ENHANCEMENT OF SHELF LIFE OF POMEGRANATE ARILS USING EDIBLE COATING

THESIS SUBMITTED IN PARTIAL FULFILMENT OF THE
REQUIREMENTS FOR THE DEGREE OF

Master of Science
IN
Food Science and Technology
(Dairy Science and Food Technology)

Supervisor
Dr. Arvind
Assistant Professor

Submitted By
Ajeet Kumar

DEPARTMENT OF DAIRY SCIENCE & FOOD TECHNOLOGY
INSTITUTE OF AGRICULTURAL SCIENCES
BANARAS HINDU UNIVERSITY
VARANASI-221005
INDIA

I. D.No.:18412FST002 **2020** **Enrolment No.:410534**

Dr. Arvind
Assistant Professor

DEPARTMENT OF DAIRY SCIENCE & FOOD TECHNOLOGY,
INSTITUTE OF AGRICULTURAL SCIENCES
BANARAS HINDU UNIVERSITY
VARANASI - 221005,
U.P. (INDIA)

Ref. No.:................ Date:…………

CERTIFICATE

The Registrar
Banaras Hindu University,
Varanasi-221005, U.P. (India).

Through: **The Head**,
Department of Dairy Science & Food Technology, I. Ag. Sc., BHU.

Dear Sir,

I have great pleasure in forwarding the thesis entitled **"ENHANCEMENT OF SHELF LIFE OF POMEGRANATE ARILS USING EDIBLE COATING"** submitted by **Mr. AJEET KUMAR, I.D. No. 18412FST002, Enrollment No.- 410534,** in partial fulfillment of the requirement for the degree of **Master of Science, Food Science and Technology,** Department of Dairy Science & Food Technology, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi.

This is to certify that the work has been carried out solely by **Mr. Ajeet Kumar** under my supervision and guidance and his findings and data presented herein are genuine and original to the best of my knowledge and belief and no part of the work has been submitted for any other degree or institution.

Thanking You.

FORWARDED

Yours faithfully

Head of Department

Dr. Arvind
(Supervisor)

ENHANCEMENT OF SHELF LIFE OF POMEGRANATE ARILS USING EDIBLE COATING

Submitted By
AJEET KUMAR

Thesis submitted in partial fulfillment of the requirements
for the degree of
Master of Science
In
Food Science and Technology
(Dairy Science and Food Technology)
INSTITUTE OF AGRICULTURAL SCIENCES
BANARASHINDUUNIVERSITY, VARANASI.
2020

I.D.No.18412FST002 **Enrolment No. 410534**

APPROVED BY ADVISORY COMMITTEE

CHAIRMAN : **Dr. Arvind**
Assistant Professor and Advisor
Department of Dairy Science & Food Technology

MEMBER : **Dr. Abhishek Dutt Tripathi**
Assistant Professor
Department of Dairy Science & Food Technology

MEMBER : **Dr. Kalyan Barman**
Assistant Professor
Department of Horticulture

EXTERNALEXAMINE

ACKNOWLEDGEMENT

I bow my head to almighty **God** whose blessings are always with me and with which I have completed my course work and research work satisfactorily without facing any obstruction. With all praises and glory to our, I bow my head to **Buddha** who works as a spiritual support and inspiration for me to live the life with joy, courage, self-confidence, helping others, honesty and positive attitude.

With great respect and honour and devotion, I offer flowers of reverence to **Bharat Ratna Mahamana Pandit Madan Mohan Malaviya Ji**, the founder of the Banaras Hindu University, a great social reformer for his values for life, his sacrifices and struggle for the sake welfare of humanity, maintaining communal harmony and overall development.

With immense pleasure and profound sense of gratitude, I take this opportunity to express my heartfelt and sincere thanks to my esteemed supervisor, **Dr. Arvind** Department of Dairy Science And Food Technology, Institute of Agricultural Sciences, **Banaras Hindu University**, for his meticulous guidance, indelible inspiration, persistent encouragement, ingenious suggestions, mellifluous nature and indefatigable attitude. I will ever cherish the fatherly affection that he bestowed upon me throughout my tenure as a student under him which helped me to cope with many difficult situations.

I extend my indebtedness to **Prof. D. C. Rai**, Head, Department of Dairy Science And Food Technology, Institute of Agricultural Sciences, Banaras Hindu University for providing all the necessary facilities, discerning comments, valuable suggestions, co-operations and helpful attitude towards me during the course of investigation.

I wish to express my heartfelt and advent thank to the members of my Advisory Committee, **Prof. D.C. Rai,** Head, Department of Dairy Science And Food Technology and **Dr. Kalyan Barman,** Department of Horticulture for their valuable suggestions and liberal help rendered during the course of study and research work.

I deem it my privilege in expressing my gratefulness to **Dr. V. K. Paswan,** Assistant Professor, Department of Dairy Science And Food Technology, **Dr. A. Poonia**, Assistant Professor, Department of Dairy Science And Food Technology, **Er. D.S. Bunkar,** Assistant Professor, Department of Dairy Science And Food Technology, **Dr. A.D. Tripathi**, Assistant Professor, Department of Dairy Science And Food Technology, Banaras Hindu University, Varanasi for their timely help.

With profound regards in a more personal sense, I owe deepest debts to my beloved and venerable father ***Shri Ramashankar*** *and mother* ***Smt. Shusheela Devi*** *and my sister* ***Meera*** *for their blessing and exhilaration that always animated me to rise against the problems and face them smilingly, how so much formidable they may be. They kept me enthusiastic throughout my educational career, which enabled me to acquire the present gratification.*

I pay my sweet and heartiest thanks to my generous seniors, ***Mr. Sujit, Mr. Pankaj Anu ,Mr. Aman Rathor, Mr. Alok Mishra, Mr. Abhinay Jha, Miss Vina Paul, Miss Sikha Pandhi ,Mr. Ashutosh Singh, Mr. Pradeep*** *who displayed profound sense of fraternity with elderly help and valuable suggestions to complete this gigantic task.*

I acknowledge my heartfelt appreciation to my batchmates and my friend ***Neha Yadav, Suman Bharti, Vini Kasturi, Smriti Kumari, Yogita, Anjali Jaiswal, Akash, Vishal, Rabindranath Mishra, Nidhi Tomar, Subarna Deb, and Arshya Singh*** *for their affection, pains and indispensable help, which made my study period in the university cheerful. So, thanks a lot for your belief, encouragement, support and love.*

