EFFECTS OF EDIBLE COATING MATERIALS AND STAGES OF MATURITY AT HARVEST ON STORAGE LIFE AND PHYSICOCHEMICAL PROPERTIES OF TOMATO (*Lycopersicon esculentum Mill.)* FRUITS

M.Sc. THESIS

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EFFECTS OF EDIBLE COATING MATERIALS AND STAGES OF MATURITY AT HARVEST ON STORAGE LIFE AND PHYSICOCHEMICAL PROPERTIES OF TOMATO (*Lycopersiconesculentum Mill.)* FRUITS

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BY

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APPROVAL SHEET

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Title: **Effects of Edible Coating Materials and Stages of Maturity at Harvest on Postharvest shelf life and Physicochemical Properties of Tomato (*Lycopersiconesculentum Mill.)*Fruits**.

I have completed my thesis research work as per the approved proposal and it has been evaluated and accepted by my advisers. Hence, I hereby kindly request the Department to allow me to present the findings of my work and submit the thesis.

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DEDICATION

This Thesis is dedicated to my beloved father Abebe Dube.

STATEMENT OF THE AUTHOR

First, I declare that this Thesis is my original work and that all sources of materials used for this Thesis have been duly acknowledged. This Thesis has been submitted in partial fulfillment of the requirements for M. Sc. degree in Postharvest Management at Jimma University and deposited at the University Library to be made available to borrowers under rules of the library. I solemnly declare that this Thesis is not submitted to any other institutions anywhere for award of any academic degree, diploma, or certificate.

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BIOGRAPHICAL SKETCH

The author was born on January 13, 1988 at Arsi, Assela, Ethiopia. She attended her primary school education at Adama No.2 school from 1994 to 2001, and her secondary education at Adama Genet Gebu Secondary School from 2002 to 2003. She attended her preparatory school at Hawas Preparatory School from 2004 to 2005. She joined Jimma University in 2006 and completed her undergraduate studies with B.Sc. degree in Horticulture in June 2008. After her graduation, she was employed as crop production and protection expert in Lanfuro Agricultural office from September 2010/11 to August 2011 and at the Giving Tree Nursery PLC as an agronomist until she joined Jimma University in November 2012 as a postgraduate student with the specialization in Postharvest Management (perishables).

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LIST OF ABBREVIATIONS AND ACRONYMS

FAO	Food and Agriculture Organization
PHM	Post-harvest Management
EC	Electrical Conductivity
PDI	Percent of Disease Index
CRD	Completely Randomized Design
TSS	Total Soluble Solid
TA	Titratable Acidity
RH	Relative Humidity
CSA	Central Statistics Authority
CAS	Controlled Atmosphere Storage
MAP	Modified Atmosphere Packaging
UV	Ultraviolet
LSD	Least Significant Difference
ANOVA	Analysis of Variance
2, 4-DNPH	2,4dinitrophenylhydrazine
MI	Maturity Index
JUCAVM	Jimma University College of Agriculture and Veterinary Medicine

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ABSTRACT

EFFECTS OF EDIBLE COATING MATERIALS AND STAGES OF MATURITY AT HARVEST ON POSTHARVEST SHELF LIFE AND PHYSICOCHEMICAL PROPERTIES OF TOMATO (*Lycopersicon esculentum Mill.)* FRUITS

Tomato (*Lycopersicon esculentum Mill.) is among the most perishable horticultural products and, after harvest management of this crop is a crucial task to minimize losses and extend their shelf life. This work was conducted to determine effect of edible coating material, coated at different harvesting stage of tomato fruits to prolong their storage life with desired physicochemical properties. Fruits of a fresh tomato variety Barbados were obtained from Awassa Jittu farm, Ethiopia. Fruits were harvested at mature-green, turning and light red stages. Treatments from three maturity stages were coated with two types of edible coating materials (Pectin and Chitosan) and laid out in factorial arrangement. of completely randomized design with three replications. Sample fruits were evaluated periodically for different changing parameters with storage time, including weight loss, color, firmness, pH, total soluble solids (TSS), titratable acidity (TA), TSS to TA ratio, ascorbic acid, total phenolic content, lycopene and extent of disease incidence and severity. Results of the study indicated that, coating of tomato fruits either with chitosan or pectin was found to delay the ripening process and maintain fruit quality. All combined treatment combinations resulted in a significant delay in the change of weight loss, TA, TSS, disease incidence, disease severity and ripening index as compared to that of uncoated control fruits. Accordingly the shelf life was extended during ambient storage at average temperature $22^0C \pm 1$ and 74 ± 1 % relative humidity. Maximum shelf life was observed for tomatoes harvested at turning stage coated by pectin (17days) followed by chitosan coated fruit at harvested at turning stage (16 days). Minimum shelf life was for uncoated fruits at the same harvesting stage (10 days). Moreover, in respect of antioxidant properties of certain compounds, coated tomato fruits revealed higher amount of ascorbic acid, lycopene and phenolic contents. Fruits coated with either chitosan or pectin at turning maturity stage showed the best result in almost all quality parameters. Thus, it can be concluded that choosing the optimum stage of maturity of fruits plays a key role in order to achieve the full objectives of coating as it has a great influence on the quality attributes of tomato fruits. The shelf life of uncoated fruits was on average around 10 days as opposed coated fruits stayed sound for about 17 days. This provides an advantage of prolonging the shelf life of tomato fruit by one week with sole application of edible coating materials. The result can be very promising, if coating materials combined with low temperature storage.*

1. INTRODUCTION

Tomato (*Lycopersicon esculentum Mill.)* belonging to the family *Solanaceae* is a major horticultural crop. World production of fresh tomato for 2009 was about 141 million tons planted on 4.5 million hectares in 144 countries (FAOSTAT, 2013). It is one of the most widely consumed, being the second most important vegetable crop next to potato worldwide (Pantheen and Chen, 2010). Tomato is among the most important vegetable crops also in Ethiopia and both fresh and processed tomato varieties are popular and widely produced (Menberu et al., 2012). Its production has shown a marked increase and it became the most profitable crop providing a higher income to farmers compared to other vegetables (Lemma et al., 1992). In Ethiopia it is grown on 5338 ha with total production of 55,635 tones (CSA, 2011).

Tomato is one of the vegetable crops which is widely consumed either raw or after processing and can provide a significant proportion of the total antioxidants in the diet (Martinez-Valverde et al., 2002). It is known as one of the health stimulating vegetables because of the antioxidant properties of certain compounds. Antioxidants include vitamins C and E, lycopene, ß-carotene, flavonoids and other phenolic compounds (Dumas et al., 2003). It is well-known that phenolic compounds contribute to fruit quality and nutritional value in terms of modifying color, taste, aroma, and flavor, and also in providing beneficial-health effects (Hagen et al., 2007). These days the society can be characterized by having many unhealthy dietary habits. Inadequate intake of healthy foods triggers major dietary imbalance, this being a major cause of chronic diseases such as obesity, diabetes mellitus, cardiovascular disease, hypertension, stroke, and several types of cancer. Therefore, it is vital to ascertain the composition and nutritional value of these products (Mertz et al., 2009).

The nutritional value, color, and flavor of tomato fruits depend mainly on lycopene, β-carotene, ascorbic acid, sugars, acid and their ratio. The two most important carotenoids in fruits of tomato are lycopene, which determines 80- 90% of fruits red color, and β-carotene, which accounts for approximately 7% of the tomato carotenoids (Frusciante et al., 2007).

The lycopene content varies significantly with ripening and variety of tomato (Martinez-Valverde et al., 2002).

Consumers judge the quality of fresh tomatoes by their firmness, appearance, color and flavor, which are related to ripeness and shelf life. Sugars, acids, phenols and minerals are the main constituents of tomato taste, quantitatively, making the largest contribution for fresh tomatoes. The two quality attributes that are most important to buyers and consumers are texture and skin color. Softening during storage, distribution and ripening of tomatoes can be a major problem because it may increase their susceptibility to damage (Kader, 2008).

Proper harvesting stage determines the nutrient contents as well as storage durability of any fruit. The shelf life of tomato depends on postharvest conditions and also on the time of harvest. It was found that maturity stage is an important factor that influences the consumer preferences. The postharvest quality and storage life of fruits appear to be controlled by the maturity (Casierra-Posada and Aguilar-Avendaño, 2009). Depending on the market and production area, tomatoes can be harvested at different stages of maturity from mature-green stage to full ripe depending on the purpose of use. Tomato is usually harvested unripe at mature green stage or breaker stage in order to reduce their losses caused by physical damage throughout handling and transport and are then allowed to ripen just prior to or during marketing (Liplap, 2013). Therefore, maturity stage at harvest is one of the most important determinants of storage life and final fruit quality

Postharvest handling of tomato fruits is important because of their perishable nature. As such they continue to undergo both desirable and undesirable changes during handling. Since tomato fruits are highly perishable they encounters several problems during transportation, storage and marketing. There is a high production of tomato fruits during the harvest time, but post-harvest processing and preservation techniques are inefficient. Therefore, fruits spoil very early because of lack of appropriate systems of preservation and processing (Ameyapoh et al., 2008). Major losses in tomato occur mainly between harvest and consumption. Apart from physical quality, serious losses also occur in the essential nutrients, vitamins and minerals. Uneven handling of tomato fruits can result in damage of the fruit cell wall leading

to softening and reduced marketability of the product. The morpho-physiological nature of tomato fruits (high moisture content, high respiration rate, high organic acid content, soft texture) tend to predispose the product for microbiological, biochemical and mechanical damages. Substantial postharvest losses of tomato fruits in terms of quality and quantity may be encountered due to lack of appropriate postharvest techniques as well as the lack of postharvest facilities (Mutari and Debbie, 2011).

Tomato fruit has relatively short postharvest life since many processes affecting quality loss take place after harvest. Thus, the major limiting factors in the storage of tomato fruit are transpiration, fungal infection and high rate of respiration, which result in the early deterioration of fruit quality acceleration of the ripening process and senescence (Zapata et al., 2008). Moreover, during ripening the chemical composition of the fruit also changes dramatically, affecting texture, taste, flavor, antioxidant contents mainly phenolic compounds, flavonoids and ascorbic acid (Bailen et al., 2006). In tropical countries about 40-50% of post-harvest losses of tomato fruits occur between harvesting, transportation and consumption of fresh tomato (Kadir, 1992). This large annual loss of tomato fruits makes the control of the ripening process to have great economic importance (Hoberichts et al., 2002).Therefore, postharvest losses have great economic implications which do not only affect the local farmers but also the economy of the entire nation.

Postharvest technology has great importance in preventing both qualitative and quantitative losses in fruits and vegetables, which is high in developing countries. Tomato fruits deteriorate rapidly after harvest and, in some cases, do not reach consumers at optimal quality after transport and marketing. Tomato fruit ripening also can be controlled through the use of gas, temperature and humidity control. Delaying the fruit ripening process would allow producers more time for shipment and increase the shelf life of the fruit for consumers. Packaging is widely used for preserving, distributing and marketing fruit and vegetables and is often used in combination with other preservation methods (Hoover, 1997). However, the disposal of packaging materials leads to ecological problems and additional recycling costs. Thus, adequate postharvest technologies are needed. Low temperature storage is used to reduce the rate of respiration and thermal decomposition for extending storage life

of tomatoes. However, the prolonged storage at low temperature causes chilling injury and also contraction of the skin occurs as water from the skin of the fruit moves into the pulp which lowers down the taste and also damages the fruit physiology (Zapata et al., 2008). Controlled atmosphere and hypobaric storage techniques are also useful in extending the shelf-life of tomatoes but these are very expensive to run on a commercial scale (Artes et al., 2006).

In this sense, in recent years, there is an increasing interest in the use of edible coatings to maintain fruit quality with the additional benefit of reducing the volume of non-biodegradable packaging materials (Tzoumaki,et al., 2009). Edible coatings can serve as alternatives to extend the postharvest life of fresh fruits and vegetables and can also result in the same effect as modified atmosphere storage where the internal gas composition is adjusted (Park, 1999). Edible coating is a transparent film that covers the food item to generate a modified atmosphere by creating a semi-permeable barrier against O_2, CO_2, moisture and solute movement, thus reducing respiration, water loss and oxidation reaction rates, hinder solute movement, reduce metabolism, seal in flavor volatiles and improve the appearance (Arvanitoyannis and Gorris, 1999).

The use of edible coating has received more attention in recent years, due to the growing interest for reducing environmental pollution caused by plastics, the need to extend the shelf life of foods, and the increasing demand for healthier and ecological foods (Espino-Díaz et al., 2010). In developing countries there is an interest in simple, low-cost alternatives and environmentally friendly technologies, e.g. using edible coatings. On the other hand, edible coatings are also effective as postharvest treatments to preserve fruit quality, with the additional benefit of reducing the volume of non-biodegradable packaging materials (Baldwin et al., 1995) So the use of edible coatings appears to be a good alternative. Therefore objectives of this work were:

General objective

To determine combined effects of best harvesting stage and edible coating material for better quality and storage stability of tomato fruits.

Specific objectives

i. To identify type of edible coating material(s) for better storage stability and quality of tomato fruits.
ii. To determine right harvesting stage of tomato fruits which responds better for type of edible coating material for a better quality and shelf life extension

2. LITERATURE REVIEW

2.1 Pre harvest factors affecting the quality of tomato

Quality management starts in the field and continues until produce reaches the end user. The response of fruit and vegetables during storage to post-harvest factors also in part depends on pre-harvest practices such as use of natural plant extract such as compost, manure and environmental factors. Understanding and managing the various roles that pre harvest factors play on quality is very important in order to achieve maximum harvest and post harvest quality for any crop. Mostly, pre harvest conditions are of overriding importance in determining storage behavior (Fischer and Richter, 1986).

Post harvest qualities of tomatoes partly depend up on pre harvest factors such as cultural practices, genetic and environmental conditions. Cultural practices such as nutrient, water supply and harvesting methods quality of tomato before and after harvest (Fischer and Richter, 1986).

2.1.1. Genetic Factors

Quality factors are reported to be more or less genetically controlled. Genotype has an important role in fruit quality, nutrient composition and postharvest life potential (Scalzo and Mezzetti, 2010). Internal factors such as genotype and fruit maturity stage affect the expression of genes, enzymes and metabolites (Carbone et al. 2009). Several studies have underlined the primary role of genetic control both of health and taste related compounds in fruits. Soluble solids content and acidity are determined by several factors such as cultivar (Byrne, 2003). Inconsistencies exist within cultivars in their quality traits which could be attributed to plant individual differences or to changes in the fruit quality during the harvest period. Cultivar selection is important because there are often differences in raw fruit composition, durability, and response to processing (Kader, 2002). In many cases, fruit cultivars grown for fresh market sale will not be the optimal cultivars for processing. Tomatoes vary in size, shape, sugar content, acidity, dry matter, resistance to pests and diseases, susceptibility to handling damage and rate of postharvest ripening within cultivars.

They also vary in their ability to achieve the desired phenotype under differing production conditions (Kader, 2002).

2.1.2 Climatic Conditions

Climatic factors, in particular temperature, relative humidity and light intensity, have great impact on the nutritional quality of fruits and vegetables. Consequently, the location of production and the season in which plants are grown can determine their ascorbic acid, carotene, riboflavin, thiamine, and flavonoid contents (Knee, 2002). The general heading "Environmental response" has been used to describe the factors affecting plant growth and development such as temperature, water and nutrient requirements and, in some instances; day length and light intensity (Tindall, 1983).

The tomato plant as a tropical one needs a sufficiently high temperature to ensure completion of its life cycle and full fruit maturation. The duration of tomato cultivation depends mainly on climatic conditions. The environmental conditions of most concern are temperature, and relative humidity. Ideal temperatures for optimal tomato plant growth are 70 to 82^{o}F for day and 62 to 64^{o}F for night. This ideal temperature is determined by long-term averages rather than instantaneous temperatures. Periods of low temperature can be compensated for by periods of high temperature, keeping the long-term averages in the optimal range for growth (Zhang, 2010).

The optimal relative humidity levels for greenhouse tomatoes are between 60% and 70%. Relative humidity (RH) affects the transpiration rate of plants, and therefore affects uptake of water and nutrients, mainly nutrients transported through xylem like calcium and potassium. High humidity significantly reduces the hourly and daily transpiration rates and reduces crop yield. Tomato plants can be grown on many different soil types, but a deep, loamy, well drained, slightly acid soil with a pH of 6.2 to 6.8 and supplied with organic matter and nutrients is the most suitable (Pediaditakis, 1997).

Tomato has a high water requirement throughout the growing period, until fruiting occurs. Uneven levels of water application may lead to physiological disorders such as cracking and

splitting of the fruit skin. These components may be used as a general guide to both aerial and root environment conditions which affect tomato growth and development, although cultivar variation in response to climatic conditions.

