by eye estimation (Plate 2, 3, 4).

Severity on the basis of shrinkage

Shrinkage of fruits is sometimes referred as deformation of material, and it is an obvious physical phenomenon commonly observed during storage. Postharvest biopreservatives and temperatures had significant effect on shrinkage severity in mango skin (Table 8, Appendix IV). The maximum (7%, 14.56%, 21.33% and 27.22% at 6th, 9th, 12th and 15th DAS) shrinkage severity was recorded in P_0 (Controlled fruits) and minimum (0%, 3.44%, 7.00% and 11.33% at 6th, 9th, 12th and 15th DAS) value was noticed in P_2 (Beewax treated fruits). Aloe vera (P_1) treated fruits also showed the second lowest (0%, 4.44%, 8.33% and 12.33% at 6th, 9th, 12^{h} and 15th DAS) shrinkage value (Figure 15).

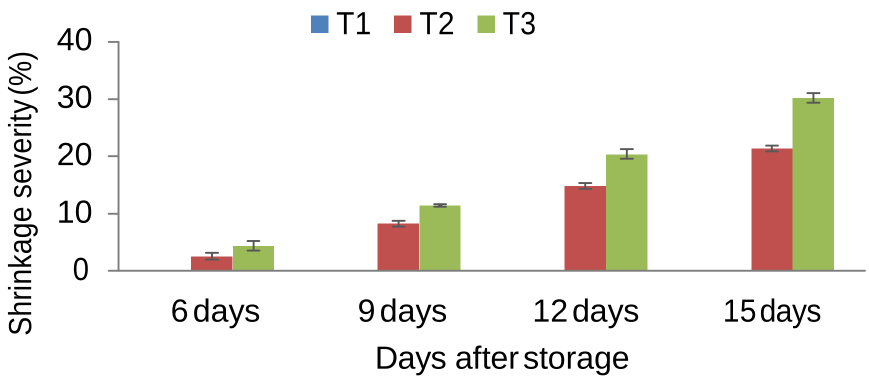
Highest (4.40%, 11.47%, 20.40% and 30.20% at 6th, 9th, 12th and 15th DAS) severity occurred in T_3 temperature and lowest (0% up to 15th day) value was recorded in T_1 temperature (Figure 16). The above finding is supported by Touil *et al*. (2014). She stated that Shrinkage occurs as a result of volume reduction due to evaporation of the moisture contained in the solid. She included that coating improves physical condition of fruits against shrinkage. As controlled fruits lost high amount of water they got huge shrinkage. But beewax and aloe vera provided with barrier reduce water loss through lenticel. Thus shrinkage was lowest. Lowering temperature by keeping fruits in the refrigerator reduces water loss which ultimately promotes good shape of mangoes. Like as, high temperature activates water loss and promote shrinkage.



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 15: Effect of postharvest biopreservatives on shrinkage severity (%) of mango at different days after Storage (DAS)

Plate 2: Visual scoring of mango on the basis of shrinkage at 15 days after storage whereas, 0= no shrinkage, 1= 1-10% shrinkage, 2= >10-20% shrinkage, 3= >20-30% shrinkage, 4= 30-40% shrinkage, 5= >40% shrinkage of mango skin



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 16: Effect of postharvest temperatures on Shrinkage severity (%) of mango skin at different days after storage (DAS)

The significant variation observed in combined effect of postharvest biopreservatives and temperatures on shrinkage severity of mango. The highest (9.33%, 15.00%, 24.00% and 33.33% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) value was observed in P_0T_3 combination and lowest value (0 up to 15^{th} day) was observed in P_0T_1 [Controlled fruits in 0 ( ) C], P_1T_1 [Aloe vera treated fruits in 0 ( ) C], P_3T_1 [Neem treated fruits in 0 ( ) C] and P_4T_1 [Chitosan treated fruits in 0 ( ) C] combinations (Table 8).

Table 8. Combined effect of postharvest bio-preservatives and temperatures on Shrinkage severity (%) of mango at different days after storage (DAS)

Treatments	**Shrinkage severity (%)**			
	6 DAS	9 DAS	12 DAS	15 DAS
P_0T_1	**0 e**[z]	**0 g**	**0 e**	**0 d**
P_0T_2	7.67 b	13.67 ab	20.00 b	27.33 ab
P_0T_3	**9.33 a**	**15.00 a**	**24.00 a**	**33.33 a**
P_1T_1	**0 e**	**0 g**	**0 e**	**0 d**
P_1T_2	0 e	4.67 efg	9.33 de	14.33 c
P_1T_3	0 e	5.67 def	11.67 cd	19.67 c
P_2T_1	**0 e**	**0 g**	**0 e**	**0 d**
P_2T_2	0 e	2.67 fg	7.00 de	12.33 cd
P_2T_3	0 e	7.67 def	14.00 d	21.67 c
P_3T_1	**0 e**	**0 g**	**0 e**	**0 d**
P_3T_2	1.00 de	5.33 efg	10.67 d	16.00 c
P_3T_3	3.33 cde	9.33 bcd	18.00 bc	26.67 b
P_4T_1	**0 e**	**0 g**	**0 e**	**0 d**
P_4T_2	4.33 bcd	10.00 cde	15.33 d	21.00 c
P_4T_3	5.33 bc	12.67 bc	20.33 b	30.67 ab
LSD (0.01)	3.56	6.83	9.44	13.90
SE	1.28	2.47	3.41	5.03
CV (%)	17.57	19.95	21.20	22.09