I am also very thankful to all the ***non-teaching staff members Shri Himanshu Rai, Shri Sonkar, Shri Chandrasekhar, Shri Anand, Shri Amresh*** *of Department of Dairy Science and Food Technology, Institute of Agricultural Sciences, Banaras Hindu University.*

Above all, my humble and wholehearted gratitude to ***Teerthanker Mahaveer*** *for his blessings.*

Date :
Place : *Varanasi.* **(Mr. Ajeet Kumar)**

CONTENTS

LISTS OF TABLES

LISTS OF FIGURES

ABBREVIATION

% : Per cent
@ : At the rate of
^{o}C : Degree Centigrade
AAS : Atomic Absorption Spectrophotometer
SFE : Supercritical Fluid Extraction
PPE : Pomegranate Peel Extracts
C : Control or Untreated samples
S.A : Sodium Alginate
A.O : Antioxidant
AOAC : Association of Official Analytical Chemists
M.P : Minimally Processed
e.g : For Example
Avg. : Average
Etc : Et cetera
g : Gram
Ca : Calcium
min : Minutes
i.e : That is
Kcal : Kilo calorie
LDPE : Low Density Poly Ethylene
N : Normality
Cu : Copper
K : Potassium
v/v : Volume per volume
c.f.u : Colony forming Units
TSS : Total soluble Solids

CMC	:	Carboxy methyl cellulose
et. al	:	And other (Latin et alii)
FAO	:	Food and Agriculture Organization
TPC	:	Total Phenolic Content
PDA	:	Potatoes dextrose agar
Fe	:	Iron
Fig.	:	Figure
HPC	:	Hydroxy propyl cellulose
g/d	:	Gram per day
RH	:	Relative humidity
L	:	Lactose
OA	:	Overall acceptability
pH	:	Negative logarithm of hydrogen ion
h	:	Hour
mg	:	Miligram
SPC	:	Soy protein concentrate
AA	:	Ascorbic acid
MC	:	Methyl cellulose

Chapter- I

INTRODUCTION

Pomegranate (*Punica granatum L.*) fruits are commonly consumed in both fresh arils and commercial processed forms, such as juice, dips and flavorings in the Middle East, China, India, and America. The fruit gives rise to three parts: seeds (about 3%), juice (about30%) and peels, each of which has interesting Pharmacologic activity (Lansky and Newman, 2007 and Palma *et al.,* 2015).

Pomegranate juice possesses Anticancer activities, including interference with tumour cell proliferation and improves lipid profiles in diabetic Patients with hyperlipidemia (Lansky and Newman, 2007; Tezcan *et al.,* 2009; Caleb *et al.,* 2013 and Li *et al.,* 2014).

Pomegranate fruit contains many different kinds of polyphenolic compounds and commercial Pomegranate juice has been shown to possess Antioxidant activity three times higher than those of Green tea (Gil *et al.,* 2000 and Caleb *et al.,* 2013).

The Soluble polyphenolic content of pomegranate juice includes anthocyanins, punicalagins, catechins, ellagic acids, tannins, and gallic acid (Aviram *et al.,* 2000; Gil *et al.,* 2000 and Caleb *et al.,* 2013). A linear relationship between total phenolic content, antioxidant capacity and antibacterial activity against several microorganisms has been established by Shan *et al.* (2007) and Palma *et al.* (2015).

Maintaining the quality of pomegranate arils is a critical challenge, they are very susceptible to textural and nutritional deterioration; this lead to reduce shelf life (Maghoumi *et al.,* 2013). Optimum cold storage condition is essential in order to minimize physiological disorders of pomegranate fruit (Caleb *et al.,* 2012).

However, the commercial shelf life of arils differs towhole fruit. Whole fruit may be stored for 3- 4 months at temperatures below 10 °C (Ghafir *et al.,* 2010), while Arils shelf life varies from 7 to 18 days at temperatures between 0 and 5 °C under different packaging conditions (Artés *et al.,* 2000; Ayhan *et al.,* 2009).

The antibacterial properties of pomegranate havebeen reported extensively for several pathogens such as *Escherichia coli, Staphylococcus aureus,* including several methicillin resistant strains like *Staphylococcus aureus, Vibrio cholera* and *Bacillus subtilis* (Braga e*t al.,* 2005; Melendez; Capriles, 2006 and Caleb *et al.,* 2013).

Scientific community reflecting consumers demand for natural antimicrobials has made efforts to investigate the possibility to use natural sources of antimicrobials (Drosionon *et al.*, 2009).

India is the largest producer of pomegranate (*Punica granatum*) next only to Iran. Pomegranate fruit is high in natural antioxidants and are known to fight against cancer, infections and other diseases in humans.

Pomegranate peels are being discarded after juice production and ready to eat arils. Pomegranate peel is nutritive rich by product which are abundant due to no more use.

Pomegranate peel attracts attention due to its apparent wound healing properties (Chidambaram et al., 2004), immune modulatory activity (Gracious et al., 2001), and antibacterial activity (Navarro *et al*., 1996) anti atherosclerotic and anti-oxidative capacities (Tzulker *et al*., 2007). Anti-oxidative activity has often been associated with a decreased risk of various diseases (Whitley et al., 2003).

The cultivar per se (genotype) behaved as the most influencing factor conditioning pomegranate sugar, organic acid profiles, antioxidant activity and total phenolic. The genotype factor should be considered as the most influencing factor in future breeding programs to enhance the synthesis of beneficial bioactive compounds (Pilar Legua *et al*., 2012).

Pomegranate peel is rich source of ellagitannin (antioxidant) and thus may serve in the prevention of cattle diseases and improvement of beef products making it an attractive component in cattle feed. Recent studies also have shown that boosting antioxidant levels in the diet of cattle may help to improve their health.

The peel packs some of the weight boosting and health enhancing effects of antibiotics and hormones without the detrimental effects and it may yield meat with higher level of beneficial antioxidants (Shabtay *et al*., 2008).

Pomegranate ellagitannin have been identified as the active antioxidant compound and anticancer activities responsible for protecting low density lipoprotein, cholesterol from oxidation in vivo a key step in the pathogenesis of atherosclerosis. Pomegranate peel and its extracts are also being investigated for their potentialuses as food bio preservatives, formulation of products in nutraceutical industry and cattle feed (N. Seeram *et al*., 2005).

Pomegranate consumption is limited due to difficulties in peeling to obtain the arils. On the other hand, pomegranate is very sensitive to sunburn, cracking, cuts, or bruises in the husk, as well as to chilling injury (Artes *et al*., 2000). Despite their excellent internal quality, these diverse external defects make the injured fruit unsuitable for fresh marketing and consumption, and usually, they are destined for industrial use or animal consumption (Artes *et al.*, 2000). The processing of the externally damaged pomegranates that are unacceptable for fresh marketing and consumption could be an excellent way to obtain a commercial profit from discarded pomegranate fruits (Lopez *et al*., 2005).