2.1.3 Cultural Practices

Soil type, mulching, irrigation, and fertilization influence the water and nutrient supply to the plant, which can in turn affect the nutritional quality of the harvested plant part (Kader, 2002). The effects of mineral and elemental uptake from fertilizers by plants are, however, significant and variable. High calcium uptake in fruit has been shown to reduce respiration rates and ethylene production, delay ripening, increase firmness, and reduce the incidence of physiological disorders and decay, all of which result in increased shelf life. High nitrogen content, on the other hand, is often associated with reduced shelf life due to increased susceptibility to mechanical damage, physiological disorders, and decay (Kader, 2002).
Among agronomic factors affecting tomato composition and flavor are water availability, soil fertility and potassium (Stevens, 1995). Careful water management could result in an increase in fruit solids. In addition there is a positive relationship between nitrogen availability and soluble solids content. Addition of potassium fertilizers increased the acid content of tomato fruits

2.1.4 Maturity stage at harvest

Deciding when to harvest a crop is often one of the most difficult decisions that a grower has to make. Often, this decision is made by pickers who are not always familiar with crop development. Maturity at harvest has a very important influence on subsequent storage life and eating quality in particular for climacteric fruits where ripening is regulated by ethylene. (Watada et al., 1984).

In postharvest physiology mature and ripe consider to be distinct terms for different stage of fruit development. Mature fruit having completed natural growth and development, for fruit, it is defined as the stage which will ensure proper completion of the ripening process. Most postharvest technologist consider that the definition should be, the stage at which a

commodity has reached a sufficient stage of development that after harvesting and postharvest handling (including ripening, where required), its quality will be at least the minimum acceptable to the ultimate consumer (Kader,1999). It's also important for deciding when a given commodity should be harvested to provide some marketing flexibility and to ensure the attainment of acceptable eating quality to the consumer (Dhatt and Mahajan, 2007).
In many fruits, such as mature (but green) tomato, the eating quality at maturity will be far less than optimal. The fruit becomes edible only after proper ripening has taken place (Kader, 1999).

The stage of fruit development at harvest is one of the major factors determining the quality of tomato fruit and its shelf life. Maturity is one of the major factors that determine the compositional quality of fruits (Lee and Kader, 2000). While ripening, the concentration of sugar, carotenes, ascorbate, rutin and caffeic acid increased whereas those in titratable acidity, chlorophylls, chlorogenic acid contents decreased which are major factors to tomato fruit quality (Gauiter et al., 2008).

The physiological maturity of the fruit at harvest is a major determinant of quality. Harvesting immature fruit curtails sugar import, and makes the postharvest degradation of starch the primary source of carbohydrates, which is both inadequate and undesirable (Balibrea et al., 2006). While picking the fruit at a later stage would permit greater sugar accumulation riper fruit is easily damaged and also has a short shelf-life. Determining the best time to harvest fruit from an eating quality perspective while reducing physical damage is not easy and varies by cultivar (Casierra-Posada and Aguilar-Avendano, 2009). Other factors to consider during harvesting include the mode of consumption, distance and time to market, and, the handling and production system (Watkins, 2006). As per USDA (1991), there are six Maturity and Ripening Stages of Tomato that are:-
Green: The tomato surface is completely green. The shade of green may vary from light to dark.
Breakers: There is a definite break of color from green to bruised fruit tannish-yellow, pink or red or 10% or less of the tomato surface.

Turning: Tannish-yellow, pink or red color shows on over 10% but not more than 30% of the tomato surface.

Pink: Pink or red color shows on over 30% but not more than 90% of the tomato surface.

Light red: Pinkish-red or red color shows on over 60% but red color covers not more than 90% of the tomato surface

Red: Red means that more than 90% of the tomato surface, in aggregate, is red

2.2 Handling

Bruising and mechanical damage to fruit occurs before, during, and after harvesting and drastically reduce quality. Tomatoes in industrial production systems may be harvested mechanically at Mature Green, packed into crates, sorted, sized, washed, cooled, stored and transported over long-distances. At each stage there are significant opportunities for mechanical damage to fruit, including bruising, scarring, scuffing, cuts and punctures (Prudent et al., 2009). The effects of physical injury are cumulative. Injury near or greater than the bio-yield point leads to cell lysis followed by unwanted chemical reactions, accelerated transpiration, respiration, ethylene production and pathogen infestation the severity of which is determined by the extent of damage (Miller, 2003). Fruit may experience internal or external injury, or both. Internal injury however may go undetected but still lead to massive fruit loss (Lee et al., 2007). Most of postharvest practices are usually more injurious and cause severe problems.

2.2.1 Major post harvest techniques being used to keep quality and extend shelf life

Harvested fruits and vegetables continue to maintain physiological activities and sustain metabolic processes which were there before harvest. Product respiration, transpiration, and ethylene production are major factors contributing to the deterioration of fresh fruits and vegetables. Reduction of these processes by technologies such as cooling, storage temperature and relative humidity management, modified and controlled atmosphere storage and 1-Methylcyclopropene (1-MCP) treatment are major techniques which help to enable the postharvest life of fresh produce to be prolonged. The climacteric burst of ethylene which

makes the fruit palatable also promotes senescence, and a goal of postharvest practices is to manage the concentration and timing of ethylene synthesis so that the fruit reaches the consumer at optimal eating quality (Lee and Kader, 2000).

2.2.1.1. Storage temperature and RH management

Temperature management is the most important tool to extend shelf-life and maintain quality of fresh fruits and vegetables. Keeping the fresh appearance of fresh fruits and vegetables after harvest is the permanent challenge imposed by both producers and consumers. One of the ways in which their quality can be kept is by controlling storage temperature. Delays between harvesting and cooling or processing can result in indirect losses such as those in flavor and nutritional quality (Lee and Kader, 2000). Temperature greatly influences the rate of respiration and transpiration of fruits and vegetables, and is certainly one of the most important factors in maintaining post harvest quality of tomato fruits. The temperature at which a commodity is stored is usually very specific to that particular product. If storage temperatures are too low, chilling injury may result. However, if temperatures are too high, metabolic processes can accelerate. In addition, a wide range of storage temperatures is also not advisable, because such conditions lead to rapid weight loss of produce. A storage temperature of 10-15°C could extend the postharvest life of fruits. At these temperatures chilling injury and ripening rate are minimal. The injury is generally followed by an increased tendency to decay, particularly when the temperature is raised (Castro et al., 2005).

Altering the relative humidity (RH) of the storage environment may also delay senescence. Perishable fruit and vegetable products should be maintained at RH levels of 90-95%. This high humidity level prevents moisture loss that may occur due to increased respiration and transpiration. Higher relative humidity increases the vapor pressure of the air and decreases physiological weight loss of commodities (Getinet et al., 2008) and limits the migration of water molecules from the product to storage room air. However, humidity levels should not exceed 95% because growth of microorganisms may be enhanced (Salunkhe et al., 1991).

2.2.1.2 Controlled atmosphere storage

Controlled atmosphere storage implies precise control of the gas concentrations of oxygen, carbon-di-oxide and ethylene inside the storage room. The controlled atmosphere storage has oxygen, carbon-dioxide and ethylene control systems which are used in order to monitor the concentrations of the gases in the chamber during the period of storage.

Keeping harvested fruit in a controlled atmosphere can help to reduce ethylene-related deterioration and also Modification of atmospheric gas levels may reduce the respiration rate of fresh produce. The gas constituents of controlled atmosphere (CA) are more precise and stable. CA may be achieved in large facilities such as storage rooms and transport vessels or in individually wrapped containers using specialized package coatings (Beaudry, 2010). In CA, carbon dioxide is increased and oxygen is decreased with the objective of reducing the rate of respiration and extending shelf life. The effectiveness of the approach depends on the variety, fruit maturity and initial quality, storage temperature, and the composition and duration of exposure to CA (Brecht et al., 2003). Low oxygen can harm the fruit by stimulating anaerobiosis (Kader and Saltveit, 2003) after the extinction point. In order to control respiration (i.e., transfer of various gases in and out of the product), food can be stored in an environment filled with various gases at appropriate, optimal temperatures.

The right gas combination can slow respiratory metabolism, and delay compositional changes in color, flavor and texture. It can also inhibit or delay microbial growth. However, this method can be quite expensive in other than large-scale stationary storage (Embuscado and Huber, 2009). CA storage of tomatoes delayed the onset of climacteric stage and slowed down the rate of respiration. During first 3 weeks of storage in CAS, respiration rate of tomatoes were reduced to about 10 percent of normal respiration (Bhowmik and Pan, 1992). Controlled atmosphere storage of tomatoes at 12°C significantly reduced the weight loss. The fruits had a higher titratable acidity and low TSS. Loss of ascorbic acid content was slower and lycopene synthesis was delayed in addition to increase in the storage life to 40 days (Tasdelen and Bayindirli, 1998).

2.2.1.3 Modified atmosphere packaging (MAP)

Modified atmosphere packaging (MAP) is where a product is enclosed in a sealed box or bag filled with required atmosphere. It is a packaging technology that modifies or alters the gas composition around the products in food packages from normal air (20.95% O_2 , 78.09% N_2, 0.93% argon, and 0.038% CO_2) to provide an atmosphere for increasing shelf life and maintaining the quality of food through controlling rate of respiration. MAP for fresh fruits and vegetables are much more challenging and complicated. Because fresh fruits and vegetables are still alive after harvesting and during marketing, the successful use of MAP will be based not only on the specific O_2 and CO_2 permeation properties of polymer films but also on the respiration activity of packed food (Kader, 1986). The composition of the air in the package changes as a result of the respiratory action of the produce and permeability characteristics of the membrane.

MAP refers to the use of specialized material to enclose a product in an altered composition of gases after which there is no active efforts to modify the environment. The polymers and films used for MAP typically allow free diffusion of gases, which maintains equilibrium between the external atmosphere gas composition and that inside the package due to tissue respiration (Philips, 1995). The most commonly used materials are low density polyethylene terephthalate, polypropylene, polyvinyl chloride and polystyrene (Sandhya, 2010) and chemically modified derivatives thereof. MAP is a better approach for short term storage of small quantities of produce than controlled atmosphere storage and is often used in association with packaging. Besides being able to provide MA and control ripening, there are a number of positive benefits of MAP. These include reducing water loss, better sanitation, and, reduced bruising and spread of disease (Bailen et al., 2006).

Mature green tomatoes packed in MAP had a built in atmosphere of 4 per cent O2 and 5 per cent CO2 and delayed the fruit ripening. These fruits had a low rate of physiological loss in weight and better overall quality than control (Onwuzulu et al., 1995). Tomatoes packed with several polyvinylchloride (PVC) films had a slow rate of colour change than control, but continued to ripe normally after the packs were perforated and transferred to 20°C. The

aroma, flavor and texture of these fruits were slightly better than control fruits (Geeson et al., 1985). Modified atmosphere packs sealed with breaker tomatoes delayed the changes in acidity, soluble solids, texture and color. It also resulted in a substantial reduction in fruit weight loss and spoilage (Nakhasi et al., 1991). Tomatoes enclosed in polyethylene bags and kept at low temperature resulted in the buildup of modified atmospheres and extended the ripening time, improved firmness and maintained quality in terms of appearance and taste (Hobson, 1981).

2.2.2 Other physical treatments

2.2.2. 1 Heat treatment

There has been increasing interest in the post harvest heat treatment (thermotherapy) of vegetables and fruits to control insect pests, prevent fungal decay, and to modify the ripening of commodities (Lurie, 1990). This is primarily because heat treatment substitutes as non damaging physical treatment. It is a non carcinogenic, non polluting, non damaging treatment for prevention of chilling injury and maintainace of fruits and vegetable (Akbudak et al., 2007). Methods for heat treatment of harvested fresh fruit and vegetables include hot water, vapor heat and hot air. High temperatures are known to inhibit ethylene production in tomatoes. Exposing tomato fruit to higher temperature (37–42 ◦C) before cold-storage may delay ripening and enhance pathogen resistance (Akbudak et al., 2007) and is one approach to reduce the occurrence of chilling injury (Lu et al., 2010).

2.2.2.2 Irradiation.

Irradiation is classified as non-ionizing or ionizing where the latter is high frequency and causes loss of ions from the material with which it comes into contact. At hermetic doses fruit tissues are able to deploy a range of protective mechanisms including the productions of antioxidants, which are healthful to humans (Sharma, 2004). Radiation can also minimize the colonization of fruit with pathogens due to contamination, insect infestation, postharvest

disease, as well as delay ripening (Allende et al., 2006). Ionizing radiation of 0.15-0.75 kGy has been proposed for insect disinfection (Tilton and Burditt, 1983).

2.2.2.3. Edible Coatings

Edible coating consists of a thin layer of protective that is applied to the skin surface of the fruit which is later consumed together with the fruit flesh. Edible coating is defined as a thin layer of edible material form as a film on the surface of the fruits and vegetables. This coating can affect the respiration and moisture loss (Ghasemzadeh, 2008). Any type of material used for enrobing (i.e., coating or wrapping) various food to extend shelf life of the product that may be eaten together with food with or without further removal is considered an edible film or coating. 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 (Bal, 2013). They are applied directly on the food surface by dipping, spraying or brushing to create a modified atmosphere (McHugh and Senesi, 2000). An ideal coating is defined as one that can extend storage life of fresh fruit without causing an anaerobic and reduces decay without affecting the quality of the fruit (El Ghaouth et al., 1992). The effect of coatings on fruits and vegetables depends greatly on temperature, alkalinity, thickness and type of coating and variety and condition of fruits (Guilbert et al., 1996).

A. Types of edible coating materials

Edible films can be produced from materials with film forming ability. During manufacturing, film materials must be dispersed and dissolved in a solvent such as water, alcohol or mixture of water and alcohol or a mixture of other solvents. Plasticizers, antimicrobial agents, colors or flavors can be added in this process. Adjusting the pH and/or heating the solutions may be done for the specific polymer to facilitate dispersion. Film solution is then casted and dried at a desired temperature and relative humidity to obtain free standing films (Bourtoom, 2008).

The edible films are classified into three categories taking into account the nature of their components: hydrocolloids (such as proteins, polysaccharides, and alginate), lipids (such as fatty acids, acylglycerol, waxes) and composites (made by combining substances from the two categories) (Donhowe and Fennema, 1993).

B. Mechanism of action of coating materials

The principle functions of edible coatings are to restrict the loss of moisture from the fruit to the external environment and to lessen the absorption of the oxygen by the fruit. coatings can help retard this movement of water vapor but become more permeable to water vapor and gases under conditions of high RH as explained above. Orientation of polymers to the flow of permeate can affect permeability properties. Water loss usually occurs in the vapor phase. Water vapor permeability describes the movement of water vapor through a film or coating per unit area and thickness, and determines the vapor pressure difference across the film at a specific temperature and humidity. Creation of a Modified Atmosphere for Coated Fresh Produce and Effect on Ripening Cells of plant tissues, such as harvested fruits and vegetables, are physiologically active in that they consume oxygen (O_2) and produce carbon dioxide (CO_2) as they respire (Baldwin et al., 1997). Coatings preserve the texture, aroma and flavor of the fruit by reducing the respiration rate and providing physical protection to the food product especially during handling and transport. They are alternatives to modified atmosphere packaging (MAP) to improve the shelf-life of fruits and reduce the deleterious effects not only retarding food deterioration and enhancing its quality, but also improving its safety because of their natural biocide activity or by incorporating antimicrobial compounds (Petersen et al., 1999).

a. Polysaccharides

Polysaccharides used for edible films or coatings include cellulose, starch derivatives, pectin derivatives, seaweed extracts, exudates gums, microbial fermentation gums and chitosan (Krochta and Mulder-Johnson, 1997). Polysaccharides may be regarded as condensation polymers of monosaccharides resulting in the formation of glycosidic linkages by elimination of water. As components of almost all living organisms, polysaccharides, also called hydrocolloids, are most abundant in the higher order of land plants and in

seaweeds where they constitute approximately three-quarters of the dry weight. They perform diverse roles in the physiology of plants, animals, and microorganisms (Tharanthan and Saroja, 2001). The development of coating from water soluble polysaccharides has brought a surge of new types of coatings for extending the shelf life of fruits and vegetables, because of their selective permeability to CO_2 and O_2 resulting in modified internal atmosphere and delayed ripening. This property is probably related to the dense structure and high polarity of the film. Polysaccharides are abundantly available, usually are of low cost, and are non-toxic (Tharanthan and Saroja, 2001).

i. Pectin

Pectin is commercially produced from citrus peel as a by-product from extraction of lime, lemon and orange juices; or from apple pomace, the dried residue remaining after extraction of apple juice (Embuscado and Huber, 2009).It is a class of complex water-soluble polysaccharides used to form coatings. It is a purified carbohydrate product obtained by aqueous extraction of some edible plant material, usually citrus fruits or apples.