P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan, T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30(2) C;
[z]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

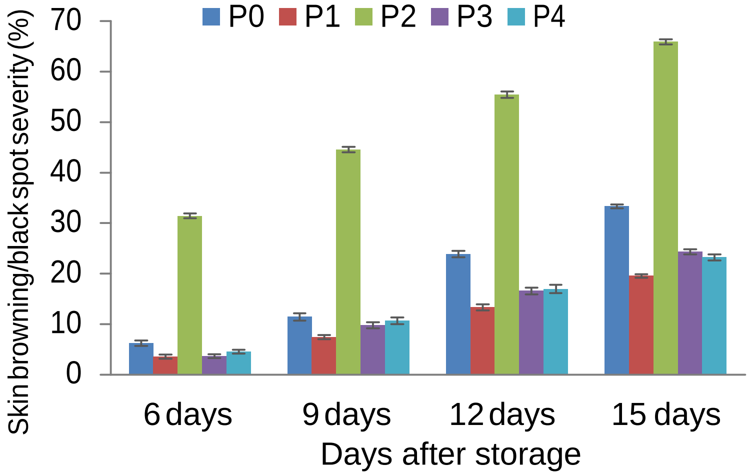
Severity on the basis of browning or black spots

Browning or black spots decreased quality of mangoes. Various biopreservatives adopted in the study exhibited significant variation in relation to browning or black spots on skin (Table 9, Appendix V). The maximum (31.44%, 44.56%, 55.44 % and 65.89% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) value was noticed from P_2 (Beewax treated fruits) followed by P_0 (Controlled fruits) which showed a value of 6.22%, 11.44%, 23.89% and 33.33% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS. Controlled fruits mainly showed black spots. But in beewax browning of external skin was highly visible. On the contrary, lowest (3.56%, 7.44%, 13.33 % and 19.56% at 6th, 9th, 12th and 15th DAS) value was obtained from P_1 (Aloe vera treated fruits) (Figure 17).

The highest (14.73%, 23.80%, 32.07% and 40.8% at 6th, 9th, 12th and 15th DAS) value of browning was found in T_3 temperature and lowest (5.46%, 12.73%, 18.80% and at 6th, 9th, 12th and 15th DAS) value was recorded in T_1 temperature (Figure 18). Beewax coated fruits had no change except the skin color. Fruits which were treated with beewax formed a brown color on their skin. The appearance got changed due to this off color. But it did not affect internal color or composition of fruit. Beewax coated fruits showed higher level of skin browning which might be occur due to-

- Beewax coating should be thin. It needs only one or two drops of wax for making thin layer around the fruit. High concentration of coating material which might be cause off-color by heavy blocking of gas exchange. CO_2 deposited on the skin, promoting off- color.
- .It causes oxidative browning of skin (Shellhammer and Krochta, 1997). However, the other preservatives showed no such type of change. Beewax can create wax whiting also called chalking on the surface of fruit due to high temperature or moisture.

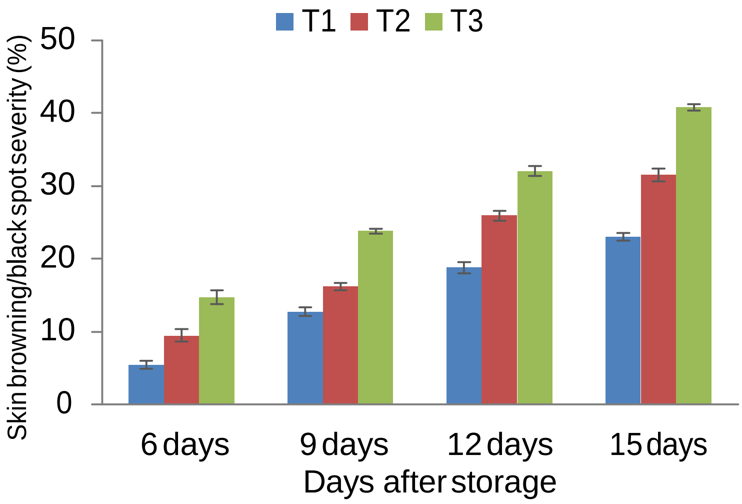
The present investigation showed that temperature highly influenced mango skin appearance. It was observed that lower was the temperature slower the rate of change in skin appearance. Higher was the temperature, faster was the rate of change occurred on the skin. The color of fruit skin is altering due to unmasking of preformed pigments by degradation of chlorophyll and biosynthesis of carotenoids and anthocyanins and their accumulation in vacuoles (Tucker and Grierson, 1987; Lizada, 1993).