Just as society has evolved over time, food system has evolved over centuries into a global system of immense size and complexity. Production of pomegranate arils in “ready-to-eat” form would be a convenient and desirable alternative to the consumption of fresh fruits and may increase pomegranate demand by consumers (Ghasemnezhad *et al*., 2013). Minimally processed pomegranate arils would be more appealing to customers than whole fruits and increase fresh consumption of pomegranate fruits (Muharrem and Nazan, 2009).

Raw material entering the food industry represents a potential source of microbial contamination. The potential growth of pathogens and microorganisms will be affected by the initial level of contamination and the processing steps in eliminating bacteria in the food. Generally, different microbial populations are present in contaminated food, and some groups become more predominant after a certain storage period. This selection might depend on several physico-chemical factors and processing and storage conditions. In the last years, different procedures have been reported for the establishment of shelf-life, mainly based on the detection of microbial alteration, as well as physico-chemical and sensorial changes (Valero *et al*., 2012).

However, maintaining the nutritional and microbial quality of pomegranate arils is a major challenge as minimally processed arils easily deteriorate in texture, color, overall quality, and consequently, shelf-life is reduced (Caleb *et al*., 2012). This is due to the active metabolic processes related to endogenous enzymatic activity, enhanced respiration rate with increased production of ethylene (Muharrem and Nazan , 2009), and increased microbial load, that some of which may be potentially harmful to human health (Caleb *et al*., 2012). If not controlled, these changes can lead to rapid senescence and deterioration of the product (Muharrem and Nazan, 2009).

Alginate is a natural polysaccharide extracted from brown seaweeds from the family of Phaeophyceae. It has several film-forming properties such as transparency and uniformity and serves as an excellent barrier to moisture. In particular, alginate has unique colloidal properties allowing its use as a thickening agent and a stabilizing material. It is widely used to enhance the shelf life of many fruits and vegetables by reducing dehydration, controlling respiration, and improving mechanical properties. Several studies reported the use of alginate for delaying the ripening process of tomato, pineapples, watermelon, and sweet cherries.

Ascorbic acid (AA) is widely used as food ingredient because of its reducing and antioxidant properties in addition to its function as an essential nutrient. Ascorbic acid has been shown to effectively scavenge superoxide, hydrogen peroxide, hypochlorite, the hydroxyl radical, per oxylradical, and singlet oxygen. Controlling lipid oxidation in peanuts using WPI coatings has been reported. However, no study has been reported about antioxidant effects of an AA-incorporated WPI coating (AA-WPI coating) on foods.

OBJECTIVES OF THE STUDY

1. To optimize the coating materials for pomegranate arils.
2. To Study the shelf life of coated pomegranate arils.

* * *

Chapter-II

REVIEW OF LITERATURE

Pomegranate (*Punica granatum* L.) is one of the most important fruits native to Iran and some parts of the Mediterranean area. It is also extensively cultivated in Spain, Egypt, Russia, India, France, China, Japan, and USA in recent years (Varasteh *et al.*, 2012). It has been cultivated since ancient times throughout the Mediterranean basin to India, and is highly adaptable to adverse climatic conditions and different soil types (Muharrem and Nazan, 2009). Aril is the edible part of pomegranate fruit that represents 50–70 % total weight of the fruit (Safa and Khazaei, 2003). It is composed of 10 % sugar (mainly fructose and glucose), 1.5 % organic acids (principally ascorbic acid, citric, and malic acid) and bioactive compounds such as anthocyanins and other phenolic compounds (Safa and Khazaei, 2003).

The skin and aril color of pomegranate genotypes, depend on the cultivar, growing region, ecological conditions during fruit maturation, and ripening (Holland *et al.*, 2009). Therefore, the arils of the pomegranate have differences in skin color and thickness, amount of water, taste, and hardness; these are the most important features of the different pomegranate genotypes. The color is most effective on consumer purchase decision. In addition, the others important parameters are texture, flavor, and nutritional value (Holland *et al.*, 2009).

Pomegranate consumption is limited due to difficulties in peeling to obtain the arils. On the other hand, pomegranate is very sensitive to sunburn, cracking, cuts, or bruises in the husk, as well as to chilling injury (Artes *et al.*, 2000). Despite their excellent internal quality, these diverse external defects make the injured fruit unsuitable for fresh marketing and consumption, and usually, they are destined for industrial use or animal consumption (Artes *et al.*, 2000). The processing of the externally damaged pomegranates that are unacceptable for fresh marketing and consumption could be an excellent way to obtain a commercial profit from discarded pomegranate fruits (Lopez *et al.*, 2005).

India is the largest producer of pomegranate (*Punica granatum*) next only to Iran. Pomegranate fruit is high in natural antioxidants and are known to fight against cancer, infections and other diseases in humans. Pomegranate peels are being discarded after juice production and ready to eat arils. Pomegranate peel is nutritive rich by product which are abundant due to no more use. Pomegranate peel attracts attention due to its apparent wound healing properties (Chidambara *et al*., 2004), immune modulatory activity (Gracious *et al.*,2001), and antibacterial activity (Navarro *et al*., 1996) ant atherosclerotic and ant oxidative capacities (Tzulker *et al*., 2007).

Antioxidative activity has often been associated with a decreased risk of various diseases (Whitley *et al.*, 2003). The cultivar per se (genotype) behaved as the most influencing factor conditioning pomegranate sugar, organic acid profiles, antioxidant activity and total phenolics. The genotype factor should be considered as the most influencing factor in future breeding programmes to enhance the synthesis of beneficial bioactive compounds (Pilar Legua *et al*., 2012).

Pomegranate peel is rich source of ellagitannin (antioxidant) and thus may serve in the prevention of cattle diseases and improvement of beef products making it an attractive component in cattle feed. Recent studies also have shown that boosting antioxidant levels in the diet of cattle may help to improve their health. The peel packs some of the weight boosting and health enhancing effects of antibiotics and hormones without the detrimental effects and it may yield meat with higher level of beneficial antioxidants (Shabtay *et al*., 2008).

Pomegranate ellagitannin have been identified as the active antioxidant compound and anticancer activities responsible for protecting low density lipoprotein, cholesterol from oxidation in vivo a key step in the pathogenesis of atherosclerosis. Pomegranate peel and its extracts are also being investigated for their potential uses as food bio preservatives, formulation of products in nutraceutical industry and cattle feed (N. Seeram *et al*., 2005).