Pectins are high molar mass hetero-polysaccharides with at least 65 % of a-(1→4)-linked d galacturonic acid-based units. These units may be present as free acid, salt (sodium, potassium calcium, ammonium), naturally esterified with methyl group, or as acid amid in a midated pectins (Lopez da Silva and Rao 2006). Furthermore, a range of neutral sugars such as l-rhamnose, d-galactose, l-arabinose, d-xylose, and small amounts of others are part of the polymer chain. l-rhamnose units exist exclusively as (1→2)-linked in the main chain, whereas all other neutralSugar residues are bond preferably at the rhamnose and galactose units to the main chain. Pectins can differ by the degree of esterification of the carboxy groups of the galacturonic acid, which is in general in the range of 20–80%. Pectins with more than 50% esterification are designated as high-esterified (high methoxylated) and distinguished from low-esterified pectins (low methoxylated) with less than 50% ester groups. Pectins are soluble in water but insoluble in most organic solvents (Baldwin et al., 1997).

Pectin is a high-volume and potentially important food ingredient available in high percentages in agricultural wastes. In addition, its nutritional benefits for human health and its

pharmaceutical activities make it interesting to use in a variety of food products (Moalemiyan et al., 2012). Pectin coatings have been also studied for their ability to retard lipid migration and moisture loss, and to improve appearance and handling of foods (Ayranci and Tunc, *2004).*

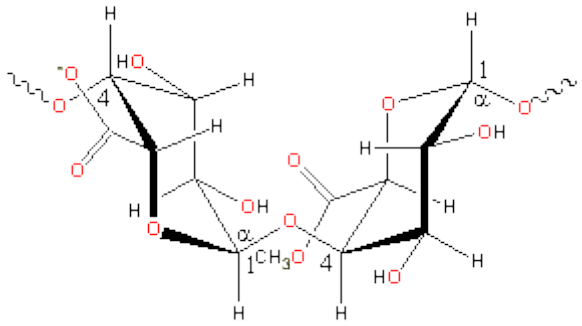

Figure 1*: Structure of pectin*

ii. Chitosan

Chitosan is the principal derivative of chitin, the material comprising the exoskeletons of crustaceans and mollusks, and is produced by alkaline deacetylation of chitin. It is an edible and biodegradable polymer derived from chitin. Next to cellulose, chitosan is the most abundant natural polymer available (Vartiainen et al., 2004). Chitosan, a linear polysaccharide consisting of (1, 4)-linked 2-amino-deoxy-β-D-glucan, is a deacetylated derivative of chitin, which is the second most abundant polysaccharide found in nature are cellulose. Chitosan has been found to be non-toxic, biodegradable, bio functional, and biocompatible, and is reported by several researchers to have strong antimicrobial and antifungal activities. Typical commercial chitosan is about 85% deacetylated. In solution, chitosan forms micelle-like aggregates from fully acetylated segments of polysaccharide chains, interconnected by blocks of almost fully deacetylated polysaccharide, stretched by electrostatic repulsion (Pedroni et al. 2003). Chitosan in the free amine form is insoluble in water at neutral pH. However, it is soluble in glacial acetic acid and dilute HCl. Chitosan carries a large number of amino groups along its chain and is, thus, capable of forming multiple complexes. At acid pH, the protonation of –NH 2 groups

converts them to $-NH\ 3^{+}$, which can associate with polyanions to form complexes and bind anionic sites at bacterial and fungal cell wall surfaces (Embuscado and Huber, 2009).

At higher pH levels (>4), chitosan can form complexes with colorants and heavy metals. These appealing features make chitosan widely applicable in wound healing, production of artificial skin, food preservation, cosmetics, and waste water treatment (Juang and Shao 2002). Some desirable properties of chitosan are that it forms films without the addition of additives, exhibits good oxygen and carbon dioxide permeability, as well as excellent mechanical properties (Suyatma et al., 2004). However, one disadvantage with chitosan is its high sensitivity to moisture. Chitosan also inhibits a number of microorganisms also exhibits antimicrobial activity against bacteria yeasts, and molds and can produce semi-permeable coatings (Vartiainen et al., 2004). Considering these superior properties of chitosan, it has been successfully used in many postharvest aspects of fruit and vegetables (Youwei and Yinzhe, 2013). Nowadays many reports involving chitosan coating mostly focus on the varieties of fruit and vegetable or compound coating based on chitosan (Riccardo et al., 2012).

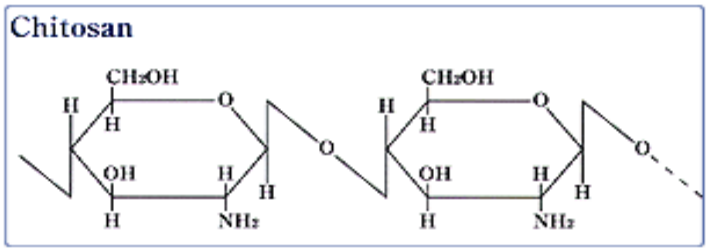

Figure 2: *Structure of chitosan*

b. Lipid coatings

Lipid compounds utilized as protective coating consist of acetylated monoglycerides, natural wax, and surfactants. The most effective lipid substances are paraffin wax and beeswax. The primary function of a lipid coating is to block transport of moisture due to their relatively low polarity. In contrast, the hydrophobic characteristic of lipid forms thicker and more brittle films (Bourtoom, 2008). Consequently, they must be associated with film forming agents such as proteins or cellulose derivatives (Debeaufort et al., 1993).

c. Composite coatings

Edible films and coatings may be heterogeneous in nature, consisting of a blend of polysaccharides, protein, and/or lipids. This approach enables one to utilize the distinct functional characteristics of each class of film former (Kester and Fennema, 1986). As composite films consisting of lipids and a mixture of proteins or polysaccharides it takes advantage of the individual component properties. The combination between polymers to form films could be from proteins and carbohydrates, proteins and lipids, carbohydrates and lipids or synthetic polymers and natural polymers (Guilbert, 1986). A gluten coating reduced softening and weight loss of strawberries, especially when lipids (beeswax, stearic, and palmitic acids) were incorporated. However, the lipid addition impaired the acceptance of the strawberries in terms of appearance and flavor (Tanada-Palmu and Grosso, 2005). Similarly, Vargas et al. (2006) observed that, although the addition of a lipid component (oleic acid) has improved the water vapor resistance of chitosan-coated strawberries, it has decreased their acceptance.

2.3 Physiological changes of tomato during fruit ripening

2.3.1 Respiration

Respiration is the chemical process by which fruits and vegetables convert sugars and oxygen into carbon dioxide, water, and heat. The heat generated by the respiration process tends to increase the temperature of the commodity. This, in turn, increases the water vapor pressure just below the surface of a commodity, leading to increased transpiration (Sastry et al., 1978). Fruit have been classified as climacteric or non-climacteric based on their respiratory and ethylene production patterns during ripening and their response to exogenous ethylene. In Early development of climacteric fruits such as tomato, the respiration rate is high and decreases to a pre-climacteric minimum during maturation. At the onset of tomato ripening, respiration increases to a maximum, the climacteric peak, before it subsequently declines slowly. This respiratory peak is preceded by or associated with a rise in ethylene production. At the pink-red stage, the climacteric process of respiration reaches the maximum level. Once

the fruit become fully ripe the rate of respiration also declined though respiration rate is one of the most important indicators of senescence in tomato fruit (Wills et al., 1998).

Harvested commodities continue to respire aerobically. However, the act of harvesting a product does create some disturbances in the normal respiration patterns. One of the main changes in the respiration of a harvested fruit is the alteration of the fruits' internal atmosphere. Normally, prior to harvest, the external tissues of a fruit are exposed to atmospheric concentrations of oxygen, nitrogen, and carbon dioxide. When the fruit is picked the protective outer cellular layer, known as the cuticle barrier, is disrupted and the gases once confined are now free to escape. During this escape a large influx of oxygen from the outside environment occurs in addition to an outflow of carbon dioxide. In the new environment containing higher oxygen and lower carbon dioxide concentrations, the respiration rates of the internal cells are no longer suppressed and respiration increases. The rapid respiratory rise depletes the metabolites used in the respiratory processes, and along with that depletion an increase in all oxidative processes occurs, which in turn will serve to hasten the fruits' ripening and eventual senescence (Phan, 1987).

2.3.2 Ethylene production

The hormone ethylene is a normal physiological product of fruit. Upon exposure to the hormone, climacteric fruits have increased respiration rates, which eventually decrease the time it takes for them to reach their climacteric respiration peak (Kader, 1987).Tomatoes are one of climacteric fruits which have a relatively short postharvest life due to high ethylene production. In climacteric fruits such as tomato, ripening process is affected by the rate of ethylene production (Carrari and Fernie, 2006). Inhibition of ethylene production delays the fruit ripening process, and increases the shelf life of the fruits for the consumer (Madhavi and Salunkhe, 1998).

Ethylene synthesis begins to increase at the onset of ripening. This takes place before any external color change at the blossom-end of green fruit becomes noticeable and precedes the synthesis of enzymes such as polygalacturonase. The climacteric peak in ethylene evolution

occurs between the ‘mature-green’ and ‘pre-breaker’ stages. Ethylene is responsible for initiating certain enzymes, including chlorophyllase and peroxidase, which once activated can alter certain fruit components, including degradation of chlorophyll. Upon ripening, ethylene production rate increases so that it accelerates to severity of changes and reduction of quality (Giovannoni, 2001). This causes changes in fruit sugar content and increases in organic acids metabolism (Kamal et al., 2001).

2.3.3 Loss of chlorophyll and synthesis of carotenoid

Tomato fruit undergo an orderly series of physiological and morphological changes as they progress in development from mature-green (MG) to red-ripe fruit. These changes include the development of red color (i.e., due to lycopene synthesis) and loss of chlorophyll. The two most important carotenoids in fruits of tomato are lycopene, which determine 80- 90% of fruits red color, and β-carotene, which accounts for approximately 7% of the tomato carotenoids. The principal pigments that are responsible for the color of tomatoes are chlorophyll and carotenoids, especially lycopene one of many carotenoids (Arias et al., 2000). Chlorophyll is the major pigment in the early stages of tomato fruit development that imparts the green color. As fruits mature and ripen the chlorophyll content decreases because of the conversion of chloroplasts to chromoplasts and the synthesis of additional carotenoids. Color change in tomato occurs in a rather circuitous pattern (Campbell and Labavitch, 1991).

In a typical fruit, loss of chlorophyll and synthesis of lycopene begin in the locular or central columella tissues, emerge externally at the stylar end of the fruit, then spread rapidly across the superficial exocarp layer, before progressing into the underlying endocarp tissues (Grierson and Kader, 1986). Lycopene and ß-carotene, represent the primary components of ripe fruit pigmentation in tomato pericarp and are responsible for the characteristic color of ripe tomatoes, conferring deep red and orange colors, respectively. These carotenoids largely influence the quality perception of fresh tomatoes (Liu et al., 2009). The chlorophyll content is reduced by 90% by the time tomatoes are red-ripe. The intensity of the bright red color of tomatoes is mainly due to the presence of lycopene which increase as tomato mature (Shi and Maguer, 2000).

2.3.4 Fruit softening due to cell wall degradation

Postharvest decrease in fruit firmness is an important component of the increase in palatability that accompanies fruit ripening. Softening is a developmentally programmed ripening process in many fruits, providing different textures as observed in various fruits, including juiciness, crispness, and stiffness (Seymour et al., 2002). It results from cell structure deterioration and changes in composition of cellular material and cell wall (Seymour et al., 2002). It is a biochemical process involving pectin and starch hydrolysis due to enzymes including wall hydrolases. Depolymerization (shortening of chain length of pectin substances) occurs with an increase in pectinesterase and polygalacturonase activities during fruit ripening (Yaman and Bayoindirli, 2002). Softening characterized by an increase in soluble pectins. Pectin is a major component of the middle lamella, which binds adjacent cells. A textural change as a result of solubilization of pectin during ripening in tomatoes (Gross, 1990). Fruit softening is a complex process that involves three sequential steps: loosening of cell wall mediated by expansions, depolymerization of hemicelluloses, and finally polyuronided depolymerization by polygalacturonase or other hydrolytic enzymes (Brummell et al., 1999).

Loss of firmness has detrimental effect and one factor for senescence of the fruit. If softening is not effectively controlled in the postharvest environment, however, fruit susceptibility to mechanical damage and pathogen attack is greatly increased (Sommer, 1982).

Firmness may also be affected by the stage of fruit maturity. In general, climacteric fruits have increased transpiration at very early (pre-climacteric) stages (Padmini, 2006). Hence, the necessary time to achieve full ripeness for fruits at different stages of maturity is a determinant of shelf life time; thereby it can directly affect changes of the fruit quality, particularly the weight loss and firmness (Casierra and Aguilar, 2009).

Softening is a biochemical process involving the hydrolysis of pectin and starch by enzymes like cell wall hydrolases. As the process of fruit ripening progresses, depolymerisation or softening of chain length of pectin substances occurs with an increase in pectinesterase and polygalacturonase activities (Yaman and Bayoindirli, 2002). It change has been associated with the degradation of the middle lamella of cortical parenchyma cells causing a marked

increase in pectin solubilization, but only slight changes in its molecular weight and small decreases in the content of hemicelluloses. During fruit ripening, depolymerization or shortening of chain length of pectin substances occurs with an increase in pectin-esterase and polygalacturonase activities (Kashappa and Hyun, 2006). The enzymatic processes are strongly associated with respiration rate of fruits.

During the ripening process, cell wall-modifying activity of several enzymes, including polygalacturonase, pectin-methyl-esterase, endo-β-mannase, α- and β-galactosidases, and β-glucanases, causes softening of the whole fruit by altering the texture due to degradation of the structural components necessary to reinforce the cell wall and the adhesion of cells (Athmaselvi et al., 2013).

2.3.5 Changes in cellular membranes

Fruit growth and ripening are complex developmental processes that involve many events contributing to the textural and constitutional changes in fruits and determining their final composition. The metabolic changes during ripening include alteration of cell structure, changes in cell wall thickness, permeability of plasma membrane, hydration of cell wall, decrease in the structural integrity, and increase in intracellular spaces (Redgwell et al., 1997).

Cell wall disassembly rate and extent are crucial for the maintenance of fruit quality and integrity (Matas et al., 2009). Among the mechanisms associated with tomato fruit ripening, changes in membrane structure play an important role. The cell membrane system (i.e. plasma membrane, endoplasmic reticulum, vacuolar membrane etc.) acts as selectively permeable barriers to the movement of compounds within and between cells. Membrane structure consists of fluid bilayers containing phospholipids and proteins. Senescence is characterized by degradation of cell membranes and a loss of membrane integrity and function, which in turn leads to loss of tissue structure, alterations in cellular metabolism and ultimately accelerated death (Paliyath and Droillard, 1992). One significant change in the membranes occurring with senescence is the change in fluidity. With the development of senescence, fluidity decreases and membranes become more rigid.

2.3.6 Degradation of starch, and synthesis of total soluble solids

In the early stages of maturation, starch accumulated in the fruit is progressively hydrolyzed in order to increase sweetness, thus affecting fruit taste during ripening (Magein and Leurquin, 2000). Decrease in starch content accompanied by an increase in soluble solids and total sugar is typical of postharvest change in climacteric fruits (Pinto et al., 2004). The amount of soluble solid in the fruit is known to increase with maturation due to the conversion of starch to sugar. Tomato fruit accumulates carbohydrate prior to the onset of ripening in the form of starch. The fruits accumulated low levels of starch in the immature stages (Yu et al., 1967). Starch accumulation continues up to the 'mature-green' stage and then rapidly decreases as ripening begin. Starch constitutes 0.10 - 0.15% in ripe tomato fruits on a dry weight basis, and was hydrolyzed during ripening.

The breakdown of starch to sugar is associated with activities of α- and β-amylases and starch phosphorylase. In addition, soluble solids content increases with fruit maturity through biosynthesis process or degradation of polysaccharides (Salunkhe et al., 1974). The increase in total soluble solids in fruits is directly correlated to the hydrolytic activities in starch, the increased activity of enzymes responsible for the hydrolysis of starch to soluble sugars, which indicates that the fruits are at the ripening process (Hassan et al., 2014). Carbohydrates constitute about 65% of the soluble solid of ripe tomato fruit. High sugar and acid are required for best flavor. The soluble sugars glucose, fructose and sucrose are the largest contributors to the total soluble solids. Soluble solids constitute a large fraction of the total solids in tomato and are indicators of sweetness (Anthon et al., 2011).