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 17: Effect of postharvest bio-preservatives on Browning or black spots severity (%) of mango at different days after storage (DAS)

Plate 3: Visual scoring of mango on the basis of browning or black spots at 15 days after storage, whereas, 0= no browning/black spots, 1= 1-10% browning/black spots, 2= >10-20% browning/black spots, 3= >20-30% browning/black spots, 4= >30-40% browning/black spots, 5= >40% browning/ black spots



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 18: Effect of postharvest temperatures on Browning or black spots severity (%) of mango at different days after storage (DAS)

The combined effects of postharvest biopreservatives and temperatures were statistically significant (Table 9). The highest (28.33%, 40.00% 58.33%, 75.00% at 6th, 9th, 12th and 15th DAS) browning or black spot severity was recorded in P_2T_3 combination and lowest (0, 2.67%, 5.33% and 10.00% at 6th, 9th, 12th and 15th DAS) value was recorded in P_1T_1 combination.

Table 9. Combined effect of postharvest biopreservatives and temperatures on Browning or black spot severity (%) of mango at different days after storage (DAS)

Treatments	**Browning or black spot severity (%)**			
	6 DAS	9 DAS	12 DAS	15 DAS
P_0T_1	2.67 def[z]	6.00 de	10.33 efg	15.67 f
P_0T_2	6.00 cdef	12.33 cd	18.67 cd	24.00 cd
P_0T_3	9.00 cd	17.00 c	26.67 bc	33.33 b
P_1T_1	**0 f**	**2.67 e**	**5.33 g**	**10.00 g**
P_1T_2	0 f	1.00 e	6.67 g	10.33 g
P_1T_3	3.67 c	8.67 cd	13.00 cde	19.33 de
P_2T_1	11.33 b	19.67 b	33.33 b	41.33 c
P_2T_2	26.67 a	38.00 ab	50.67 a	63.33 a

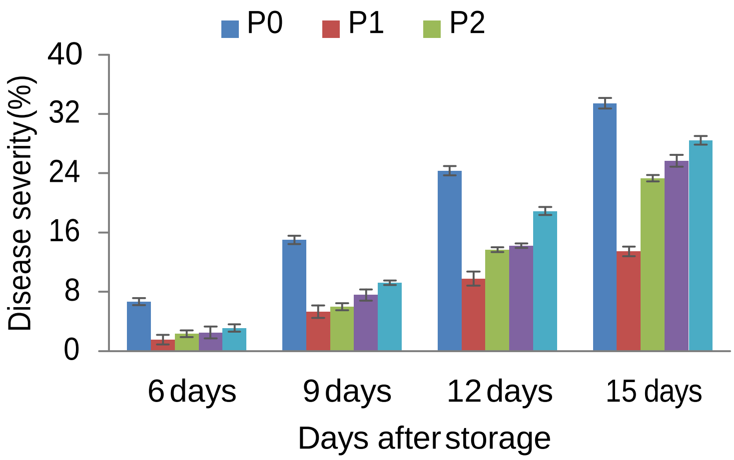
P_2T_3	**28.33 a**	**40.00 a**	**58.33 a**	**75.00 a**
P_3T_1	1.67 ef	3.33 de	7.33 fg	13.33 fg
P_3T_2	1.67 ef	6.67 de	13.67 fg	19.33 fg
P_3T_3	7.67 cde	13.33 cd	19.67 def	27.33 de
P_4T_1	1.67 cdef	3.00 cde	8.33 defg	13.00 fg
P_4T_2	3.00 def	8.00 de	14.33 fg	19.33 f
P_4T_3	7.00 cdef	13.00 cde	20.33 def	28.33 ef
LSD (0.01)	4.36	9.39	10.53	9.59
SE	2.67	2.49	2.89	2.19
CV (%)	23.02	25.27	26.34	27.57

P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3:Neem, P_4: Chitosan, T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30(2) C; zMeans with different letters significantly differ at LSD" s test at $P \leq 0.0$; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference

Severity on the basis of disease

Bio-preservatives have significant variation to disease severity. (Table 10, Appendix VI). It was observed that bio-preservatives had significant effect on disease severity of mango skin. The highest (6.67%, 15.00%, 24.33% and 33.44% at 6th, 9th, 12th and 15th DAS) value was recorded in P_0 (Controlled fruits) and lowest (1.56%, 5.33%, 9.78% and 13.44% at 6th, 9th, 12th and 15th DAS) value was obtained from P_1 (aloe vera treated fruits) (Figure 19).