Table 2.1 Pomegranate Nutrition Chart

Nutrient	**Unit**	**Value per 100 g**
Water	g	77.93
Energy	kcal	83
Protein	g	1.67
Total lipid (fat)	g	1.17
Carbohydrate, by difference	g	18.70
Fiber, total dietary	g	4.0
Sugars, total	g	13.67
Calcium, Ca	mg	10
Iron, Fe	mg	0.30
Magnesium, Mg	mg	12
Phosphorus, P	mg	36
Potassium, K	mg	236
Sodium, Na	mg	3
Zinc, Zn	mg	0.35
Vitamins		
Vitamin C, total ascorbic acid	mg	10.2
Thiamin	mg	0.067
Riboflavin	mg	0.053
Niacin	mg	0.293
Vitamin B-6	mg	0.075
Folate, DFE	microgram	38
Vitamin B-12	microgram	0.00
Vitamin A, RAE	microgram	0
Vitamin A, IU	IU	0
Vitamin E (alpha-tocopherol)	mg	0.60
Vitamin D (D2 + D3)	microgram	0.0
Vitamin D	IU	0
Vitamin K (phylloquinone)	microgram	16.4
Lipids		
Fatty acids, total saturated	g	0.120
Fatty acids, total monounsaturated	g	0.093
Fatty acids, total polyunsaturated	g	0.079
Fatty acids, total trans	g	0.000
Cholesterol	mg	0
Other		
Caffeine	mg	0

As per United States Department of Agriculture (USDA)

Kotamballi *et al.*, (2002) Studied Antioxidant Activity of Pomegranate (*Punica granatum*) Peel Extract Using *in Vivo* Models and demonstrated that peel extracts possess significant antioxidant activity in various in vitro models. Methanolic extract of pomegranate peel at 50 mg/kg (in terms of catechin equivalents) followed by CCl4 treatment causes preservation of catalase, peroxidase, and SOD to values comparable with control values, where's lipid peroxidation was brought back by 54% as compared to control . Histopathological studies of the liver were also carried out to determine the hepatoprotection effect exhibited by the pomegranate peel extract against the toxic effects of CCl4. Histopathological studies of the liver of different groups also support the protective effects exhibited by the MeOH extract of pomegranate peel by restoring the normal hepatic architecture.

Nam *et al.* (2002) demonstrated hem preventive and adjuvant therapeutic potential of pomegranate (*Punica granatum*) for human breast cancer. Their actions, and of the crude whole oil and crude fermented and unfermented juice concentrate, were assessed *in vitro* for possible chemo preventive or adjuvant therapeutic potential in human breast cancer. The ability to effect a blockade of endogenous active estrogen biosynthesis was shown by polyphenols from fermented juice, pericarp, and oil, which inhibited aromatase activity by 60–80%. Fermented juice and pericarp polyphenols.

Fernando *et al.* (2006) performed In vitro susceptibility of *Entamoeba histolytica* and, *Giardia lamblia*to plants used in Mexican traditional medicine for the treatment of gastrointestinal disorders. species showed selectivity and significant antiprotozoal activity were *Chiranthodendron pentadactylon*, *Annonacherimola* and *Punica granatum* were the most active on *Entamoeba histolytica* with IC50 <30_g/ml. *Dorstenia contrajerva*, *Sennavillosa* and *Rutachale pensis*were the most active toward *Giardia lamblia* with IC50 <38_g/ml. The potency of *Chiranthodendron pentadactylon* (IC50 2.5_g/ml) on *Entamoeba histolytica* was close that of to emetine, but far less than metronidazole, drugs used as control.

Surinderkumar *et al.* (2007) evaluated Antihepatotoxic Effect of *Punica granatum* Acetone Extract against isoniazid and rifampicin induced hepatotoxicity.

Study has investigated the effect of 70% acetone extract of *Punica granatum* L. fruits on hepatic marker enzymes, antioxidants, and tissue per oxidative damage during isoniazid- and rifampicin-induced hepatotoxicity. These findings demonstrated the hepatoprotective potential of the acetone extract of *Punica granatum* fruits on tissue defense systems during isoniazid- and rifampicin-induced hepatotoxicity in rats.

Qna *et al.* (2007) studied antidiarrheal Activity of the aqueous extract of *Punica ngranatum* (Pomegranate) peels. The antidiarrheal effects of the aqueous extract of *Punica granatum L.* (Punicaceae) peels were evaluated in rats. Studies were carried out on the isolated rat ileum, gastrointestinal motility in vivo, and on castor oil–induced diarrhea in rats. The results revealed that the extract exhibited a concentration-dependent inhibition of the spontaneous movement of the isolated rat ileum and attenuated acetylcholine-induced contractions. The extract (100, 200, 300, and 400 mg=kg) also caused a dose-dependent decrease of gastrointestinal transit and markedly protected rats against castor oil–induced diarrhea enter pooling.

Yasoubi *et al.* (2007) reported total phenolic contents and antioxidant activity of Pomegranate (*Punica granatum* L.) peel extracts. The phenolic compounds of pomegranate (*Punica granatum* L.) peel extracted by two methods (solvent and ultrasound-assisted) with five solvents (acetone, methanol, ethanol, water and ethyl acetate) were compared with supercritical fluid extraction (SFE). The total phenolic compounds were determined according to the Folin-Ciocalteu reagent using tannic acid as standard. The overall results showed that acetone with sonication produced the maximum amount of phenolic compounds from pomegranate peel extracts (PPE).

Naqvi *et.al.* (2009) has studied the ant amoebic activity of water soluble fractions of the rind and flowers of *Punica granatum usinge* *in vitro* and *in vivo* studies, on a virulent strain of *Entamoeba histolytica*, exhibited encouraging results.

Al-Zoreky (2009) studied antimicrobial activity of pomegranate (*Punica granatum* L.) fruit peels against some food-borne pathogens by various extracts from pomegranate fruit peels using both *in vitro* (agar diffusion) and *in situ* (food) methods. The 80% methanolic extract of peels (WME) was a potent inhibitor for *Listeria monocytogenes*, *S. aureus*, *Escherichia coli* and *Yersinia enterocolitica.* The minimum inhibitory concentration (MIC) of WME against *Salmonella enteritidis*was the highest (4 mg/ml). WME afforded > 1 log10 reduction of *L. monocytogenes*in food (fish) during storage at 4 °C. Phytochemical analyses revealed the presence of active inhibitors in peels, including phenolics and flavonoids. The activity of WME was related to its higher content (262.5 mg/g) of total phenolics.