2.4 Antioxidant capacity of tomato fruit

Antioxidants are important in disease prevention in both plants and animals, inhibiting or delaying the oxidation of biomolecules by preventing the initiation or propagation of oxidizing chain reactions (Yahia et al., 2007). Fresh fruits and vegetables are rich sources of antioxidants. As such, a high intake of fresh fruits and vegetables has been demonstrated to be

protective against both heart disease and certain types of cancer due to some important constituents present in the fruit (Giovannucci, 1999).

Tomato antioxidants include carotenoids such as β-carotene, a precursor of vitamin A, and mainly lycopene, which is largely responsible for the red color of the fruit, vitamins such as ascorbic acid, and phenolic compounds such as flavonoids and phenolic acids (Borguini and Torres, 2009). These include antioxidant activities, such as the quenching of singlet oxygen or the scavenging of peroxyl radicals, induction of cell to cell communication and growth control (Wills and Ku, 2002). These compounds may play an important role inhibiting reactive oxygen species responsible for many important diseases, through free-radical scavenging, metal chelation, inhibition of cellular proliferation, and modulation of enzymatic activity and signal transduction pathways (Crozier et al., 2009). Lipid peroxidation occurs by oxidation of fatty acids in the presence of enzymes and by exposure to reactive oxygen species and to transition metal ions in a free radical chain reaction.

Results from recent research have shown that the diverse phenolic compounds present in fruits and vegetables are responsible for the high antioxidant capacity. Phenolic acid and flavonoid compounds are secondary metabolites in plants with the ability to protect human body tissue against oxidative attacks (Romanazzi et al., 2002). Phenolic compounds, because of their structure, are very efficient scavengers of peroxyl radicals. Moreover, the action of phenolic compounds can be related to their capacity to reduce and chelate ferric iron which catalyzes lipid peroxidation (Subhash et al., 2007). The flavonoids have been confirmed as a group of polyphenols important in conferring antioxidant benefits (Luthria et al., 2006).

Tomato flavonoids, in particular rutin and naringenin due to their high antioxidant power and to the significant biological activities, can have a substantial role in the health benefits attributed to the tomato consumption (Bourne, 1998). The antioxidant and free radical-scavenging properties of polyphenol compounds in several plant extracts have been recently reported, suggesting possible protective roles of polyphenol compounds in reducing risk of cardiovascular diseases in humans. Whereas lycopene is the most efficient singlet oxygen quencher among the biological carotenoids (Khachik et al., 2002).

Although natural antioxidants appear important in disease prevention, only limited data are available on their occurrence and distribution in tomatoes. Information about changes in bioactive compounds composition and their total antioxidant capacity during storage is required to offer consumers nutritionally sound fresh fruits. Tomatoes are perceived by the general consumer as an antioxidant rich fruit that is highly perishable, having a short shelf life. This opinion is based on the observable alterations in the outer appearance of the fruit namely the rapid change in firmness, texture and color and the propensity to develop rottenness. Less obvious to the public are the changes that occur at the level of composition after the detachment of the fruit from the plant. These are undoubtedly the most relevant because they affect the nutritional value of tomatoes. Moreover, during ripening the chemical composition of the fruit also changes dramatically, affecting texture, flavour, antioxidant contents mainly phenolic compounds, flavonoids and ascorbic acid (Bailén et al., 2006). Since the taste, color and nutrient qualities of tomatoes can also depend on their antioxidant contents, further insights into the factors likely to affect their composition should help to define the quality of tomatoes more clearly (Dumas et al., 2003).

3. MATERIALS AND METHODS

3.1. Description of the study area

The study was conducted in Jimma University College of Agriculture and Veterinary Medicine (JUCAVM). JUCAVM geographically located 346 Km southwest of Addis Ababa at about 7^0, 33'N latitude and 36^0, 57' E longitude at an altitude of 1710 m.a.s.l. The mean maximum and minimum temperature of the area are 26.8^{o}C and 11.4^{o}C, respectively and the mean maximum and minimum relative humidity is 91.4% and 39.92%, respectively (BPEDORS, 2000). During the present study, the average temperature and relative humidity of the PHM laboratory was 22^0C$\pm$1 and 74$\pm$1 % RH respectively.

3.2 Experimental material

The variety used in this study was selected on the basis of perishability. Tomato fruits ((*Lycopersiconesculentum* Mill.) fresh type, of variety Barbados which was produced in green house were collected from Hawassa Jittu farm at different stages of maturity (mature green, turning and light red). This variety was selected among other varities due to its highly perishable nature unlike other fresh type tomatoes varieties produced at Jitu farm. After harvesting the fruits were transported to Addis Ababa by refrigerated cold truck which was adjusted to 15^{o}C then to JUCAVM postharvest management laboratory using a Track exposed to ambient air condition. Maximum care was taken to minimize mechanical damage during harvesting, transportation and handling of fruits.

3.3 Preparation of experimental material

Fruit maturation level was precisely selected and the fruit color was compared in the field using biological color chart (USDA, 1991). Harvesting was carried out manually in the morning. Disease-free fruits having uniform shape, size and weight without any injuries or defects were selected and hand washed with tap water, blotted with soft cloth. The three harvesting stages of tomato fruits used in this experiment included mature green (tomato surface is completely green), turning (Tannish-yellow, pink or red color shows on over 10% but not more than 30% of the tomato surface) and light red (Pinkish-red or red color shows on

over 60% but red color covers not more than 90% of the tomato surface). From each stage of maturity for each treatment 18 uniform fruits were washed with tap water containing 2% (w/v) sodium hypochlorite solution, and rinsed with sterile water, bloated with cheese cloth and surface dried at ambient condition.

Figure 3. The three harvesting stages of tomato fruits used in the Experiment

3.4 Preparation and application of edible coating materials to treatments

3.4.1 Preparation of pectin solution

Pectin solution preparation was conducted as indicated in Felix and Mahendran (2009). Briefly, commercially available pectin (30 g) Degree of esterification 50% was dissolved in 1000 ml warm water (40°C) whilst stirring with magnetic stirrer to prepare 3% (w/v) pectin solution and allowed to homogenize, with moderate stirring until the solute completely dissolved.

3.4.2 Preparation of chitosan solution

The chitosan solutions were prepared according to El Ghaouth et al. (1992). Twenty gram of chitosan was dispersed in 900 ml of distilled water and 50 ml of glacial acetic acid was added to dissolve the chitosan. The solutions were homogenized with stirring using magnetic stirrer to remove undissolved particles. In order to guarantee the stability of the emulsions, the pH

value was adjusted to 5.6 with 1N NaOH solution. Tween 80 (0.l% v/v) was added to improve wettability of the solution during coating.

3.4.3 Application of coating treatments

Fruits were uniformly dipped in each solution when the temperature of the solutions reached at room temperature (25^{o}C). Fruits were dipped for 2–3 min to ensure uniformity coating of the whole surface. Meanwhile control fruits were dipped in distilled water for the same duration and excess water/solution from the fruits were removed were air dried for 3 h at room temperature. A dry layer with plastic texture and general appearance of the fruits were used as criteria to determine the end of surface drying. Surface dried coated fruits were then packed in cardboard boxes with a size of 12 cm L x10 cm Hx 8.5cmW. Cardboards have 6 openings of 7 cm^3 size on sides (except bottom and top parts). The data were recorded before treatment (day 0) and in 5 days interval for all physicochemical parameters for 20 days.

3.6 Data collected

Data were collected for both physical and chemical parameters. First the non distractive parameters were measured then the distractive measurements were taken.

3.6.1 Physical parameters

3.6.1.1. Physiological weight loss

Weight loss of fruits was recorded from day zero of treatment through storage time under ambient storage conditions and recorded at 5 day intervals. Relative percentage weight loss was calculated using Eq. 1 and the cumulative weight loss was expressed as the cumulative percentage for the respective treatments (Athmaselvi et al., 2013).

$$\%\, WL = \frac{WI - WF}{WI} x\, 100 \qquad (1)$$

where % WL=percentage weight loss, WI=initial fruit weight in g, WF=final fruit weight in g at the indicated period.

3.6.1.2 Fruit firmness

The method given by Fan et al. (1999) was used to determine fruit firmness using Texture Analyzer (Micros Sable TA-XT plus, UK). The force required for the plunger to press into the fruit to a depth of 5 mm was recorded, and expressed in Newton. Firmness stable Microsystems with 2 mm plunger tip, with flat head stainless-steel cylindrical probe was used for the measurement of tomato fruit firmness. The machine was set for compression with a speed of 1.5 mm/sec. For the present research from each treatment two fruits were used from all treatments at a time and the average result was used for the analysis. The start of penetration test was the contact of the probe with the surface of tomato fruits and finished when the probe penetrated the tissues to a depth of 5 mm. The point where the maximum force at time of penetration was recorded as the value for the fruit firmness in Newton.

3.6.1.3 Fruit color change

Total colour change of samples were determined using CIE (Commission Internationale de L'E clairage) L*a*b* color space to evaluate the effect of coating on color change of samples using tri-stimulus colorimeter (Accu probe HH06), which was calibrated using white tiles. The instrument was standardized with standard white tile (L=83.14, a*=-3.67 and b*=10.79). Total color change were expressed in terms of "L*" value (lightness and blackness , ranging from 100 to zero respectively), "a*" (redness to greenness) value and "b*" (yellowness to blueness) value. Color measurement was made on day zero as a target value and other measurements were collected on specified day intervals. Multiple readings (3-5) per fruit were taken from each sample by changing the position of the tomato fruits to get average color measurements values (Maftoonazad and Ramaswamy, 2005).The total color change (ΔE) was calculated using Eq. 2.

$$\Delta E = \sqrt{(L^* - L)^2 + (a^* - a)^2 + (b^* - b)^2} \qquad (2)$$

where, ΔE = represents the total color change as compared to target value; L^* and L are lightness values for the target and sample respectively; a^* and a are target and sample redness values respectively; b^* and b are target and sample yellowness values respectively.

3.6.1.4 Disease incidence

Disease incidence was calculated as number of infested fruits showing any disease symptoms out of total numbers of tomato fruits stored. Five separate tomato fruits were allocated and used for the assessment of disease incidence and percent disease index was evaluated as indicated in Hossain et al. (2010).

$$Disease\ incidence\ (I)(Frequency) = \frac{number\ of\ \ fruits \inf ected * 100}{total\ number\ of\ \ fruits\ assesed} \quad (3)$$

3.6.1.5 Percent disease index (PDI)

For estimation of fruit area affected by disease, the whole fruit area was considered as 100 and thereby the infected area was visually estimated in order to determine the Percent of Disease Index (PDI), i.e. severity was determined as per the scale of Amadi et al. (2009). A quantitative severity index (0 – 4 rating) was used, in which the numbers 0, 1, 2, 3, and 4, indicated that no infected surface area scored 0, whereas the infected surface areas 1-25%, 26- 50%, 51-75%, 76% or more of the fruit surface areas affected by the disease, respectively.

$$percent\ \ disease\ \ index\ (Area) = \frac{sum\ of\ \ all\ disease\ \ ratings * 100}{total\ number\ of\ \ fruits * \max imum\ \ rating\ value} \quad (4)$$

3.6.1.6 Determination of shelf life of stored fruits

The shelf life of tomato fruits was calculated by counting the days required for them to attain the last stage of ripening, but up to the stage when they remained still acceptable for commercial marketing. About 10% physiological loss in weight was considered as an index of termination of the shelf life (threshold level) of fruit commodities (Pal et al., 1997; Acedo, 1997).

3.6.2 Chemical parameters

3.6.2.1 Determination of pH

The pH of the sample tomato fruits was determined following the method described in Rangana (1979). Tomatoes were crushed and made into juice. Then the pH meter was standardized with pH 4.0 and 7.0 buffer solutions. After standardization, the pH of each juice sample was measured by using digital pH meter (CP-505, Poland).

3.6.2.2 Determination of Titratable Acidity (TA)

Titratable Acidity (TA) was determined by titration (AOAC, 2000). Fruits were crushed and made into juice, and 5 ml of sample from the pulp was taken and added in to 250 ml conical flask. Then 10 ml distilled water was added to make the fruit color light to facilitate clear end point identification. To determine the total titratable acidity of the pulp, fresh 0.1N NaOH was used. TA of tomato fruits was expressed as percentage of citric acid equivalent, since this organic acid is a predominant acid in the fruit. Titrable acidity was determined using Eq. 5.

$$\%\ acid = \frac{(ml\ NaOH)(N\ of\ the\ base\ in\ mol\ per\ liter)(0.0064)}{Sample\ volume\ in\ ml} \text{ X } 100 \qquad (5)$$

Where 1ml 0.1N NaOH is equivalent to 0.0064g citric acid.

3.6.2.3 Determination of total soluble solid

The total soluble solid (TSS) content of tomato fruit pulp was determined using a digital hand held refractometer (Bellingham + Stanley 45-2, UK). The percentage of TSS was obtained from direct reading of the refractometer in degree brix . First the refractometer was calibrated with distilled water, and TSS of juice was determined. Before measurement, homogenous sample was prepared by blending the tomato flesh in a blender. The sample was thoroughly mixed and drops of juice were placed on the prism surface then prism lid was closed. The position at which the demarcation line between the light and dark regions crosses the vertical scale gives the percentage of soluble solids readings. Multiple measurements (3-5)

were taken per a replicated treatment and the average values were used. After each test, the prism plate was cleaned with distilled water and wiped with a soft tissue for subsequent measurement.

3.6.2.4 Determination of TSS/TA ratio (TSS: TA)

The ratio between TSS and TA was determined by dividing the value of TSS to that of TA in order to have a sugar-acid balance of samples for each treatment. In order to calculate the Sugar to acid ratio, Eq.6 was used:

$$sugar\ \ acid\ ratio = \frac{^{o}Brix\,value}{percentage\ acid} \tag{6}$$

3.6.2.5 Determination of ascorbic acid content

Ascorbic acid content was determined by spectrophotometric method (Mohammed et al., 2009). Five grams of tomato sample was mixed with 100 ml of 6% trichloro acetic acid and quantitatively transferred into a 200 mL volumetric flask and shaken gently to homogenize the solution. The obtained solution was filtered and centrifuged at 4000 rpm for 15 minutes, then the sample transferred to a conical flask and 1-2 drops of saturated Bromine solution were added and aerated. Then to each 10 ml aliquot 10 ml of 2% thiourea was added. By using pipette from 10 ml aliquot 4 ml added into each of test tubes. Then 1 ml of 2, 4- DNPH solution was added to form

(7)

Where: A_s *Absorbance of samples*
A_b *Absorbance of blank*
$A_{10\ \mu g\ Std}$. *Absorbance of 10 µg AA standard*

3.6.2.6 Estimation of lycopene content

The Lycopene content of the sample tomato fruits was analyzed according to the method described by Nagata and Yamashita (1992). First tomato fruits were crushed and well homogenized, seeds were separated and then one gram of the sample (tomato pulp) was taken.

All pigments in the sample were extracted with acetone and hexane (ratio of 4:6). After homogenizing extracted samples, samples were placed into a beaker and allowed to stand for about 15 minutes till it made a phase separation. Finally the pigment from top part was collected into a glass curvet (1cm path length) and their absorbance was measured at 663nm, 645nm, 505nm and 453nm using spectrophotometer (T80 UV/VIS, UK). The absorbance readings taken at different wave lengths were used to determine the total lycopene content using Eq.8 as indicated in Nagata and Yamashita (1992):

$$Lycopene\ \left(mg/100g\right) = -0.0458A_{663} + 0.204A_{645} + 0.372A_{505} - 0.0806A_{453} \quad (8)$$

3.6.2.7 Determination of total polyphenol content

Total phenols were measured spectrophotometrically using Folin–Ciocalteu reagent with gallic acid as a standard (Gao et al., 2011). Briefly, 50 µl of tomato extract were added to 3 ml of deionized water plus 250 µl of Folin–Ciocalteu reagent (1N). After a 5 min reaction time, 750 µl of 20 % Na_2CO_3 solution was added. The mixture volume was made up to 5 ml with deionized water. Then the total phenolic content was measured at 760 nm after a 30 min reaction time using spectrophotometer (T80 UV/VIS, UK). The results are reported in terms of mg of gallic acid equivalent (GAE) per 100 g of fresh weight. Pure Gallic acid (GA) was used as a standard (covering the concentration range between 0.1 and 1.0 mg/mL) (R^2 = 0.993) and results were expressed as milligrams of GAE per gram of fresh weight.

3.5 Design of the experiment and Data analysis

In this study, all the experiments were laid in a Completely Randomized Design (CRD) with a factorial treatment combination, replicated three times, whereby 18 tomato fruits were used per replication. Analysis of variance (ANOVA) was performed using SAS statistical program (Version 9.2). First the data were checked whether they were fulfilled the assumption or not. The Least Significant Differences (LSD) test will be performed following the ANOVA for Treatments showing statistically significant difference at $P < 0.05$ level. Data for disease

incidence and severity were analyzed using non parametric test. As those disease data did not fulfilled the ANOVA assumption

4. RESULTS AND DISCUSSION

4.1 Physical characteristics

4.1.1 Physiological weight loss

Moisture loss occurred due to vapour phase diffusion driven by a gradient of water vapour pressure between inside and outside of fruit (Nisperos- Carriedo *et al*., 1992). Water loss from fruits equates to loss of saleable weights thus it cause direct loss in marketing.