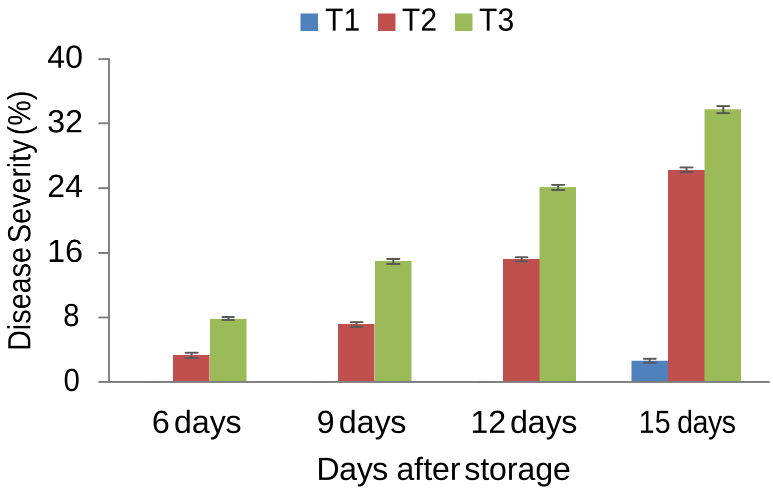
It was observed that temperatures had significant effect on disease severity of mango skin. The highest (7.87%, 14.93%, 24.13% and 33.73% at 6th, 9th, 12th and 15th DAS) value was recorded in T_3 and lowest (0, 0, 0 and 2.67% at 6th, 9th, 12th and 15th DAS) value was obtained from T_1 temperature (Figure 20). From the above discussion it is clear that severity of disease on mango fruits increased with the advancement of time. When the temperature was low, infection was low. Higher was the temperature, faster was the rate of infestation. This finding is very much similar with Alkan *et al*. (2015). He stated that among postharvest disease in mango, anthracnose caused by *Colletotrichum gloeosporioides* is predominant under humid growth conditions. From the analysis it is seemed coated fruits are tend to infect less than controlled fruits. This finding is similar with Molla *et al*. (2011). Fungal diseases account for one of the main causes of loss during commercialization of tropical fruits. The extracts collected from different medicinal plants like neem, garlic and aloe vera were found most effective to check the mycelial growth of *C. gloeosporioides* and these findings were strongly supported by Raheja and Thakore (2002). Scientist EI-G haouth *et al*. (1992) who reported that aloe vera coating prevent attack of tomatoes by *Penicillium spp*., *Aspergillus spp*., *Rhizopus stolonifer* and *Botrytis cinerea*. The investigation by Eissa (2007), also stated that wax emulsion helps extend the shelf life by limiting the growth of fungi and decrease the spoilage without affecting ripening process of fruits.



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 19: Effect of postharvest bio-preservatives on disease severity (%) of mango at different days after storage (DAS)

Plate 4 Visual scoring of mango on the basis of disease severity at 15 days after storage whereas, 0= no infection, 1= 1-10% infected, 2= >10-20% infected, 3= >20-30% infected, 4= >30-40% infected, 5= >40% infected by disease.



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 20: Effect of postharvest temperatures on disease severity (%) of mango at different days after storage (DAS)

The combined effects of postharvest biopreservatives and temperatures were statistically significant (Table10). Highest (20.33%, 30.67% 45.33% and 65.00% at 6_{th}, 9_{th}, 12_{th} and 15_{th} DAS) disease severity was recorded in P_0T_3 combination. On the contrary, no infection was found in P_1T_1 [Aloevera treated fruits in 0 ( ) C], P_2T_1 [Beewax treated fruits in 0 ( ) C], P_3T_1 [Neem treated fruits in 0 ( ) C] and P_4T_1 [Chitosan treated fruits in 0 ( ) C] combination.

Table 10. Combined effect of postharvest biopreservatives and temperatures on disease severity (%) of mango at different days after storage (DAS)

Treatments	**Disease severity (%) (DAS)**			
	6 DAS	9 DAS	12 DAS	15 DAS
P_0T_1	0 f[z]	0 i	0 h	7.33 h
P_0T_2	8.67 b	16.33 b	25.67 b	35.00 b
P_0T_3	**20.33 a**	**30.67 a**	**45.33 a**	**65.00 a**
P_1T_1	**0 f**	**0 i**	**0 h**	**0 i**
P_1T_2	0 f	5.33 h	13.67 g	19.67 gh
P_1T_3	4.67 d	10.67 fg	15.67 f	20.67 g