Hülya (2011) studied antibacterial and antifungal activity of pomegranate (*Punica granatum* L.) peels. The antibacterial activity of peel extracts was tested against three bacteria strains, which were named *Staphylococcus aureus* (ATCC 25923), *Escherichia coli* (ATCC 25922) and *Salmonella, Enteritidis*(ATCC 13076). The antifungal activity wastested against two fungal strains, which were named *Aspergillus parasiticus* NRRL 2999 and *Aspergillus parasiticus* NRRL 465. All extracts possessed remarkable antibacterial and antifungal activities against all tested bacterial and fungal strains.

2.2 EDIBLE COATING

An edible coating is defined as a thin layer of edible material applied to the surface of food products to extend its shelf life, by reducing moisture and solute migration, gas exchange, respiration, and oxidative rates, as well as by reducing or even suppressing physiological disorders (Quirós-Sauceda *et al.*, 2014; Kester and Fennema., 1986). The main advantage of using edible films and coatings is that several active ingredients can be incorporated into the produce, thus enhancing safety or even nutrition and sensory attributes.

Edible coating is used to improve food appearance and provide safety to the food by its environmental friendly nature. It may be obtained from both animal and vegetable sources. Coating may be of protein, lipid, polysaccharide, resin, nature alone or in combination. They act as a barrier for moisture and gases during processing, handling and storage. It reduces food deterioration and enhances safety by their activity or by incorporation of antimicrobial compound. Other advantages of using edible coating is to reduce packaging waste, to extend the shelf life of fresh and minimally processed product and protect it from harmful environmental effect by maintaining the transfer of oxygen, carbon dioxide, moisture, aroma and taste compound in a food system. Edible coating may carry functional ingredient such as antioxidants, nutrients and flavor to enhance food stability, quality, functionality and safety. Fruits and vegetables are generally coated by dipping in, brushing or by spraying with edible material so that a semi permeable membrane is produced on the surface by which it suppress the respiration rate, controls moisture loss and provides other function (Raghav *et al.*, 2016)

Edible coating extends the post-harvest life of fresh fruits and vegetables. It is used to improve food appearance and provide safety to the food by its environmental friendly nature. It may be obtained from both animal and vegetable sources. Nature of edible coating may be of protein, lipid, polysaccharide, resin alone or in combination. It acts as a barrier for moisture and gases during processing, handling and storage. It reduces food deterioration and enhances safety by their activity or by incorporation of antimicrobial compound. Other advantages of using edible coating is to reduce packaging waste, to extend the shelf life of fresh and minimally processed product and protect it from harmful environmental effect by maintaining the transfer of oxygen, carbon dioxide, moisture, aroma and taste compound in a food system. According to this review, Edible Coatings extends shelf life, reduce water and moisture loss, delayed ripening process and also prevent microbial growth specifically in fresh fruits and vegetables

2.2.1 Edible coating should

- Not contain any toxic, allergic substance and should be digestible.
- Liable to mechanical damage during handling, display and transportation.
- Have good adhesion property.
- Have good water barrier properties.
- Provide semi permeability to maintain internal equilibrium of gases which is involved during anaerobic and aerobic respiration, thus retarding senescence not affect the nutritional and organoleptic properties of fruit and vegetable.
- Be capable of being used as a carrier for desirable additives such as flavor, nutrients, coloring and vitamin.

- Have antimicrobial and antibacterial properties and be easily manufactured and economically viable.

2.2.2 Requirements from a Coating Material

The characteristics required from an edible coating depend on the specific requirements of the product to be coated, including the primary degradation modes to which it is most susceptible. Fresh and minimally processed fruits have complex requirements concerning packaging systems, since such products are still metabolically active. The main requirements for a fruit coating are described in the following: Moderately low permeability to oxygen and carbon dioxide in order to slow down respiration and overall metabolic activity, retarding ripening and its related changes. On the other hand, the metabolic activity must not be reduced to a degree that creates anaerobic conditions, which promote physiological disorders and accelerate quality loss (Kester and Fennema, 1986; Debeaufort *et al*., 1998). Edible coatings for fruits should control the ripening by reducing oxygen penetration in the fruit rather than by decreasing CO2 and ethylene evaporation rates, that is to say, the CO2/O2 permeability ratio (related to selectivity) should be as high as possible. Proteins and polysaccharide coatings present much higher ratios (from 10 to 25) than those of conventional plastic films (lower than 5.73) (Debeaufort *et al*., 1998). The decreased metabolic activity provided by edible coatings has also been known to retard softening changes (Conforti and Zinck, 2002; Zhou *et al.,* 2011), which result from the loss of turgor pressure and degradation of cell walls, contributing to a decrease in fruit brittleness and firmness (Zhou *et al.,* 2008). The degradation of cell wall structure has been attributed to activity of enzymes such as pectin methyl esterase, cellulase, and polygalacturonase on polysaccharides present in the cell wall (Goulao and Oliveira, 2008). Low water vapor permeability in order to retard desiccation (Garcia and Barret, 2002). In the case of minimally processed fruits, this is especially difficult, since the product surface usually has a very high water activity, which tends to decrease the performance of hydrophilic coatings (Hagenmaier and Shaw, 1992). Sensory inertness or compatibility. Edible coatings were traditionally supposed to be tasteless so would not interfere with the flavor of the product (Contreras Medellin and Labuza, 1981). Alternatively, they may have sensory properties compatible with those of the food. For instance, fruit purees have been studied as film forming edible materials (McHugh *et al.*, 1996; Senesi and McHugh, 2002; Rojas-Graü *et al.*, 2006, 2007a; Azeredo *et al.*, 2009) which can be used as edible coatings for fruits due to the presence of film forming polysaccharides in their compositions. Edible coating for fruits and Vegetables Biopolymer such as lipids, polysaccharide, protein and resin are common coating forming materials that can be used alone or in combination. The functionality of coating is greatly influenced by physical and chemical characteristics of polymer (Sothornvit, 2000). Coating material selection is based on water solubility, hydrophilicity and hydrophobicity nature, ease in formation of coatings and sensory properties.

2.2.3 Edible coating for fruits and Vegetables

Biopolymer such as lipids, polysaccharide, protein and resin are common coating forming materials that can be used alone or in combination. The functionality of coating is greatly influenced by physical and chemical characteristics of polymer (Sothornvit, 2000). Coating material selection is based on water solubility, hydrophilicity and hydrophobicity nature, ease in formation of coatings and sensory properties.