Weight loss is an important index of postharvest storage life in fresh produces. It is mainly attributed to the loss of water during metabolic processes like respiration and transpiration. Physiological weight loss appeared to be the major detrimental factor of storage life and quality of tomato fruits in particular and horticultural crops in general. In the current study the interaction between maturity stages and types of coating materials used resulted in a significant ($P< 0.05$) difference in physiological weight loss of tomato fruits (Appendix Table 1). Weight loss of coated tomato fruits was relatively lower than the uncoated fruits. However, pectin and chitosan treated samples showed less weight loss as compared to the control (Table 1). This shows that coating fruits with edible coating materials like with chitosan and pectin reduces weight loss of tomato fruits through reducing rate of transpiration weight losses from fruits. When harvesting stage is taken into consideration with types of coating materials, the highest weight loss (18.7%) was observed from fruits harvested at light red stage (maximum maturity stage in this study) with no coating, but the lowest (8.5%) was for fruits harvested at turning stage coated with pectin. It was found that as the storage duration extended the weight loss percentage was also increased and the maximum weight loss was recorded on 15^{th} day of storage from control fruits.

Table 1**:** Effects of pectin and chitosan coatings on Physiological weight loss (%) of tomato fruits harvested at different stages of maturity

Coating materials	Harvesting Stages	Days after application of coating materials			
		Day 5	Day10	Day 15	Day 20
Control	Mature green	3.4^{cd}	10.0^{c}	14.0^{c}	

	Turning	3.8[c]	11.1[b]	15.8[b]	
	Light Red	5.3[a]	13.3[a]	18.7[a]	
Chitosan	Mature green	3.0[e]	6.97[f]	10.2[de]	14.5[c]
	Turning	3.4[d]	8.1[e]	9.5[e]	13.5[d]
	Light Red	4.3[b]	9.1[d]	11.0[d]	17.4[a]
Pectin	Mature green	2.9[e]	6.7[f]	9.5[e]	14.7[c]
	Turning	3.7[cd]	8.1[e]	8.5[f]	13.0[d]
	Light Red	4.3[b]	9.0[d]	10.9[d]	15.8[b]
LSD (5%)		0.4	0.6	0.8	0.6
CV (%)		6.4	4.2	4.3	2.5

**After 15 day of storage all uncoated control fruits were spoiled and discarded.*

Note: *Means with the same letter (s) within a column are not significantly different.*

Moisture loss and gaseous exchange from fruits is usually controlled by the epidermal layers provided with guard cells and stomata. Coating helps to reduce this further because it forms a film on the top of the fruit skin and act as an additional barrier to moisture loss (Togrul and Arslan (2004) which in turn reduce rate of moisture migration from fruits. This barrier property also reduces the oxygen uptake by the fruit which in turn slows down rate of respiration and associated weight loss from the fruit surface (Abbasi et al., 2009). Water losses from transpiration may also be affected by the stage of fruit maturity. This result is in line with Getinet et al. (2008) who found the highest weight loss was recorded in Marglobe tomato fruits harvested at light-red stage and the lowest weight loss was from Roma VF variety harvested at mature-green stage.

When stage of harvesting is considered alone, the highest weight loss was recorded on tomato fruit harvested at light red stage and the lowest value for fruits harvested at green stage. This might be associated with thin epidermal layer of more matured fruit as compared to less degraded the skin of relatively less matured fruits.

In general, the observed weight loss in control (uncoated) fruits might be associated with effects of transpiration associated with high water vapor pressure difference between the fruit surface and ambient air due to absence of protective layer that slows down the rate of

migration of water molecules. On the other hand a low water vapor pressure difference was created between surfaces of coated fruits as compared to control. Therefore, for uncoated fruits, weight loss can lead to wilting and shriveling resulted in shorter storage life and poor fruits for market and consumption. Since tomato fruit is rich in moisture content, a 10% moisture loss can be translated as 10% loss in market value. However, using edible coatings would be advantageous because they are not only act as barriers, reducing water transfer (Baldwin et al., 1999), but also can seal small wounds on skins and thus inhibiting mold infections (Mario et al., 2014).

Differences pertaining to physiological weight loss between coated and uncoated tomatoes were also reported by Lin and Zhao (2007) who were observed that edible coatings provide an effective barrier to water vapor transmission thus helping to alleviate the problem of moisture loss. This result is also in agreement with the findings of Srinivasa et al. (2006) for tomato and bell pepper packaged in cartons covered with chitosan film which extended the storage life of both tomato and bell pepper through reduction of water loss and modification of the internal atmosphere. Ali et al. (2010), who reported that gum Arabic coating on tomato fruits extended storage life through reduction of water loss and modification of the internal atmosphere using coating materials. Weight loss was also noticed by Athmaselvi et al. (2013) and it was lower in tomato fruits coated with Aloe vera and higher in fruit without any treatment. Salunke et al. (1991) also indicated that slower rates of moisture loss in coated fruits can be attributed to the barrier properties for gas diffusion through cuticle and lenticels, the organelles that regulate the transpiration process and gas exchange between the fruit and its environment.

Shriveling symptoms (Wills et al., 1981) of tomato fruits may become evident when the weight loss becomes extreme as it was observed on uncoated fruits of the present study. Shriveling was caused due to water loss by respiratory and water transpiration (Woods, 1990). Tomatoes are very susceptible to moisture loss because they have very thin skin, which offers little resistance to mass transfer of water and causing shrinkage which affects its sensory and marketable values. In the current study, no severe symptoms of shriveling were observed on coated fruits until the 15^{th} day of storage for all harvesting stages, which is so important from

quantitative and qualitative point of views. However, after the 15th day of storage the green and light red fruits showed some shriveling symptom while those fruits at turning stage remained acceptable until day 20. The light red uncoated fruits showed significant shriveling after 10 days of storage time. On the 15th day of storage fruits of all harvesting stage showed significant shriveling symptom. As a result, all the control fruits turned unattractive, which was associated with the high moisture loss of water from the fruits.

Furthermore, coating imparted an attractive glossy shine appearance on fruits, and coated fruits maintained wholesomeness appearance even after 15 days of storage which have a market value advantage. This might be attributed to the fact that edible coatings decrease the water vapor transmission rate by forming a barrier which prevents skin texture change, as water is essential for the preservation of cell turgor (Perez-Gago et al., 2010).

4.1.2 Fruit firmness

Fruit firmness is a major attribute that dictates the postharvest life and quality of fruits. It is an important factor indicating the internal freshness of tomatoes and influences acceptability of the fruit by consumers. It is related to water content and cell wall strength of fruit skins which is mainly affected by metabolic changes during storage.

Data in Table 2 shows that a significant ($P < 0.01$) difference was observed in tomato fruit firmness due harvest stages, coating materials and their interaction. Firmness of tomato fruits was better preserved by the application of coatings (Table 2.). At the beginning of storage period (before coating) the values recorded for tomato fruit fresh firmness were 9.01N, 7.47 N and 6.3N for mature green, turning and light red fruits respectively. These values decreased sharply in control fruits until day 15 of storage (Table 2). At the end of the storage (15th day) period, control fruits clearly showed the lowest firmness. The adequate fruit firmness was maintained by pectin and chitosan coatings till 15 days after coating application. Present study showed that tomatoes at green stages are firmer than tomatoes at turning and light red stages during the storage period.

Table 2 Firmness (N) of coated and uncoated tomato fruits during storage time at ambient condition

Coating materials	Harvesting Stages	Days after application of coating materials			
		Day 5	Day10	Day 15	Day 20
Control	Mature green	6.64^{cd}	4.87^{e}	3.63^{d}	
	Turning	4.85^{f}	4.02^{f}	3.06^{e}	
	Light Red	4.02^{g}	3.04^{g}	2.21^{f}	
Chitosan	Mature green	7.8^{b}	7.02^{a}	4.61^{c}	4.49^{b}
	Turning	8.0^{d}	5.6^{c}	5.2^{b}	4.94^{ab}
	Light Red	5.39^{e}	5.0^{de}	3.57^{d}	3.4^{d}
Pectin	Mature green	8.63^{a}	7.2^{a}	5.87^{a}	4.68^{b}
	Turning	6.9^{c}	6.05^{b}	5.64^{a}	5.2^{a}
	Light Red	5.79^{e}	5.22^{cd}	3.86^{d}	3.92^{c}
LSD		0.4	0.4	0.35	0.2
CV		3.8	2.9	5	5.7

**After 15 day of storage all control fruits were spoiled and discarded.*

Note: *Means with the same letter (s) within a column are not significantly different.*

This is obvious that stage of harvesting at maturity has direct effect on retaining firmness of tomato fruits. Firmness decreased notably with advance in maturity stage of tomato fruits. Maftoonazad and others (2008) also indicated that as the length of storage period extended, peach fruits showed a significant decrease in firmness while loss of texture and softening were delayed in coated fruits. In similar study Tilahun (2013) showed that the highest value of firmness was recorded in mature green tomato while the lowest value in full ripen stage.

Better firmness retention was observed from coated fruits at different harvesting stages. The rate of firmness degradation was high in case of uncoated fruits. Preservation of firmness associated with rate of respiration and cell wall degradation of tomato fruits. The lesser the rate to better fruit firmness retained. This might be associated that, coating of fruits can be expected to modify the internal gas composition of fruits, especially reducing diffusion and availability of O_2 from ambient air to cells respiration (Salunkhe et al., 1991). Limited O_2 availability limits the rate of respiration of cells of coated fruits which in turn better cell wall retention. Furthermore, the decrease in water loss due to fruit coating might have resulted in maintenance of fruit firmness.

In terms of harvesting stage, fruits harvested at different stages of maturity showed significant difference in firmness and decline with an increase in stage of harvesting from green mature to light red stage. However, results of this study showed that, coating works better with stages of harvesting as compared to uncoated fruits. Maftoonazad and Ramaswamy (2005) reported that firmness value in coated samples was almost 1.5 times higher than that of uncoated fruits, as reported for avocados coated with methylcellulose. Similarly, Chauhan et al. (2013) indicated that Shellac based surface coating retained tomatoes' firmness better than control fruits. Generally, the combined treatment effect of coating and early harvesting stage showed a significant beneficial effect on firmness retention as compared to uncoated fruits. Even though coating materials and stages of maturity showed significant interaction effects, but relatively better fruit firmness was observed when pectin coating combined with green mature harvesting stage (after 5 and 10 days storage) and turning stage (after 20 days storage). This might be storage stability of pectin coating on fruits surface as compared to chitosan film.

4.1.3. Color change

Color is a very important determinant of quality and consumer acceptability. It is most important characteristic to assess ripeness and postharvest life of tomato and has major importance in making purchase decision. It is apparent that tomato fruits harvested at different stages exhibit color difference. However, original fruits color immediately after coating were taken as a bench mark to evaluate color changes of fruits with time for each treatment combination. Compared to initial color of fruits, coated fruits showed significant delay on change of color as compared to uncoated ones (Appendix Table 2). But no significant ($P>0.05$) interaction effects was observed between coating and stage of harvest. Most often, fresh tomato fruits are consumed at their maximum organoleptic quality which is attained when the fruit reaches at full red stage before excessive softening. This stage is attained after complex metabolic processes. However, color change of tomato fruit is associated with chlorophyll degradation as well as lycopene synthesis and accumulation (Dumas et al., 2003) which are influenced by rate of respiration. During the course of ripening, chloroplasts in the peel are transformed into chromoplasts containing red and yellow pigments (Lizada, 1993).

The total color difference (ΔE) extensively used to characterize variations in color perception due chlorophyll degradation and formation of lycopene. Figure 4, a and b summarize progressive change of total color with time from initial values as affected by types of coating materials and stage at harvesting.

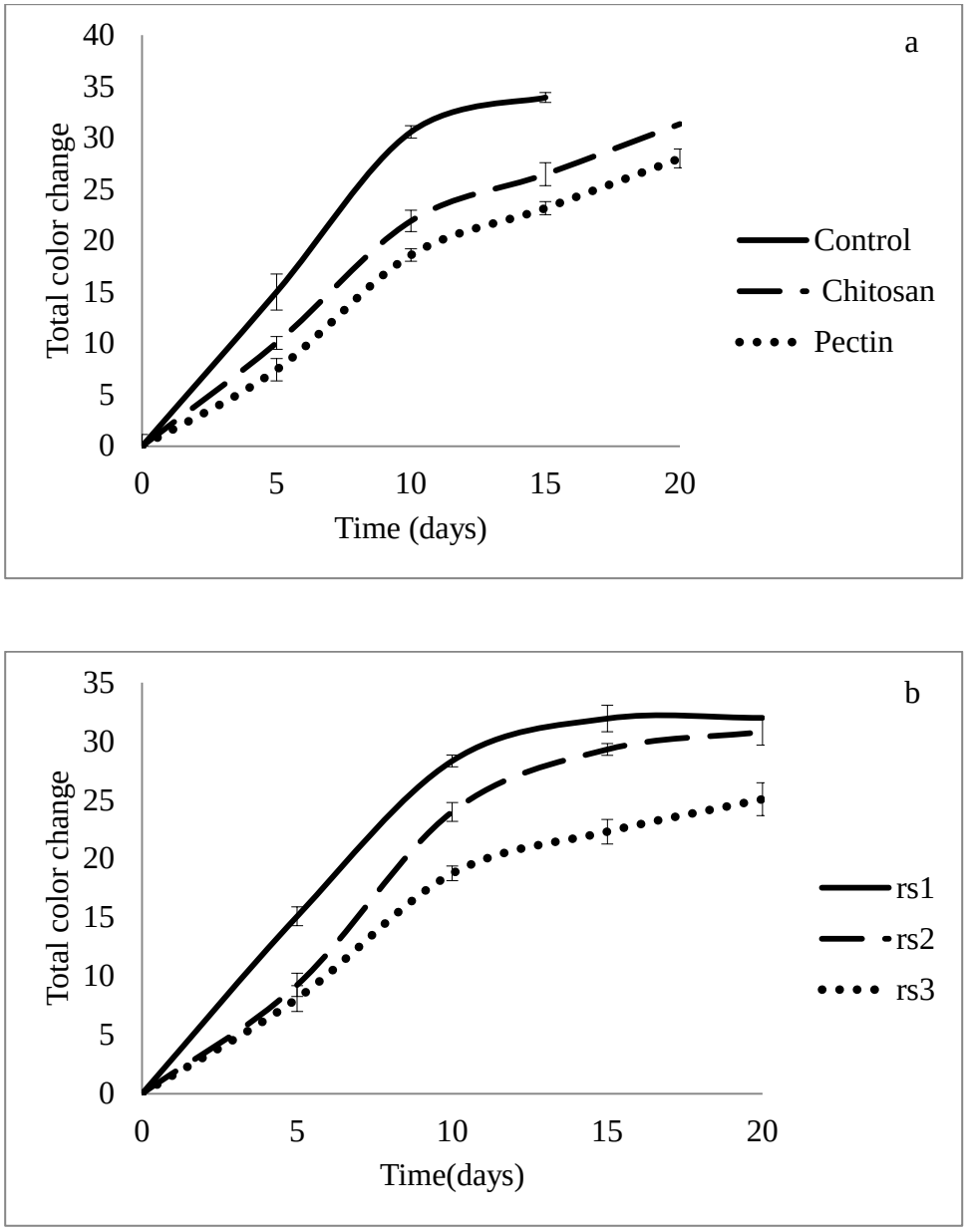

Figure 4 The effects of fruit coating materials (a) and maturity stages at harvest (b) on total color change of tomato fruits stored under ambient condition.

The total color change in control tomato fruits was enhanced and they become red within 2-5 days of storage compared to those fruits which were coated with chitosan and pectin (5-12 days) depending on the harvesting stage. Generally uncoated fruits harvested at different maturity stages changed their color rapidly during storage. The highest rate of color change was observed in tomato fruits harvested at green mature stage followed by turning and red stage. This was mainly because of color difference from turning and light red stages were small as compared to green color from green mature stages. Total color change increased sharply in control fruits until day 10 of storage and the rise became slower after towards. Similar results were also indicated in Ali et al. (2011), a retardation of color development in papaya fruits which were treated with higher concentrations of chitosan due to slow rate of respiration and reduced ethylene production. This, in turn, delayed the ripening and senescence of the fruits, resulting in reduced color change.