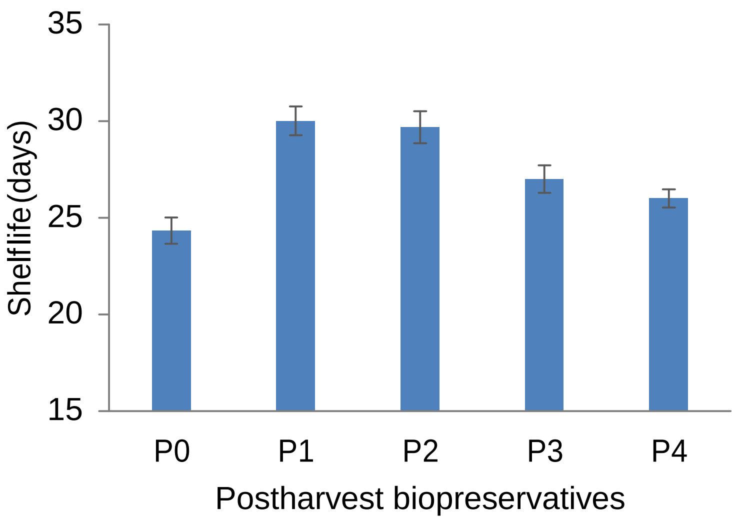
P_2T_1	**0 f**	**0 i**	**0 h**	**0 i**
P_2T_2	2.00 e	9.67 g	21.33 de	30.00 f
P_2T_3	5.00 d	11.33 f	21.67 d	30.00 de
P_3T_1	**0 f**	**0 i**	**0 h**	**0 i**
P_3T_2	0 f	10.00 g	20.00 e	35.00 ef
P_3T_3	0 f	12.67 e	22.67 d	42.00 d
P_4T_1	**0 f**	**0 i**	**0 h**	**0 i**
P_4T_2	0 f	12.33 d	24.33 c	36.33 cd
P_4T_3	9.33 c	19.33 c	39.67 b	51.00 bc
LSD (0.01)	1.31	1.32	1.87	6.84
SE	0.47	0.48	0.68	2.47
CV (%)	15.57	5.87	4.68	10.88

P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan, T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; [z]Means with different letters significantly differ at LSD" s test at P ≤ 0.0 ;CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference

Shelf life:

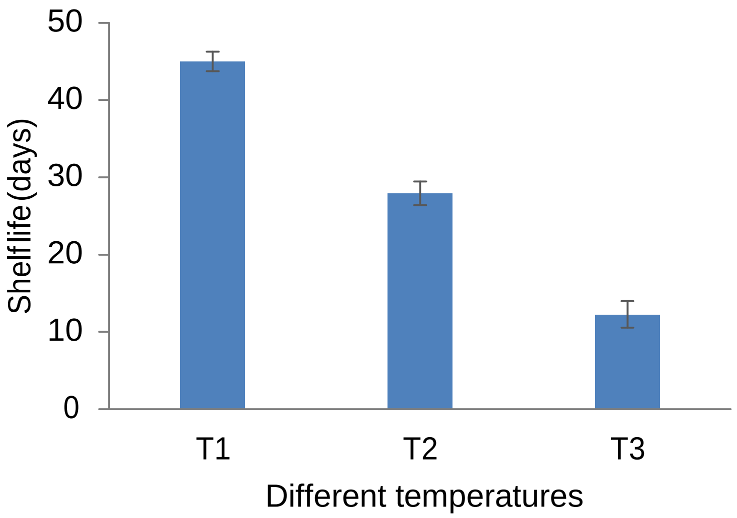
The basic quality index of fruit is shelf life and it is the most important parameter in loss of biochemical reaction of fruit. This shelf life period begins from the time of harvesting and extends up to the start of rotting of fruit (Mondal, 2000). In this present study shelf life was determined by eye estimation. Highly significant variation was observed in respect of shelf life of mango due to the effect of different postharvest biopreservatives and temperatures (Table 11, Appendix VII). It could be mentioned that highest (30 Days) shelf life of mango fruits was belong to aloe vera extract (P_1) followed by beewax emulsion (29.67 Days) treated fruits and lowest (24.33 Days) shelf life was declared in controlled fruits (P_0) (Figure 21). This finding is similar with Muangdech (2017). He studied on the effect of aloe vera gel on quality and shelf life of mango (*Mangifera indica* L.) fruits cv.Nam Dok Mai. He discovered that coating with 20% Aloe vera gel gave the longest shelf life with good quality at 14 days at a storage temperature of 25 °C and 75±5 % relative humidity as well as slowing down the weight loss, firmness and changed in chemical composition such as Titratable Acidity (TA) and Total Soluble Solids (TSS) significantly compared to control and other treatment (p ≤ 0.05). These findings are very much similar with the present study. He also explained that the use of the Aloe vera gel coatings did not make alteration to the quality of the fruit when ripe. That"s the reason aloe vera coated fruits showed an excellent result in all temperatures. From the above observation beewax coating performed second highest shelf life. Penchaiya *et al*. (2006) stated that wax coating not only reduces the moisture loss and enhances product by adding a bright shiny appearance, but also protects the fruits from postharvest decay which ultimately extends the shelf life. He also added that small cracks and dents in the rind or skin can be sealed by wax emulsion and establishes a barrier against the entrance of fungal and bacterial pathogens into the product. It also generates a non-water compatible surface which is not conducive to growth and development of pathogens.