2.2.4 Lipid based coatings

Lipid compounds include neutral lipids of glyceride which are esters of glycerol and fatty acid and waxes which are esters of long chain monohydric alcohols and fatty acids. Resins are a group of acidic substances that are secreted by special plant cell into long resin duets or canals in response to injury or infection in many trees and shrubs (Debeaufort *et al.*, 1998). Edible lipids are neutral lipids, waxes and resin which are traditional coating material for fresh produce and provide effective moisture barrier property and also improve surface appearance (Hernandez, 1994; Morillon, 2002).

2.2.5 Polysaccharide based coating

Polysaccharide coating have been produced from starch and starch derivatives, cellulose derivative, alginates, carrageenan, various plant and microbial gums, chitosan and pectinates (Nisperos, 1990). These coatings reduce respiration rate of fruits and vegetables (Motlagh, 1988). Due to its hydrophilic nature it has high gas barrier property.

(a) Starch and its derivative

Starch is a natural polysaccharide used for food hydrocolloid (Whistler 1965) because of its high functionality and relatively low cost. Starch film is generally transparent, odourless, tasteless and colorless and has low permeability to O_2. Dextrin is derived from starch and has smaller molecular size and used in coating formation. Dextrin coating has better resistance to water vapour as compared to starch coating (Allen, 1963). Pullulan is an extracellular polysaccharide of starch which is edible and biodegradable. Pullulan films are colourless, odourless, and tasteless and show high O_2 barrier properties. Pullulan coating has been applied for preserving strawberries and kiwi fruits (Diab, 2001). Starch coating is effective for fruits and vegetables because they have high respiration rate.

(b) Cellulose and its derivative

Cellulose is a structural material of plant cell walls (Nisperos, 1994). Cellulose derivative shows excellent film forming property but they are too expensive for large scale application. Most common cellulose derivatives are carboxymethyl cellulose (CMC), methyl cellulose (MC), hydroxypropyl cellulose (HPC) and hydroxypropyl methyl cellulose (HPMC). These are water soluble polysaccharide, non-ionic and compatible with surfactants.

(c) Seaweed extracts

Alginates are the major structural polysaccharides of brown seaweed, also known as Phaeophyceae. Alginate consists of good film forming property, transparent and soluble in water. Alginates have low permeability to oil and fats but have high permeability to water vapour (Valero *et al.*, 2013). It also act as a sacrificing agent. Alginates have good adhesion property. Calcium alginate coatings enhance the quality of fruits and vegetables by retarding shrinkage, oxidative rancidity, moisture migration and oil absorption. It reduces weight loss and improves appearance and colour (Hershko, 1998). Carrageenan is extracted from several red seaweeds mainly Chrondruscrispus (Whistler, 1985) and it is complex mixture of polysaccharide. Carrageenan based coating reduces moisture loss and oxidation of apple slices (Lee et al., 2003). It inhibits microbial growth. It reduces moisture loss from grape fruit (Bryan, 1972).

(d) Chitosan

It is a linear polymer of 2-amino-2-deoxy-βD-glucan, is a deacetylated form of chitin. It is a naturally occurring cationic biopolymer Bemiller (1965), Davis (1988). It is a shell component of crab and shrimp, skeletal substances of invertebrates and cell wall constituent of fungi and insects (Raghav *et al.,* 2016). Chitosan is one of the best coating material for fresh produce because of its excellent film forming properties, antimicrobial activity and its compatibility with other substances such as minerals, vitamins and antimicrobial agents (Li, 1992; Shahidi ,1999). Chitosan based coating delay ripening and decreased respiration rate of fruits and vegetables and retard weight loss, colour wilting and fungal infection in cucumber and tomatoes (EI Ghaouth, 1991).

(e) Aloe vera

Aloe vera is a tropical and subtropical plant. Aloe vera contains medicinal and therapeutic properties and has been used for centuries (Eshun, 2004). Aloe vera gel is used as edible coating for fruit and vegetables. It has antifungal properties (Saks, 1995). Aloe vera gel based edible coatings prevent moisture loss and retains firmness, decreases respiration rate, delays oxidative browning and reduces the growth of microorganisms in table grapes.

2.2.6 Protein based edible coating

Edible coating can be made from animal protein (such as milk protein) and plant protein (such as zein, soy protein and wheat protein). They show excellent oxygen, carbon dioxide and lipid barrier properties particularly at low RH (Lin et al., 2007). Protein based edible coatings are brittle and susceptible to cracking. Protein based edible coating shows poor water barrier properties (Mohamoud and Savello, 1992).

(a) Plant origin

Zein and soy protein are the two main plant origin protein and they are used as edible coating for fruits and vegetables. Zein is the storage protein of corn and comprises of 45 to 50% of the protein in corn. Soy protein concentrate (SPC) or Soy protein Isolate (SPI) is produced from defatted protein and contain 65 to 72% and 90% protein respectively (Mounts ,1987). Soy protein based edible coating shows bad resistance to moisture because of its hydrophilic characteristics. Soy protein based coating exhibits good barrier properties to O_2 at low RH.

(b) Animal origin

Whey protein and Casein are the main milk protein. They are used as edible coating for fruit and vegetables. Casein comprises 80% of the total milk protein (Chen, 1995). Casein films are transparent, flavorless, and flexible and are attractive for food applications. Whey proteins comprise 20% of the total milk protein. Whey proteins are purified to produce whey protein concentrate. It can have 25 to 80% protein content or whey protein isolate has 90% protein content. Whey protein films show good oxygen barrier properties in low and intermediate relative humidity.

2.2.7 Commercial application of edible coating

The impact of cellulose edible coating having different pH on lightly processed carrot was studied by Li and Barth (1998). Their study showed that lower pH EC had highest CO_2 and lowest O_2 concentration. A study was conducted by Baldwin *et al*. (1999) to determine the impact of edible coating produced from polysaccharide and carnauba wax on mango fruit. Their results indicated that polysaccharide was less permeable to respiratory gas such as O_2. Both coatings delayed ripening and improved appearance. Therefore it can be concluded that shelf life was extended by both coatings. Water loss was also comparatively reduced by carnauba wax and polysaccharide coatings.

Ascorbic acid (AA) is widely used as a food ingredient because of its reducing and antioxidant properties in addition to its function as an essential nutrient. Ascorbic acid has been shown to effectively scavenge superoxide, hydrogen peroxide, hypochlorite, the hydroxyl radical, peroxyl radical, and singlet oxygen. Controlling lipid oxidation in peanuts using WPI coatings has been reported. However, no study has been reported about antioxidant effects of an AA-incorporated WPI coating (AA-WPI coating) on foods.