Elevated CO_2 levels (>1%) in fruit tissues (which could be achieved by coating material) have been shown to retard fruit ripening by inhibiting ethylene synthesis (Martínez-Romero et al., 2006; Zapata et al., 2008) which is a key plant hormone for degradation of chlorophyll and development of carotenoids . It was also observed differently that color changes in pears were retarded by O_2 depression rather than increases in CO_2 (Amarante et al., 2001). A study indicated that, delayed synthesis of anthocyanins has been reported in papaya, as well as strawberry, litchi, sweet potatoes, bell pepper, pear and mango associated with coating with chitosan (Chien et al., 2007).

Moalemiyan et al. (2012) also reported that the color changes in control mango sharply changed from green to yellow in the very early days of storage but in pectin based coated fruits showed retardation in color development, which is in agreement with this study result as indicated in figure 3a.. In others study, Maftoonazad and Ramaswamy (2005) showed the effect of methyl cellulose-based coating on the color of avocados stored at room temperatures. Their results revealed that coated fruits had more green color than control. In general, this study also confirmed that, pectin and chitosan coatings were effective in preserving green color and also the color of coated tomato fruits showed much glossier and brighter than the control fruits, and thus imparted an attractive and natural-looking fruits.

4.1.4 Disease incidence (%)

The results in Table 3 indicate that percent incidence of diseases was significantly ($P<0.05$) affected by the interaction effect of coating and harvest stages. The incidence was significantly lower on coated tomato fruits as compared with uncoated ones. On the control fruits harvested at light red stage the first disease occurrence was observed on the 5th day of storage which was 6.7%. As the fruit become ripen they are more susceptible to fungal contamination. On the control fruits harvested at light red stage 100% incidence was observed after 15 days of storage while coated fruits with chitosan and pectin harvested at the same stage of maturity exhibited 40% and 33.3% incidences respectively. On the other hand, after 15 days of storage at ambient conditions the disease incidence on mature green fruits of control, chitosan and pectin coated fruits were 53.33%, 26.6% and 6.6 %, respectively.

Table 3: Effect of coating materials on disease incidence of tomato fruits harvested at different maturity stages during storage

Coating materials	Harvesting stages	Days after application of coating materials			
		Day 5	Day10	Day 15	Day 20
Control	Mature green	0.0^{b}	6.7^{c}	53. 3^{c}	
	Turning	0.0^{b}	33.3^{b}	80.0^{b}	
	Light Red	6.7^{a}	53.3^{a}	100^{a}	
Chitosan	Mature green	0.0^{b}	0.0^{c}	26. 7de	33. 7^{bc}
	Turning	0.0^{b}	0.0^{c}	33. 3de	40.0^{ab}
	Light Red	0.0^{b}	13.3^{c}	40. 0^{cd}	53. 3^{a}
Pectin	Mature green	0.0^{b}	0.0^{c}	6.7^{f}	11.4^{d}
	Turning	0.0^{b}	0.0^{c}	20. 0^{ef}	31.4^{cd}
	Light Red	0.0^{b}	6.7^{c}	33. 3de	40.0^{ab}

**After 15 day of storage all control fruits were spoiled and discarded.*

Note: *Means with the same letter (s) within a column are not significantly different.*

Abbasi et al. (2009) also observed that the decay control of irradiated chitosan coated mango was better as compared to uncoated fruits. The fruit-spoiling fungi were observed in untreated control fruits after 2 weeks and in irradiated chitosan coated fruits after 5 weeks of storage (Abbasi et al., 2009).

El-Ghaouth et al. (1991) suggested that chitosan induces chitinase, a defense enzyme, which catalyzes hydrolysis of chitin, a common component of fungal cell walls, thus preventing the growth of fungi on the fruit. Similarly, Zhang et al. (2011) stated that Chitosan could effectively inhibit postharvest diseases of fruits by direct inhibition of spore germination, germ tube elongation and mycelial growth of phytopathogens as well indirect inducement of defense-related enzymes. Antimicrobial capacity of edible coating materials also reported for gum Arabic. Fruits treated with 10% gum arabic coating remained disease free even after 20 days of storage. Many of the control fruits (67%) were spoiled after 16 days of storage (Ali et al., 2010).

Other works also indicated that antimicrobial properties of pectin as a coating materials. Moalemiyan et al. (2012) reported that on control fruits, on the 5^{th} day of storage 50% of the fruits had anthracnose symptoms and at the end of the experiment, 90% of the control fruits showed symptoms of anthracnose in comparison to, 3% of pectin coated fruits .Though both Chitosan and pectin do have some sort of antimicrobial property, results in this study showed that, changes in disease incidence occurred more slowly in Chitosan and pectin coated fruits as compared to the higher rates observed in the control samples (Table 3).

As indicated in the above sections, application of coating delayed the rate of firmness lose due to preserving cell wall integrity. Furthermore coating can reduce rate of respiration and ethylene synthesis. These conditions in turn retain the tolerance of fruit tissues for diseases and hence this inhibitory action can provide better protection against postharvest decay in fruits (Hassan et al., 2014). Furthermore, coating helps to delay senescence, which makes the commodity more vulnerable to pathogenic infection as a result of loss of cellular or tissue integrity (Tanada-Palmu and Grosso, 2005).

4.1.5 Percent disease index (PDI)

Percent disease index was used to assess the effectiveness of coating materials in retarding severity of fruit disease. Coated fruits were better maintained and had low severity of disease

symptoms, whereas non-treated fruits showed increased fruit deterioration (Table 4). In this study significant ($P < 0.05$) differences were observed in terms of percent of disease index of fruits due to the interaction effect of coatings and maturity stages.

After 15 days of storage the highest fruit disease incidence (83.3 %) was recorded from the uncoated light red tomato fruits while the lowest value (6.7%) was recorded from tomato fruits harvested at green stage and coated with pectin after 20 days of storage. No disease signs were observed until the 5th day of storage period for fruits of all harvesting stages. The disease symptoms appeared on the control fruits after 5-10 days depending on the harvesting stages and after 15 days most of the fruits were spoiled due to severe disease infection. As could be observed from Table 4, there was a steady increase in fruit disease severity with prolonged storage period for all treatments. The highest fruit severity was recorded after 15 days of storage on uncoated light red fruits (83.3%), followed by uncoated turning (61.6%) and green mature fruits (40%). On 15th days of storage, fruits coated with chitosan were showed 6.7%, 11.7%, 23.3% and with pectin 1.7%, 5%, 18.3% with an increase in harvesting stages.

Table 4: Effect of coating material during storage on percent of disease index on tomato fruit harvested at different stages of maturity.

Coating materials	Harvesting stages	Days after application of coating materials			
		Day5	Day10	Day 15	Day 20
Control	Mature green	0.0^{b}	1.7^{c}	40.0^{c}	
	Turning	0.0^{b}	13.3^{b}	61.6^{b}	
	Light Red	5.0^{a}	28.3^{a}	83.3^{a}	
Chitosan	Mature green	0.0^{b}	0.0^{c}	6.7^{f}	21.3^{c}
	Turning	0.0^{b}	0.0^{c}	11.7^{ef}	26.7^{b}
	Light Red	0.0^{b}	3.3^{c}	23.3^{d}	33.3^{a}
Pectin	Mature green	0.0^{b}	0.0^{c}	1.7^{g}	12.9^{d}
	Turning	0.0^{b}	0.0^{c}	5.0^{g}	10. 0^{c}
	Light Red	0.0^{b}	1.6^{c}	18.3^{de}	25.0^{b}

**After 15 day of storage all control fruits were spoiled and discarded.*
Note: *Means with the same letter (s) within a column are not significantly different.*

Maftoonazad et al. (2007) also studied the effect of pectin-based coating on the physical and physiological changes in avocados as influenced by *Lasiodiplodia theobromae* infection. Their results showed that pectin-based coating can reduce the rate of disease progress than the control ones. Study of El-Anany et al. (2009) noted that coated fruit with jojoba wax, paraffin oil, soybean oil gum, glycerol and Arabic gum on Anna apple can reduce the disease progress as compared to the control. It is also possible that coating can form a physical barrier against new pathogenic infections, reducing the incidence of postharvest diseases (Amarante et al., 2001).

Genanew (2013) stated that delaying the harvest may lead to higher tendency of increasing the disease progress which in turn results in poor quality and low market value. Since those fruits harvested in the green and turning stage were firmer than those in the light red stage, they were less susceptible to decay. Based upon these supportive evidences, from the present study it can be concluded that edible coatings being a good emulsifier protecting the rate of disease incidences. This might be associated with mechanical barrier of the film for pathogen invasion and created modified atmosphere, delayed the ripening process and maintained tissue firmness.

4.1.6 Shelf life

The time period, whereby a product is not only safe to eat, but still has acceptable taste, texture and appearance after being removed from its natural environment, is defined as shelf life (Embuscado and Huber, 2009). The shelf life of tomato fruits was considerably influenced by the coating and harvesting stages. About 10% loss in weight is considered a reference index for termination of the shelf life (threshold level) of fruit commodities (Acedo, 1997; Pal et al., 1997). As shown in Figure 5, maximum shelf life was observed for tomatoes harvested at turning stage coated by pectin (17days) followed by chitosan at the same harvesting stage (16 days). However, tomatoes harvested at light red stage coated with pectin had a maximum marketable storage life 13 days followed by chitosan having 12 days. However, minimum shelf life was recorded for control fruits harvested at light red stage (10 days).

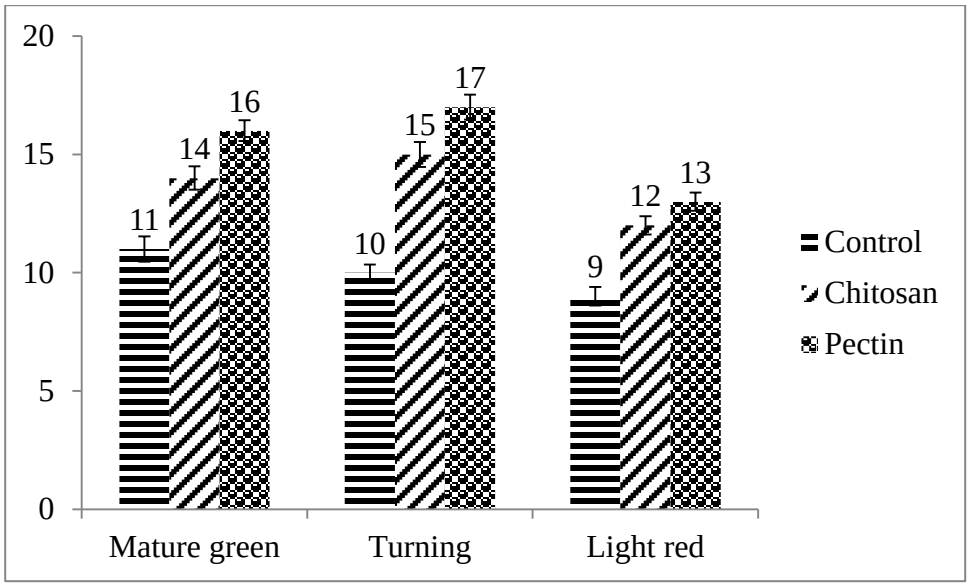

Figure 5: Shelf life (days) of tomato fruits harvested at different maturity stages and treated with pectin and chitosan and stored at ambient condition

Diaz-Sobac et al., (1996) used a coating emulsion including maltodextrins, carboxymethyl cellulose, propylene glycol and a mixture of sorbitan fatty acid esters on Manila mango. The emulsions were used to form films, which were employed to coat mangoes, to study their post-harvest life under different storage conditions (15 and 25^{o}C and 80–85% RH). Their results showed that this treatment can extend the post-harvest storage ability of Manila mangoes at least 20 days more than uncoated fruits, without the need of refrigerated storage.

Maftoonazad and Ramaswamy (2008) also used a pectin-based composite coating on avocados and evaluated the extent of quality changes under different storage temperatures for predicting the quality loss. Their results showed that pectin-based composite coatings significantly reduced the rate of physical, chemical and physiological changes in avocados during storage and extended the shelf life by more than a month at 10^{o}C storage. In addition to these reports, Moalemiyan et al., (2012) recommended that, chlorophyll retention and extension of shelf life of mangoes can be achieved by pectin coatings. Felix and Mahendran (2009) in their study showed also that coated red tomatoes took 15 days to ripe at 30°C whereas the uncoated fruits took 5 days to ripe.

4.2 Chemical parameters

4.2.1 pH

The pH increased in the storage time for both coated and uncoated fruits. The pH of tomatoes is determined primarily by the acid content of the fruit that determine the product safety and taste. In general, with an increase on days of storage and harvesting stages regardless of coating materials, pH of samples was showed an increase in value. In line with this result, Borji et al. (2012) and Moneruzzaman et al. (2009) also indicated that the pH of tomato fruit increased with advancement in maturity stage from mature- green to full-ripe stage. However, the rate of increase of pH in control fruits was higher than coated fruits (Table 5). Significant ($P < 0.05$) difference in pH value of tomato fruit was observed due to the interaction effect of maturity stages and coating (Appendix Table 6). The lowest pH values after 15 days of storage were observed for fruit samples coated with pectin at different stages of harvesting, whereas the highest for uncoated ones (Table 5).

Table 5: Effect of coating with pectin and chitosan and stage of maturity at harvest on pH of tomato fruit pulp during storage under ambient condition

Coating materials	Harvesting stages	Days after application of coating materials			
		Day 5	Day10	Day 15	Day 20
Control	Mature green	4.10^{e}	4.38^{d}	4.53^{d}	
	Turning	4.27^{c}	4.49^{bc}	4.76^{b}	
	Light Red	4.40^{a}	4.63^{a}	4.85^{a}	
Chitosan	Mature green	4.07^{ef}	4.24^{f}	4.38^{f}	4.41^{d}
	Turning	4.19^{d}	4.38^{d}	4.45^{e}	4.64^{b}
	Light Red	4.36^{ab}	4.52^{b}	4.6^{c}	4.74^{a}
Pectin	Mature green	4.03^{f}	4.26^{f}	4.29^{g}	4.38^{d}
	Turning	4.16^{d}	4.33^{e}	4.39^{ef}	4.5^{c}
	Light Red	4.33^{b}	4.49^{c}	4.53^{d}	4.64^{b}
LSD (5%)		0.04	0.02	0.06	0.11
CV (%)		0.6	0.3	0.7	0.5

After 15 day of storage all control fruits were spoiled and discarded.
Note: *Means with the same letter (s) within a column are not significantly different.*

The pH increase during storage might have been resulted from a decrease in TA content in fruits and while the higher levels of TA in coated fruits may have been due to protective O_2 barrier or reduction of O_2 supply to the internal fruit surface inhibiting respiration rate (Jiang and Li, 2001). But an increase in pH value may be due to break-down of organic acids due to respiration process during storage. Generally, a decline in acidity demonstrates advancement of maturation and ripening; thus, edible coating contributes to delaying the fruit maturation and ripening through reduction of respiration rate and lower utilization of organic acids stored in the vacuoles as respiratory substrate (Medlicott et al., 1987). Athmaselvi et al. (2013) also stated that, Aloe vera treated tomato fruits were better in keeping pH and showed a better effect in comparison with untreated fruit. Maftoonazad and Ramaswamy (2005) in their study elucidated that, pH of peache fruits increased at a higher rate in control samples as compared to coated fruits.

4.2.2 Titrable acidity (TA)

The acidity of tomato plays the major role and imparts taste to the fruit. Titratable acidity (TA) is an important consumer variable as the balance of TSS and TA relates to overall taste and consumer acceptability. TA is directly related to the concentration of organic acids present in fruits. The predominant acids in ripened tomato fruit is citric acid. The TA values of coated and uncoated fruits decreased with storage time (Table 6) and the value was significantly higher ($P \leq 0.05$) in chitosan and pectin treated fruits compared to the control due to the interaction effect of maturity stages and coating materials (Appendix Table 4).

The quantity of TA of fruits at the time of storage varied for both harvesting stages and coatings. As shown in Table 6, acidity decreased for both coated and uncoated fruits with an increase in fruit maturity and storage time. On the 10^{th} and 15^{th} days of storage the value of TA was significantly higher ($P \leq 0.05$) in chitosan and pectin treated fruits as compared to the control. Getinet et al. (2008) indicated that higher value in TA (0.67%) in fruits harvested at turning stage and the lowest value (0.58%) was from fruits harvested at mature green stage.
In coated fruits harvested at turning and mature green stage TA increased and peaked after 5 days of storage and showed a decline in concentration. On 15^{th} date of storage, the highest TA

values were observed fruit samples harvested at turning stage but coated with Chitosan and pectin. These values almost double of that of uncoated fruits harvested at the same maturity stage. This confirms that edible coating materials reduce the rate of acid metabolism (Yaman and Bayoindirli, 2002) as compared to control. Since organic acids, such as malic or citric acid, are primary substrates for respiration, a reduction in acidity is expected in terms of rate of increase in respiration of cells of fruits (El-Anany et al., 2009).