The highest (45 Days) shelf life was noted down in T_1 and lowest (12.27) shelf life was found in T_3 temperature (Figure 22). In 0 ( ) C almost all the fruits were remained in their excellent condition except slight spots on the untreated control fruits. This was because of low temperature. It prohibited all the microorganism, spoilage and shrinkage. But the control fruits were slightly spoiled due to lack of any treatment.



P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3: Neem, P_4: Chitosan; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 21: Effect of postharvest bio-preservatives on shelf life of mango



T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; Vertical bars represent the standard errors of the treatment means at $p \leq 0.0$.

Figure 22: Effect of postharvest temperatures on shelf life of mango

The combination effect also showed significant differences among biopreservatives and temperatures. As 0 ( ) C provided with no disease or other quality deterioration due to low temperature so shelf life of treated and non-treated mango fruits all stored in 0 ( ) C showed 45 days. It was also spotted down in combination table. Likeas, P_1T_1 [Aloevera treated fruits in 0 ( ) C], P_2T_1 [Beewax treated fruits in 0 ( ) C] , P_3T_1 [Neem treated fruits in 0 ( ) C] , P_4T_1 [Chitosan treated fruits in 0 ( ) C] and P_0T_1 [Controlled fruits in 0 ( ) C] combinations showed 45 days of shelf life and lowest (9.33 days) shelf life was observed in P_0T_3 [Controlled fruits in 30 (2) C] combination (Table 11).

Table 11. Combined effect of postharvest biopreservatives and temperatures on shelf life of mango fruits

Treatments	**Shelf life (Days)**
P_0T_1	45.00 a^z
P_0T_2	22.67 d
P_0T_3	**9.33 g**
P_1T_1	**45.00 a**
P_1T_2	30.00 b
P_1T_3	15.00 e

P_2T_1	45.00 a
P_2T_2	29.67 b
P_2T_3	14.33 e
P_3T_1	45.00 a
P_3T_2	27.67 c
P_3T_3	11.33 f
P_4T_1	45.00 a
P_4T_2	27.67 c
P_4T_3	11.33 f
LSD (0.01)	1.08
SE	0.39
CV (%)	1.68

P_0: Control, P_1: Aloe vera, P_2: Beewax, P_3:Neem, P_4:Chitosan, T_1: 0 ( ) C, T_2: 20 (2) C, T_3: 30 (2) C; [z]Means with different letters significantly differ at LSD″ s test at P ≤ 0.0 ; CV: Coefficient of Variation; SE: Standard Error; LSD: Least Significant Difference.

CHAPTER V

SUMMARY AND CONCLUSIONS

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 storage temperatures and biopreservatives on shelf life of mango cv. „Amrapali‟ and to evaluate the quality parameters of mango fruits after storage. In this two factorial experiment preservatives were denoted as Factor A and temperatures were denoted as Factor B. Four different postharvest preservatives used in this study are: i) 100% Aloe vera extraction (P1), ii) 6% Beewax emulsion (P2), iii) 40% Neem solution (P3), iv) 2% Chitosan solution, untreated fruits marked as Control (P0) and three different temperatures such as i) 0 ( ) C ii) 20 (2) C and iii) 30 (2) C were used in this experiment. The experiment was laid out in Completely Randomized Design (CRD). In this study observations were made on external and internal fruit attributes, physiochemical properties such as total weight loss, moisture content, pH, total soluble solid content, Ascorbic acid, Visual scoring of mango skin on the basis of shrinkage severity, browning or black spots severity, disease severity and shelf life. In this research work mango 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 elucidated. The results of the experiment expressed that almost all the parameters studied were significantly influenced by the above factors.