Roasted peanuts are highly susceptible to lipid oxidation because of a high content of polyunsaturated fatty acids. Lipid oxidation in peanuts makes them unacceptable because of rancidity formation, which is a concern for the shelf life stability of many confections containing peanuts. Peanuts have been used as a model food for studying effects of whey protein coatings on lipid oxidation of foods.

Chapter -III

MATERIALS AND METHODS

The present investigation entitled, “Enhancement of shelf life of pomegranate arils using edible coating” was carried out in the Dairy Science and Food Technology, Institute of Agriculture Science, Banaras Hindu University, Varanasi, Uttar Pradesh, India. During the investigation the work was planned to extend the shelf life of pomegranate arils by improving the overall retention of physiochemical and microbiological properties.

3.1 Plant material and Processing

Fresh pomegranate (Arakta Variety) used for this study was procured from Varanasi, India .Fruit were transported to the laboratory within 30 min of the procurement and the pomegranate was graded for their uniformity in size and shape . Selected pomegranate were first washed with running water and then cut to remove upper hard layer of pomegranate and after that remove arils properly. Pomegranate arils treated with 50 ppm sodium hypochl oride solution for half an hour for surface disinfection. The samples were then dried at room temperature.

All the chemicals such as sodium alginate, Sodium bicarbonate, Glacial Acetic acid etc. were purchased from Himedia, Mumbai, India. Glass wares and metal wares were available in the laboratory of food Dairy Science and Food Technology related to project. Various instruments that have been used in this project are enlisted in TABLE 3.2.

Table 3.2- List of the instruments with its company, model and country name

Name of Instrument/Equipment	**Company, model and country**
Electric weighing balance	Metler Toledo,JB1603-C/Fact, Switzerland
pH meter	Termo Scientific, Sn B21899,Singapure
Refractometer	Bellingham and Stanley, UK
Spectrophotometer	Shimadzu, Japan
Laminar air flow	Labtech LCB 120V,Daihan Pvt, Ltd, India
Centrifuge machine	Sigma,3-30K,Germany
Autoclave	Tomy Digital, UK
Incubator	Remi, India
Colorimeter	Hunter Lab, Color Quest XE

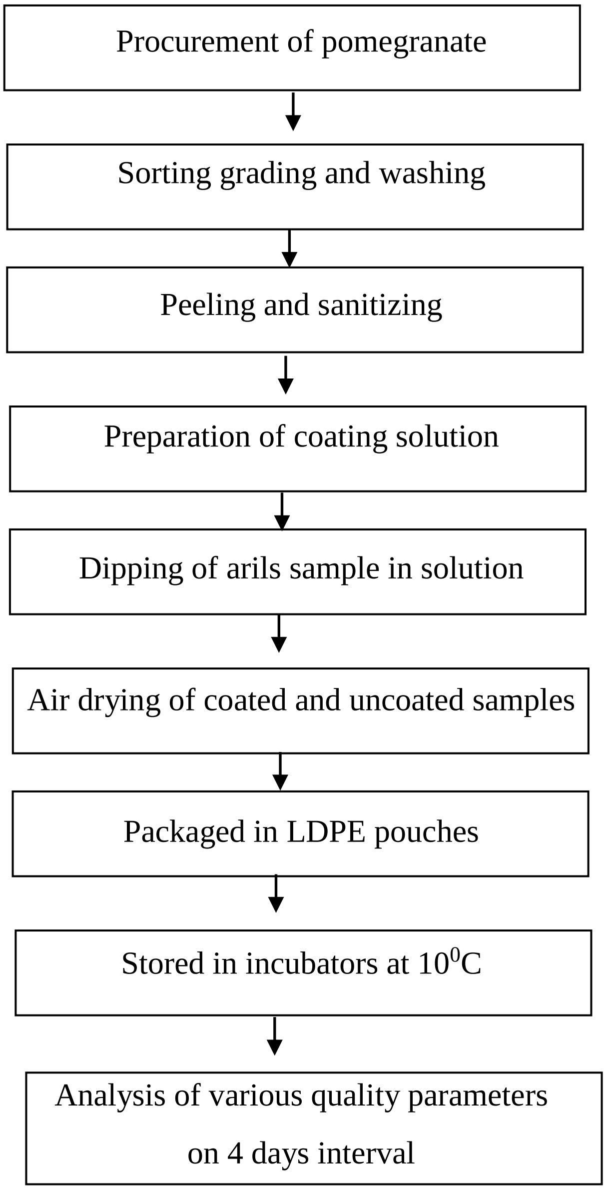
3.3 Coating solution

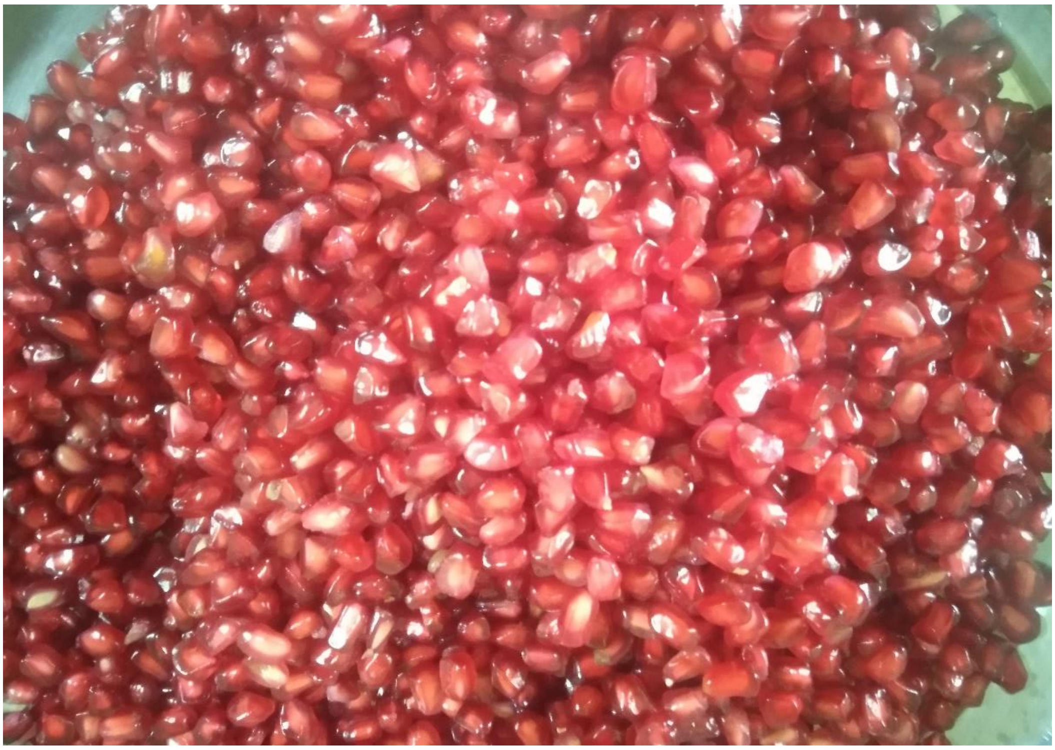
To prepare alginate solution, known quantity of sodium alginate powder was weighted using citizen balance and solution was prepared by dissolving alginate powder in aqueous solution of 0.5 % acetic acid. The resulting mixture was stirred continuously at room temperature (25 ^{0}C) until alginate was dissolved. Coatings were applied by immersion of arils in the alginate solution for 5 min, depending on treatments whereas the control samples were washed with distilled water. The arils were then strained out and kept on the blotting paper to remove the excess moisture at room temperature. Then arils were packed in LDPE pouches and kept in refrigerated storage at 10^0 C. Chemical parameters viz. TSS (%) total sugars (%), reducing sugar (%), non-reducing sugars (%), titratable acidity (%), anthocyanin content (%), Ascorbic acid (mg/100g), Total phenolic content (mg gallic acid/100gm) as well as organoleptic characters of treated arils were evaluated during the investigation at 4th, 8th, 12th and 16th day of storage. Data obtained on various characters were analyzed statistically according to the analysis of variance techniques for CRD (Fisher 1950).