Table 6: Effect of coating with pectin and chitosan and stage of maturity at harvest on titrable acidity (%) of tomato fruits

Coating materials	Harvesting stages	Days after application of coating materials			
		Day 5	Day10	Day 15	Day 20
Control	Mature green	0.36^{d}	0.28^{c}	0.19^{c}	
	Turning	0.42^{b}	0.24^{d}	0.14^{d}	
	Light Red	0.23^{f}	0.14^{f}	0.087^{e}	
Chitosan	Mature green	0.39^{c}	0.36^{b}	0.28^{b}	0.24^{b}
	Turning	0.45^{a}	0.34^{b}	0.33^{a}	0.19^{c}
	Light Red	0.29^{e}	0.21^{e}	0.18^{c}	0.16^{d}
Pectin	Mature green	0.41^{b}	0.40^{a}	0.27^{b}	0.27^{a}
	Turning	0.47^{a}	0.36^{b}	0.31^{a}	0.22^{b}
	Light Red	0.31^{e}	0.28^{c}	0.25^{b}	0.18^{c}
LSD (5%)		0.024	0.020	0.028	0.025
CV (%)		3.8	4.0	7.3	6.5

After 15 day of storage all control fruits were spoiled and discarded.

Note: *Means with the same letter within a column are not significantly different.*

The decreasing acidity at the end of storage might be due to use of the acids as energy source with an increase in ripening (Wills et al., 1998; Castro et al., 2005). Similar result was also reported by Felix and Mahendran, (2009) who found that TA declined over the ripening stages due to the climacteric rise in respiration over the degree of ripeness and with maturity evolution where the tomatoes coated in pectin stored had the lowest mean value. In another study, Abassi et al. (2009) reported that chitosan coatings slowed the changes on TA of mango, but on control fruits the rate of decline were significantly higher. Our result is also in

agreement with those reported by Ali et al., (2010) who analyzed the effects of gum arabic as an edible coating for preservation of TA in tomato fruit.

4.2.3 Total soluble solids

Total soluble solids are an important factor to be considered with respect to consumer acceptance. It is expected to increase during ripening and decrease towards senescence (Tasdelen and Bayindirli, 1998). It has been reported that TSS increases with stage of ripeness at harvest (Znidarcic and Pozrl, 2006) and also it generally increases with advancement in maturity during storage (Getinet et al., 2008) which is in agreement with the current result. Borji et al. (2012) noted that, maturity stages at harvest could affect the TSS content of the fruit. The authors found that the TSS content of mature green and full ripe tomatoes was 5.1 and 6.2 ^{o}Brix respectively.

In the present study we observed a significant ($P < 0.05$) interaction effect between coating and maturity stages on the TSS content of the tomato fruits (Appendix Table 5). TSS of control fruits at the end of the storage period (15^{th}day) was 4.8, 4.6, and 4.2 °Brix for fruits harvested at mature green, turning and light red stages, respectively. Whereas tomato fruits coated with pectin resulted in 4.9, 5.4 and 4.9 ^{o}Brix and that of chitosan coated having a 5.1, 5.5 and 5 °Brix for the same stage of harvesting respectively (Table 7). In all cases, fruits harvested at turning stages showed relatively higher TSS values, which might be associated with, higher concentration of organic acid and soluble sugar balance at this stage as compared to early or late mature nature of fruits at both stages of harvesting.

Table 7: Effect of treatment with pectin and chitosan and Maturity stage at harvest on TSS (^{o}Brix) of tomato fruits

Coating materials	Harvesting stages	Days after application of coating materials			
		Day 5	Day10	Day 15	Day 20
Control	Mature green	3.7^{e}	4.5^{d}	4.8^{c}	
	Turning	4.4^{c}	5.1^{ab}	4.6^{d}	
	Light Red	5.1^{a}	4.9^{b}	4.2^{e}	

Chitosan	Mature green	3.3^{f}	4.4^{d}	4.9^{c}	4.4^{c}
	Turning	4.2^{d}	4.9^{b}	5.4^{a}	4.8^{ab}
	Light Red	4.8^{b}	5.2^{a}	4.9^{c}	4.5^{c}
Pectin	Mature green	3.2^{f}	4.1^{e}	5.1^{b}	4.6^{c}
	Turning	4.2^{d}	4.7^{c}	5.5^{a}	5.1^{a}
	Light Red	4.6^{c}	5.2^{a}	5.0^{bc}	4.7^{bc}
LSD		0.21	0.16	0.22	0.23
CV		3	2.0	2.5	2.8

After 15 day of storage all control fruits were spoiled and discarded

Note: *Means with the same letter (s) within a column are not significantly different*

Coatings provide an excellent semi-permeable film around the fruit, modifying the internal atmosphere by reducing O_2 availability for respiration and degradation of macromolecules. Decreased respiration rates slow down the synthesis and use of metabolites resulting in slower rate of increase on TSS (Yaman and Bayoindirli, 2002). The lowest TSS at the end of the storage period was recorded in control fruits at all harvesting stages. The decrease in TSS is caused by a decline in the amount of carbohydrates and pectins, partial hydrolysis of protein and decomposition of glycosides into sub-units during respiration causing a decrease in TSS (Athmaselvi et al., 2013). Similar results in TSS were observed when mangoes were coated with pectin (Moalemiyan et al., 2012).

4.2.4 TSS/TA ratio as a ripening index

The TSS/TA ratio is an important factor for quality parameters of tomato fruits, since it is known that sweetness and sourness are important criteria for tomato flavour (Stevens and Kader, 1995). The relationship between total soluble solids and TA which could be taken as maturity ripening index (RI) showed a significant differences ($P<0.05$) as a function of maturity stage, coating and their interaction (Appendix Table 10). The TSS/TA ratio increased significantly along with increased storage time in both uncoated and coated fruits (Table 8). TSS/TA at green stage for control, pectin and chitosan treated fruits at day 5 was 10.09, 8.52, and 7.91, respectively and subsequently reached 25.65, 17.57, and 19.10 by the end of the storage period, with a significant interaction effect between maturity stages and coatings. Generally coated tomato fruits revealed relatively small ratio changes for all

harvesting stages (Table 8). Similar results were also reported by Al-Mughrabi (1994) who demonstrated that harvesting at mature-green stage had lower TSS/TA ratio values in comparison with red-ripe fruits. In general from result of our study (Table 8), a ratio above 10, can be used as index to determine degree of ripeness of tomato fruits. For instance, uncoated fruit at light red harvesting stage of 5^{th} of storage showed almost equivalent ratio for coated samples at 15^{th} day of storage. As index of ripening, the ratio can be used to investigate the positive effect of coating materials on preserving of total soluble compounds in fruits as compared to uncoated ones.

Table 8 TSS/TA ratio for control, pectin and chitosan coated fruits harvested at different maturity stages and stored at ambient condition

Coating materials	Harvesting stages	Days after application of coating materials			
		Day 5	Day10	Day 15	Day 20
Control	Mature green	10.0^{de}	16.1^{d}	25.6^{c}	
	Turning	10.6^{d}	20.8^{c}	32.7^{b}	
	Light Red	22.0^{a}	36.0^{a}	49.4^{a}	
Chitosan	Mature green	8.5^{f}	12.3^{ef}	19.3^{d}	18.4^{d}
	Turning	9.4^{f}	14.5^{de}	17. 6^{d}	25.0^{b}
	Light Red	17.0^{b}	24.5^{b}	27.7^{c}	29.1^{a}
Pectin	Mature green	7.9^{f}	10.3^{f}	19.1^{d}	16.8^{d}
	Turning	8.8^{f}	13.1^{e}	16.6^{d}	21. 1^{c}
	Light Red	15.1^{c}	19.1^{d}	19.8^{d}	24.8^{b}
LSD		1.49	2.52	4.42	2.54
CV		7.2	7.9	10.2	6.4

After 15 day of storage all control fruits were spoiled and discarded.

Note: *Means with the same letter (s) within a column are not significantly different.*

4.1.5 Ascorbic acid content

Table 9 shows the changes in the ascorbic acid content of tomato fruits at three maturity stages, treated with chitosan and pectin in 20 days of storage time at ambient temperature. Significant differences were observed among treatments ($P<0.05$) for their interaction as indicated in Appendix Table 9. For tomatoes at turning stage, the mean value of ascorbic acid content was 15.80, 34.38 and 38.08 mg/100 g fresh weights for control, chitosan and pectin,

respectively (after 15 days of storage). Green tomatoes showed ascorbic acid values of 17.94, 30.60, and 30.10 mg/100 g fresh weight for control, chitosan, and pectin treatments after 15 days of storage. However, light red tomatoes showed ascorbic acid values of 13.03, 22.20, and 26.70 mg/100 g fresh weight for control, chitosan, and pectin treatments for the same storage period. Sharma et al. (1996) reported ascorbic acid content ranged from 11.21to 53.29 mg/100g in tomato genotypes which is in agreement with values indicated in this study.

Table 9: Changes in ascorbic acid content (mg/100 g) of tomato fruits harvested at three maturity stages coated with pectin and chitosan and stored at ambient condition

Coating materials	Harvesting stages	Days after application of coating materials			
		Day 5	Day10	Day 15	Day 20
Control	Mature green	14.6^{d}	16.7^{e}	17.9^{f}	
	Turning	31.6^{a}	29.6^{b}	15.8^{g}	
	Light Red	21.0^{c}	14.6^{f}	13.0^{h}	
Chitosan	Mature green	9.6^{egf}	20.0^{d}	30.5^{c}	21.1^{d}
	Turning	12.9^{de}	34.7^{a}	34.3^{b}	27.1^{b}
	Light Red	23.5^{b}	25.5^{c}	22.2^{e}	15.7^{f}
Pectin	Mature green	8.3^{g}	26.3^{c}	30.1^{c}	22.6^{c}
	Turning	11.2^{def}	31.0^{b}	38.0^{a}	29.1^{a}
	Light Red	21.0^{c}	30.1^{b}	26.7^{d}	18.3^{e}
LSD		1.87	1.63	1.72	1.41
CV		4.9	3.6	3.9	3.6

After 15 day of storage all control fruits were spoiled and discarded.

Note: *Means with the same letter (s) within a column are not significantly different.*

Similar results were reported by Tigist et al. (2011) who found a general trend of increase in ascorbic acid content, followed by a falling during full ripening stage. The results illustrated in Table 9 show a reduction in ascorbic acid content along with the storage period not only for coated fruits but also for the control. However, a decrease in ascorbic acid content was significantly higher in control as compared with coated fruits. High ascorbic acid in coated fruits could be attributed with slow ripening rate due to semi-permeable membrane films of chitosan and pectin, since coatings serve as a protective layer and control the diffusion of O_2 (Srinivasa et al., 2002) which is critical to initiate respiration processes (Ayranci and Tunc,

2004). The reduction in the extent of loss of ascorbic acid in coated tomato fruits as well could be due to the low oxygen penetrability of the coatings, which might have caused reduced activity of the enzymes and prevented the oxidation of ascorbic acid and hence the rate of conversion of ascorbic acid into dehydro-ascorbic acid could be slowed down during storage (Bal, 2013). Ali et al. (2010) reported a similar slowing down of ascorbic acid degradation for gum Arabic coated tomato during ripening. Similar results have been reported with a high CO_2 storage atmosphere for tomatoes (Mathooko, 2003), where a slowing down of the increase in ascorbic acid during ripening was observed. Commonly, keeping away oxygen from the food delays the deteriorative oxidation reaction of vitamin C (Ayranci and Tunc, 2004). Likewise in Ali et al (2011) papaya fruits coated with chitosan showed a slower initial increase in ascorbic acid as compared to uncoated fruits. This suggests that chitosan and pectin coatings slowed down the synthesis of ascorbic acid during ripening and also slowed down the rate of loss in coated fruits which can be attributed with O_2 availability for respiration and oxidation. Moneruzzaman et al. (2008) also reported that ascorbic acid content was decreased with the advancement of ripening of tomato fruits. It was found that half ripe tomato fruits contained the highest quantity of ascorbic acid while the mature green tomato fruits contained the lowest quantity, which is in line of our result, the highest values from fruits harvested at turning stage.

4.1.6 Lycopene content

Lycopene is the major carotenoid compound in tomatoes, it gives the fruit its characteristic red color (Frusciante et al., 2007). The lycopene content of tomatoes has been previously reported to be in the range of 0.88 to 4.2 mg per 100 g of fresh weight (Clinton, 1999). Apart harvesting stages and coating materials, various factors may influence lycopene content. For example variety, growing condition, maturity, season, geographic origin, fertilizers, soil type, amount of sunlight received and experimental conditions (Storage, extraction) among others might be responsible for the differences (Clinton, 1999).

During ripening the chlorophyll content decreases, and there is a rapid synthesis of the red pigment lycopene. Table 10 shows the changes in the lycopene content of tomato fruits at three maturity stages coated with chitosan and pectin over 20 days of storage at ambient

condition . In the current study, significant ($P < 0.05$) difference was observed on the lycopene content of tomato fruits due to the interaction effect of maturity stages and edible coating materials (Appendix Table 8).

Generally lycopene content of the tomato fruits increased with the storage time in all treated and untreated fruits (Table 10) which was associated with ripening stages. However, the content of untreated fruits increased sharply and reached to a maximum level after 15 days of storage. But similar lycopene concentration was noted from pectin and chitosan coated fruits on 20th day of storage. The ripening and antioxidant index of the tomatoes (lycopene) also varies from one ripening stage to the other and the variations were also observed with coated and uncoated fruits. The results of the study (Table 10) established that the content of lycopene from all treatments increased with storage time but at different rates. The lowest concentration of lycopene (0.11 mg/100 g) was recorded in pectin coated fruits harvested at green stage after 5 days of storage while the highest concentration of 1.1 mg/100 g was measured in chitosan coated fruits which were harvested at light red stage after 20 days of storage.

Table 10: Lycopene (mg/100 g) of tomato fruits harvested at three maturity stages coated with pectin and chitosan and stored at ambient condition

Coating materials	Harvesting stages	Days after application of coating materials			
		Day 5	Day10	Day 15	Day 20
Control	Mature green	0.22^{c}	0.35^{d}	0.81^{c}	
	Turning	0.27^{b}	0.44^{b}	0.88^{b}	
	Light Red	0.30^{a}	0.61^{a}	0.95^{a}	
Chitosan	Mature green	0.14^{e}	0.26^{h}	0.42^{f}	0.93^{ab}
	Turning	0.19^{d}	0.31^{e}	0.51^{e}	0.97^{ab}
	Light Red	0.27^{b}	0.40^{c}	0.59^{d}	1.11^{a}
Pectin	Mature green	0.11^{f}	0.23^{g}	0.37^{g}	0.88^{b}
	Turning	0.20^{d}	0.28^{f}	0.43^{f}	0.94^{ab}
	Light Red	0.23^{c}	0.39^{c}	0.62^{d}	1.05^{a}
LSD (5%)		0.019	0.015	0.038	0.16
CV (%)		3.9	4.7	4.1	9.6

After 15 day of storage all control fruits were spoiled and discarded.

Note: *Means with the same letter (s) within a column are not significantly different.*

The early increase in lycopene content in or control fruits might be due to faster ripening rate of fruits which lead to the conversion of chloroplasts to chromoplasts and lycopene accumulation, in internal membrane system (Grierson and Kader, 1986). Results of our study also in line with Ali et al. (2013) who reported that lycopene content of uncoated tomatoes increased sharply and reached to a maximum peak after 12 days of storage but those coated with gum arabic stayed for 16 days. It has also been reported that the formation of lycopene depends on the rate of respiration during storage (Javanmardi and Kubota, 2006). As indicated in above section, coatings reduce rate of respiration of fruits. Since uncoated fruits exposed to atmospheric oxygen, the lycopene content of red light fruits after 15^{th} days of storage was 0.95, 0.59 and 0.62 mg/100 g, for control, chitosan and pectin coated fruits.

4.1.7 Total polyphenol content

Polyphenols are common constituents of foods of plant origin and are major antioxidants in the human diet. These compounds possess diverse biological properties which provide a number of benefits, including antioxidant, apoptotic, anti aging, anti carcinogenic and anti inflammatory activities, cardiovascular protection, and improvement of endothelial function. Polyphenols also inhibit angiogenesis and cell-proliferation (Han et al.,2007).