Total fifteen postharvest treatments were applied in this experiment with control. Among all those treatments highest total weight loss (3.94%, 5.94%, 7.88%, 10.15% and 12.28% at 3rd, 6Th, 9th, 12th and 15th DAS) was observed in controlled fruits (P_0) and lowest value (1.01%, 2.02%, 2.62%, 4.25% and 5.35% at 3rd, 6Th, 9th, 12th and 15th DAS) was noticed in beewax treated fruits (P_2). The highest moisture content (85.43%) was found in beewax (P_2) and lowest (79.076%) was found in controlled fruits. Again, pH was found to be the highest (5.04) at the end of shelf life in untreated fruits (P_0) whereas beewax coating (P_2) represented the lowest value (4.28). TSS value was mostly influenced by aloe vera extract (P_1) to keep its peak lowest

level (14.89%) and highest value (19.22%) was obtained by untreated controlled fruits (P_0). TA value which was an important quality parameter of mango showed maximum value (0.48%) for both aloe vera (P_1) and beewax (P_2) treated fruits and minimum value (0.36%) for controlled fruits (P_0). Ascorbic acid content was found to be the highest (28.71 mg/100g) at the end of shelf life in case beewax (P_2) treated fruits where controlled treatment (P_0) represented the lowest ascorbic acid content (22.51 mg/100g). However, 6% beewax (T_2) treated fruits represented the highest β-carotene content (3.63 mg/100 g) and controlled fruits represented lowest (2.28 mg/100g) β-carotene content. Shrinkage severity was maximum (7%, 14.56%, 21.33% and 27.22% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) in Controlled fruits (P_0) and minimum (0%, 3.44%, 7.00% and 11.33% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) in Beewax treated fruits (P_2). Maximum browning or black spots (31.44%, 44.56%, 55.44 % and 65.89% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) were also found in Beewax treated fruits (P_2). On the contrary, minimum (3.56%, 7.44%, 13.33 % and 19.56% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) value was found in P_1 (Aloe vera treated fruits). Disease severity was recorded to be significantly maximum (6.67%, 15.00%, 24.33% and 33.44% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) in P_0 (Controlled fruits) fruits and minimum (1.56%, 5.33%, 9.78% and 13.44% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) in P_1 (aloe vera treated fruits). Above parameter indicated that highest shelf life of mango (30 days) was belonged to 100% aloe vera extract (P_1) and 6% beewax emulsion (P_2) treated fruits and lowest (24.33) shelf life was declared in controlled fruits (P_0).

Total weight loss (3.86%, 6.29%, 8.47%, 10.98%, and 13.51% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS), moisture content of pulp (83.338%), pH value (5.41), TSS (19.93%), shrinkage severity (4.40%, 11.47%, 20.40% and 30.20% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS), browning of mango skin (14.73%, 23.80%, 32.07% and 40.8% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) and disease severity (7.87%, 14.93%, 24.13% and 33.73% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) value was found to be the highest in 30 (2) C (T_3) and lowest weight loss (1.05%, 1.43%, 1.64%, 2.93% and 3.17% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS), Moisture content of pulp (81.44%), pH value (3.82), TSS (14.27%), shrinkage severity (0% up to 15^{th} day), browning or black spots severity (5.46%, 12.73%, 18.80% and 23.00 at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) and disease severity (0, 0, 0 and 2.67% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) was recorded in 0 ( ) C (T_1). On the other hand, Highest value of TA (0.54%), Ascorbic acid (42.59 mg/100g), shelf life (45

Days) was found in 0 ( ) C (T_1) and lowest TA (0.35%), Ascorbic acid (14.66 mg/100g), shelf life (12.27 days) was found in 30 (2) C temperature.

The combined effect between the postharvest biopreservatives and temperatures were found that maximum (5.69%, 8.2%, 10.83%, 13.95% and 17.07% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS) rate of weight loss, pH value (5.83), β-carotene (5.67mg/100 g) was observed in P_0T_3 and minimum weight loss (0.22%, 0.54%, 0.54%,1.06% and 1.06% at 3^{rd}, 6^{Th}, 9^{th}, 12^{th} and 15^{th} DAS), pH value (3.64), β-carotene (0.90 mg/100 g) was recorded in P_2T_1. In case of moisture content 86.413% was observed in P_2T_1 and lowest was determined in P_0T_3 combination. Again the significant effect of treatments on TSS gave the maximum value (22.33%) in P_0T_3 and minimum (12.67%) in P_1T_1. In case of interaction effect, highest (0.56%) TA value was recorded in P_1T_1 and the lowest (0.24%) value was noticed in P_0T_3 combination. In the present study, the maximum (48.02 mg/100 g) value of Ascorbic acid was observed in P_1T_1 and minimum (12.31 mg/100 g) in P_0T_3 combination. In case of shrinkage severity maximum value (9.33%, 15.00%, 24.00% and 33.33% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) was determined in P_0T_3. Highest (38.33%, 50.00% 68.33%, 95.00% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) browning or black spot severity was recorded in P_2T_3 and lowest (0, 2.67%, 5.33% and 10.00% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) value was found in $P_1T_{1.}$ Highest (20.33%, 30.67% 45.33% and 65.00% at 6^{th}, 9^{th}, 12^{th} and 15^{th} DAS) disease severity was recorded in P_0T_3 combination but no infection was found in Aloevera, Beewax, Neem and Chitosan treated fruits in 0 ( ) C. Combined effect declared that highest shelf life was recorded in all preservatives at 0 ( ) C treated fruits and lowest (9.33 days) was found in controlled fruits in 30 (2) C.

Conclusion

In an effort to maintain the freshness and quality of mango fruits, beewax seemed to be the best preservative. It effectively reduced weight loss, delayed ripening, locked up moisture, and checked the pH, TA, Ascorbic acid content of mango. But considering the appearance and quality both, Aloe vera should be the best option as it reduced loss of moisture, lowered TSS value, shrinkage severity and disease severity and improved shelf life. Moreover, no skin browning was appeared. So, it can be concluded that Aloe vera in 0 ( ) C (P_1T_1) is best for long time storage, home consumption and the possibility of use in processing industry.

Suggestions

- Further experiment regarding mangoes appearance in beewax treatment, could be done
- Further research could be done on purpose of reduction of disease severity

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APPENDICES

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

Mean square	Degrees of freedom	Mean square of weight loss at different days after harvest				
		3	6	9	12	15
Replication	2	0.02**	0.01**	0.00**	0.06**	0.03**
Factor A	4	10.49**	23.24**	36.77**	42.538**	59.538**
Factor B	2	32.20**	107.26**	203.73**	272.74**	471.112**
AB	8	1.71**	3.27**	5.346**	5.428**	10.267**
Error	28	0.18**	0.19**	0.278**	0.287**	0.503**

**Significant at 1% level of significance

Appendix II: Effect of postharvest biopreservatives and temperatures on moisture content (%) and pH of mango pulp at 15 days after storage

Mean square	Degrees of freedom	Mean square at the end of shelf life	
		Moisture	pH
Replication	2	0.10**	0.00**
Factor A	4	76.9903**	0.8812**
Factor B	2	15.4538**	10.0858**
AB	8	5.2383**	0.0864**
Error	28	0.0996**	0.0071**

**Significant at 1% level of significance

Appendix III: Effect of postharvest biopreservatives and temperatures on TSS, TA, Ascorbic acid and β-carotene of mango pulp at 15 days after storage

Mean square	Degrees of freedom	Mean square at the end of shelf life			
		TSS	TA	Vit-C	β-carotene
Replication	2	0.16**	6.89**	0.39**	0.002**
Factor A	4	27.76**	0.021**	63.57**	2.79**
Factor B	2	123.89**	0.15**	3313.86**	40.01**
AB	8	2.39**	6.78**	9.11**	0.29**
Error	28	0.37**	7.52**	0.41**	0.01**

**Significant at 1% level of significance

Appendix IV: Effect of postharvest biopreservatives and temperatures on shrinkage severity (%) of mango at 15 days of storage

Mean square	Degrees of freedom	Mean square of shrinkage severity (%) of mango skin at different days after harvest			
		6	9	12	15
Replication	2	1.87**	21.36**	64.16**	105.27**
Factor A	4	77.06**	176.30**	478.47**	658.30**
Factor B	2	73.40**	790.16**	2442.82**	4968.60**
AB	8	23.46**	46.77**	129.52**	210.93**
Error	28	2.49**	9.17**	17.54**	37.96**

**Significant at 1% level of significance

Appendix V: Effect of postharvest preservatives and temperatures on skin browning or black spots severity (%) of mango at 15 days of storage

Mean square	Degrees of freedom	Mean square of skin browning or black spots severity (%) at different days after harvest			
		6	9	12	15
Replication	2	42.76**	25.49**	92.29**	178.07**
Factor A	4	1317.06**	2123.19**	3260.64**	5667.13**
Factor B	2	324.02**	480.62**	1093.09**	2767.27**
AB	8	53.02**	49.04**	134.23**	361.85**
Error	28	10.66**	30.20**	35.96**	26.40**

**Significant at 1% level of significance

Appendix VI: Effect of postharvest biopreservatives and temperatures on disease severity (%) of mango at 15 days of storage

Mean square	Degrees of freedom	Mean square of disease severity (%) of mango at different days after harvest			
		6	9	12	15
Replication	2	0.600**	0.16**	0.29**	20.60**
Factor A	4	188.922**	268.30**	482.94**	1190.63**
Factor B	2	233.867**	1184.16**	3646.82**	7304.27**
AB	8	56.089**	68.93**	138.04**	139.93**
Error	28	0.338**	0.35**	0.69**	9.20**

**Significant at 1% level of significance

Appendix VII: Effect of postharvest biopreservatives and temperatures on shelf life:

Mean square	Degrees of freedom	Mean square of Shelf life
Replication	2	0.47**
Factor A	4	11.86**
Factor B	2	88.87**
AB	8	4.01**
Error	28	0.11**

**Significant at 1% level of significance