The total polyphenols expressed as mg of gallic acid equivalents per 100 g of fruit sample showed some sort of increase in concentration with stage of harvesting and storage time (Table 11). In the current study significant ($P < 0.05$) difference on the total content phenolic content of tomato fruit was observed due to the interaction effect of maturity stages and coatings materials (Appendix Table 7). After 10^{th} days of storage higher values of total phenolic content was observed on fruits harvested at turning stage but coated with pectin (93.2 mg/100 g sample) and followed by chitosan (79.6 mg/ 100 g). The same trend was followed after 15^{th} and 20^{th} days of storage in terms of harvesting stages, but values were decreased when storage time increased to 20 days (Table 11). At this stage of harvesting, fruits could perceived coatings materials as a potential abiotic stress, thereby resulting in production of secondary metabolites like phenols in coated samples (Gonzalez-Aguilar et al.,

2010). Previous studies also showed that low O_2 and high CO_2 concentrations increased the production of phenolic compounds during the storage of fresh cut melons, which was related to oxidative stress on the fruit (Frusciante et al., 2007).

Table 11: Total phenolic contents (mg/g) of tomato fruits harvested at different maturity stages and coated with pectin and chitosan films before storage at ambient condition

Coating materials	Harvesting stages	Days after application of coating materials			
		Day 5	Day10	Day 15	Day 20
Control	Mature green	56.0^{e}	61.4^{ef}	60.8 ^{f}	
	Turning	69.8^{c}	64.4^{e}	50.2^{g}	
	Light Red	64.9^{d}	58.5^{f}	44.3^{h}	
Chitosan	Mature green	52.4^{f}	61.3^{ef}	69.8^{f}	57.5^{e}
	Turning	67.3^{cd}	79.6^{b}	76.8^{b}	70.8^{b}
	Light Red	75.8^{b}	73.3^{c}	68.0^{d}	49.2^{f}
Pectin	Mature green	47.0^{g}	68.1^{d}	73.9^{e}	65.4^{c}
	Turning	68.9^{c}	92.3^{a}	85.4^{a}	79.0^{a}
	Light Red	79.1^{a}	75.8^{c}	70.7^{c}	61.9^{d}
LSD		0.030	0.032	0.025	0.018
CV		2.1	2.7	2.4	1.6

After 15 day of storage all control fruits were spoiled and discarded.

Note: *Means with the same letter (s) within a column are not significantly different.*

The accumulation of phenolic compounds may be promoted by PAL enzyme (Phenylalanine ammonia-lyase) activity, which is activated under stress conditions (Wu and Lin, 2002). Edible coatings can produce abiotic stress on produce, modifying its metabolism and affecting the production of secondary metabolites such as phenolic and flavonoid compounds due to the oxidative stress created by coating (Gonzalez-Aguilar et al. 2010). In grapes treated with edible chitosan coatings, an increase in the PAL enzyme was observed (Romanazzi et al., 2002).

Since phenolic compounds contribute to fruit quality in terms of color, taste, aroma and flavor (Tomás-Barberán and Espín 2001), those coated fruits with higher phenolic content would have higher quality than controls. Furthermore, from health point of view, an increase in total phenolic content is related with the enhancement of antioxidant capacity (Reyes and Cisneros-

Zevallos, 2003) of fruits. Low amount of total phenolic content after 10 days of storage for control fruits might be due to the higher rate of respiration which resulted in the loss of degradation of certain phenolic compounds (Day, 2001). In terms of effects of edible coatings on polyphenol contents, this result is also with work of Liu and others (2007), tthey indicated that production of phenolic compounds was induced in tomato fruit, treated with chitosan. Similarly, Ali et al. (2010) reported the maximum amount of total phenolic content was observed on gum arabic coated fruit and reached to a peak after 12 days and decreased sharply at the final days of storage.

5. SUMMARY AND CONCLUSIONS

Tomato being a highly perishable fruit possesses very short shelf life and reaches to respiration peak of the ripening process. To prevent high postharvest losses, especially in the developing countries, like Ethiopia where losses are very high, the application of simple technologies is very beneficial. In view of easy adoption and sustainability of technologies, the use of edible coatings could be a good alternative since it is simple, low-cost and environmentally friendly alternative to extend shelf life and maintain quality. The additional benefit conferred by edible coatings is that these are natural products and are not chemically synthesized.

Application of edible films on a highly perishable commodity like tomato extends shelf life which in turn will promote the sustained availability of the produce and provide higher returns for producers, retailers and the country. Coating can lowered both the rate of substrate catabolism and the ability to generate the energy required to drive the biochemical reactions associated with fruit ripening. Coatings also favorably influenced several physiological properties of the fruits during storage. They slowed down the rate of respiration, color changes, softening of the tissue and increased the shelf-life of the fruits.

In brief in this study, it was observed that surface coating of tomato fruits using pectin and chitosan solution can significantly delay changes in quality attributes such as weight, firmness, total color change, TA, TSS, pH, ascorbic acid, phenolic content, lycopene content, and disease incidence and severity which all together extended the shelf life of fruits during ambient storage. Maximum shelf life was observed for tomatoes harvested at turning stage coated with pectin followed by chitosan. Thus, the present study indicated that coated tomato fruits had the longest shelf life with better and acceptable quality,. Extending the storage life of tomatoes may enable growers, wholesalers and retailers to have a relatively longer period of time to transport and market their produce.

The most suitable stage of harvest to apply coating, for both chitosan and pectin, was turning stage. Harvesting at turning stage and coating preserved the quality of tomato fruits to the

greatest extent. In conclusion tomato fruits harvested at turning stage and coated with either chitosan or pectin can satisfactorily preserve fruit quality and extend the shelf life of fresh tomato fruits. As both Chitosan and Pectin are found to be equally effective, the choice between the two depends on price and availability. This study recommends pectin coating since cost wise it is cheaper than chitosan.

Edible coating technique seems to be very promising as long as consumers accept this technique as safe. The coatings also offer an attractive glossy sheen to the produce and may protect from bruising and injury. However, all the above conclusions were derived from results of this study conducted only once and with a single variety of tomato. Therefore, further studies could be conducted with consideration of more type of coating materials for a wide range of tomato varieties in order to draw a comprehensive recommendation of relatively cheap and most available but effective coating material(s). Further combination studies are also essential to investigate effect of coating materials with various type of packaging materials and storage temperature for further extension of storage life and better quality product..

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7 APPENDICES

Appendix Table 1 Overall ANOVA table for weight loss

Source of variation	DF	Mean Square	F Value	Pr> F
Coating material	2	36.36090370	243.78	<.0001
Harvesting stage	2	14.94213704	100.18	<.0001
Coating * Harvesting stage	4	0.52908148	3.55	0.0266

CV = 4.2

Appendix Table 2: Overall ANOVA for total color change

Source of variation	DF	Mean Square	F Value	Pr> F
Coating material	2	344.5350184	61.48	<.0001
Harvesting stage	2	207.2460103	36.98	<.0001
Coating * Harvesting stage	4	2.2188161	0.40	0.8089

CV = 10.0

Appendix Table 3 Overall ANOVA for firmness

Source of variation	DF	Mean Square	F Value	Pr> F
Coating material	2	11.07349259	256.40	<.0001
Harvesting stage	2	6.42634537	148.14	<.0001
Coating * Harvesting stage	4	0.43741065	10.06	0.0002

CV = 5.0

Appendix Table 4 Overall ANOVA for titrable acidity

Source	DF	Mean Square	F Value	Pr> F
Source of variation	2	0.03604444	256.11	<.0001
Harvesting stage	2	0.04674444	332.13	<.0001
Coating * Harvesting stage	4	0.00042222	3.00	0.0464

CV = 4.1

Appendix Table 5 Overall ANOVA for total soluble solid

Source of variation	DF	Mean Square	F Value	Pr> F
Coating material	2	1.12000000	73.76	<.0001
Harvesting stage	2	0.49333333	32.49	<.0001
Coating * Harvesting stage	4	0.12166667	8.01	0.0007

CV 2.5

Appendix Table 6 Overall ANOVA for pH

Source of variation	DF	Mean Square	F Value	Pr> F
Coating material	2	0.05512593	201.14	<.0001
Harvesting stage	2	0.14588148	532.27	<.0001
Coating * Harvesting stage	4	0.00102037	3.72	0.0223

CV 0.37

Appendix Table 7 Overall ANOVA for phenolic compound

Source of variation	DF	Mean Square	F Value	Pr> F
Coating material	2	677.694815	186.56	<.0001
Harvesting stage	2	530.589259	146.06	<.0001
Coating * Harvesting stage	4	107.822593	29.68	<.0001

CV = 2.7

Appendix Table 8 Overall ANOVA for Lycopene content

Source of variation	DF	Mean Square	F Value	Pr> F
Coating material	2	0.01234746	136.27	<.0001
Harvesting stage	2	0.04478775	494.28	<.0001
Coating * Harvesting stage	4	0.00087290	9.63	0.0002

CV = 4.6

Appendix Table 9 Overall ANOVA for Ascorbic acid

Source of variation	DF	Mean Square	F Value	Pr> F
Coating material	2	368.6772620	356.62	<.0001

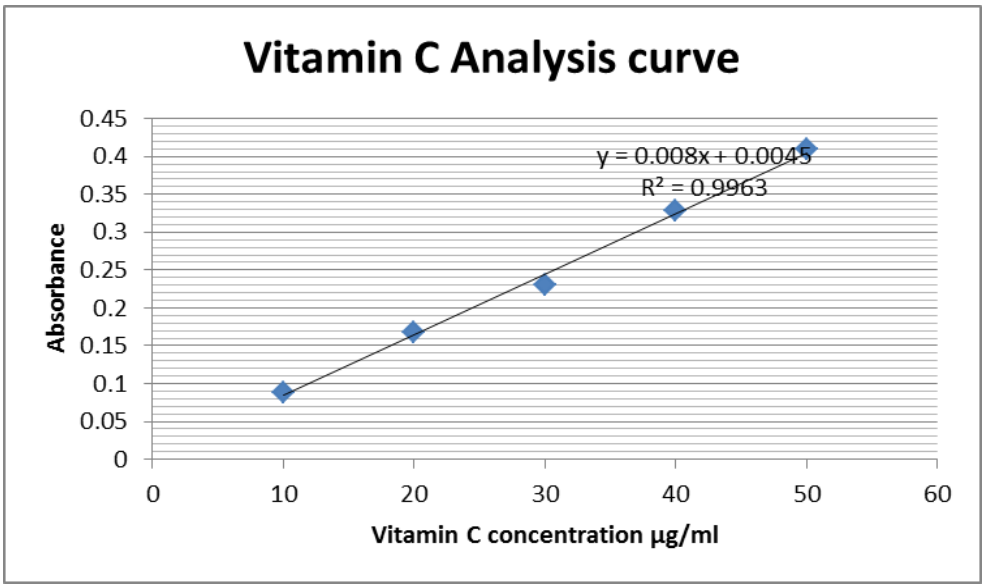
Harvesting stage	2	198.2554578	191.77	<.0001
Coating * Harvesting stage	4	21.1895711	20.50	<.0001

CV 3.29

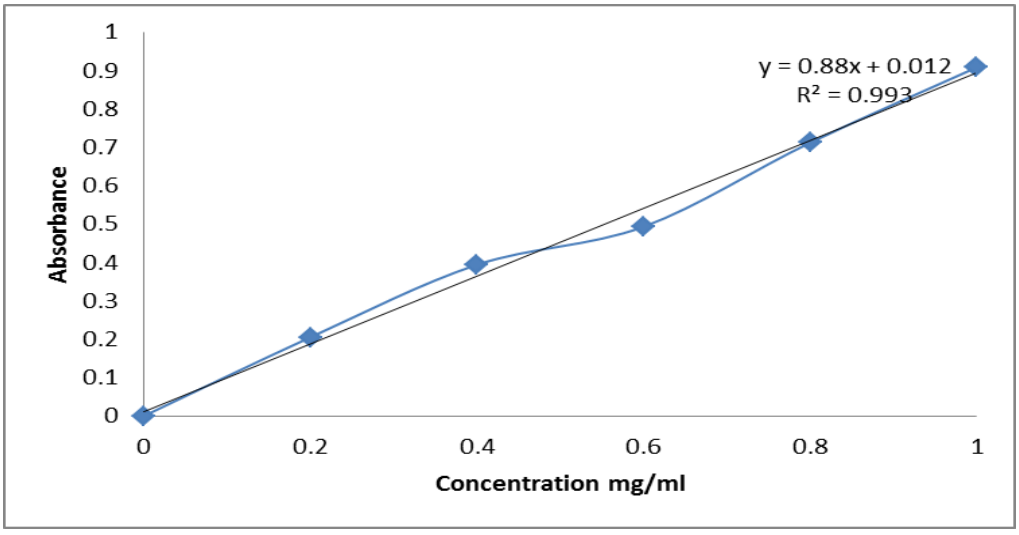
Appendix Table 10: Overall ANOVA for maturity index

Source of variation	DF	Mean Square	F Value	Pr> F
Coating material	2	777.691393	116.93	<.0001
Harvesting stage	2	341.440180	51.34	<.0001
Coating * Harvesting stage	4	101.892446	15.32	<.0001

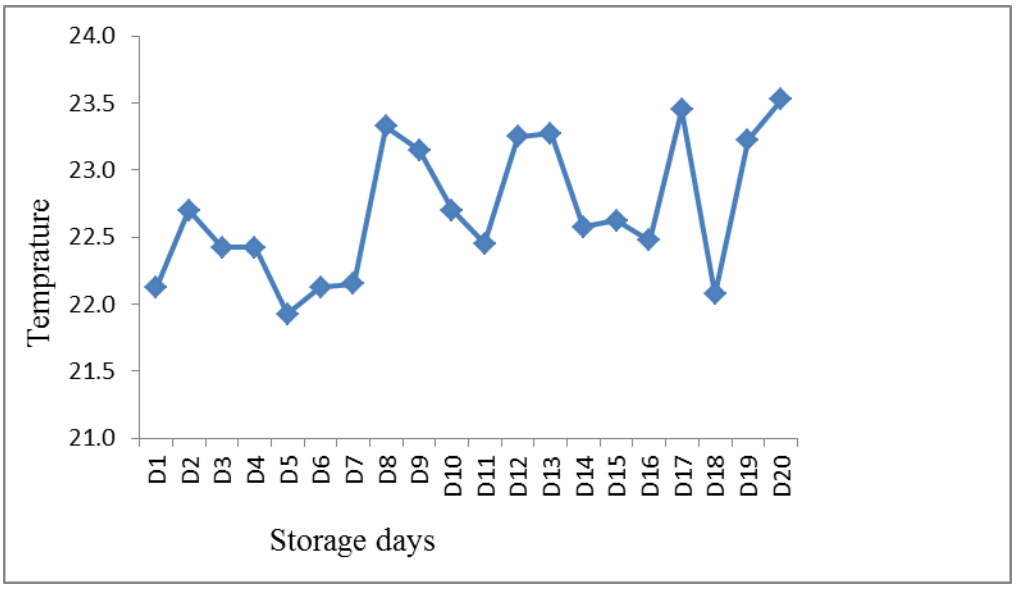
CV 10.2



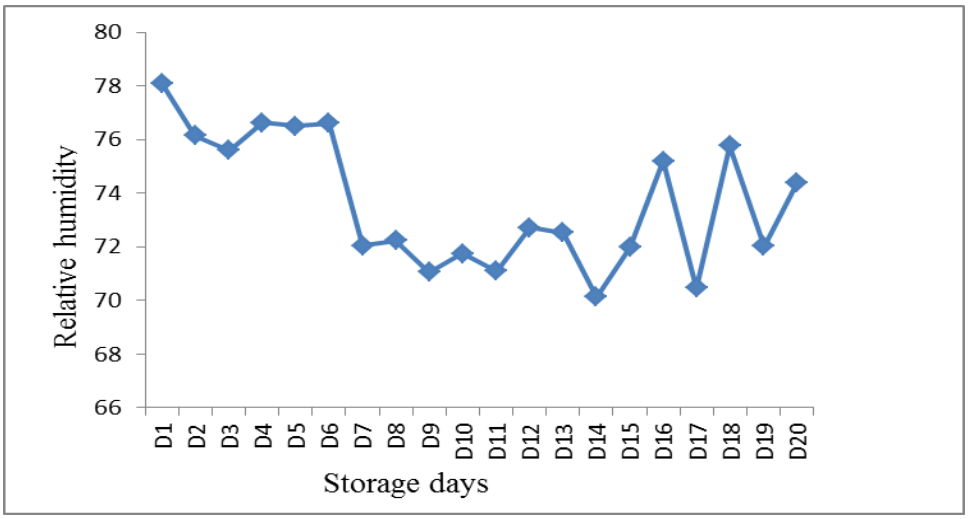
Appendix Figure 1: Standard curve of vitamin C analysis



Appendix Figure 2: Standard curve of phenolic content analysis



Appendix Figure 3: The average daily temperature of the storage room for 20 days



Appendix Figure 4: The average daily relative humidity of the storage room for 20 days