Table 3.4: Concentration of six different treatments

Treatments	Concentrations
T_1	1% sodium alginate
T_2	1% sodium alginate + 1% Ascorbic acid
T_3	1.5% sodium alginate
T_4	1.5 sodium alginate + 1% Ascorbic acid
T_5	2% sodium alginate
T_6	2% sodium alginate + 1% Ascorbic acid
T_7	Untreated sample

Flow diagram showing the sodium alginate coating of pomegranate arils





Coated Pomegranate arils

Coated Pomegranate arils

Coated Pomegranate arils packed in LDPE pouches

Coated Pomegranate arils packed in LDPE pouches

3.5 Physicochemical Analysis of Pomegranate arils

3.5.1 Total soluble solids (TSS)

TSS measurement of pomegranate arils juice was done with the help of hand refractometer using the method as recommended by (**Srivastava and Kumar, 2004**). Sample was placed on the prism looking through the eye piece with the projection inlet facing towards light. The point on the scale was noted where the boundary line of the shaded area intersects the unshaded area, total soluble solids indicated by degree Brix was directly read from refractometer and calculated with the following equation-

TSS (^{0}Brix)=Refractometer reading

3.5.2 Titratable acidity

Acidity of various samples were determined using the method as recommended by **(Ranganna, 2001)**.5ml sample of pomegranate ails juice was dissolve in a500 ml of distilled water and out of this 20 ml aliquot was taken and titrated with 0.1 N NaOH using a few drops of phenolphthalein solution as indicator. The end point was judged by the appearance of pink colour. The titer value was noted and result was calculated as percent total acids using the following equation.

$$\textbf{Acidity (\%)} = \frac{VOL.\ of\ NaOH\ used\ (ml) X\ Normal\ of\ NaOHX\ Equi.wt.of\ citric\ acidX 1000}{Vol.of\ sample\ (ml) titred}$$

3.5.3 Total sugar

Sulfuric Acid-UV Method

Procedure

1. One ml aliquot of carbohydrate solution is rapidly mixed with 3ml of concentrated H_2SO_4.
2. The temperature of the mixture rises rapidly within 10-15 second after addition acid.
3. The solution was cooled in ice for 2min to bring to room temperature.
4. Finally UV light absorption at 315ppm.

3.5.3.1. Reducing sugar

Principle-

The DNS (Dinitro salicylic acid) method for estimating the ion concentration of reducing sugar in a sample was originally inketed by **(G. Miller, 1959)**. Reducing sugar has the property to reduce many of reagent. A reducing sugar in one that in a basic solution forms an aldehyde or ketone. The aldehyde group of glucose convert 3, 5 dinitro salicylic acid (DNS) to 3-amino-5 nitro salicylic acid when is reduced from DNS water is used up a reactant and oxygen gas is released during the reaction

The formation of 3-amino-5nitro salicylic acid result in a change in the amount of light absorbent wave length 540 nm. The absorbance measured using a spectrophotometer is directly proportional to the amount of reducing sugar.

Material

Sodium Potassium Tartrate

Dissolve 45gm of NaOH tartrate in 75ml of H_2O.

3, 5 DNS Solution

Dissolve 1.5gm of DNS reagent in 30ml of 2ml/liter NaOH.

2 molar NaOH

80gm of NaOH dissolved in 1 liter of H_2O.

DNS reagent

Prepare fresh by mixing the reagent (1) and (2) make the Volume to 150ml with a water standard sugar sodium.

Standard Sugar Sodium-

(a) Stock the standard sugar sodium 250mg of glucose in water and make up the volume to 100ml.

(b) **Working standard sodium-**Take 10ml from this stock solution and make up the volume to 100ml.

Procedure

1. Seven clean and dry test tubes were taken.
2. Pipette out the standard solution in the range of to 2ml (0,0.5,1.5,2.0).
3. Make the final volume in all the tube to 2ml with distilled water concentration ranging from 0 to 750mg.
4. 1 ml DNS reagent was added to all the test tube and mix will and capped it with cotton (To avoid the loss of liquid to Due to evaporation)
5. Test tubes were kept in boiling water bath for 5-10 minute (temperature 100^0C)
6. Taken the tube and cool to room temperature read extinction at **540nm** against the blank.
7. Prepare the standard curve of the sugar provided and use them to estimate the concentration of Unknown provided.

Result-The 100ml of unknown solution contain-------mg of glucose.

3.5.3.2 Non-Reducing sugar

Non-Reducing Sugar=Total Sugar-Reducing Sugar

3.5.4 Estimation of total anthocyanin content

Principle of the assay:

The differential assay method for anthocyanin is based on measuring the absorbance at two different pH values. It is based on the structural transformations of the anthocyanin chromophore as a function of ph. In order to determine the total monomeric anthocyanin Content the absorbance at pH 1.0 and 4.5 is measured at 700nm.

Assay procedure

Total monomeric anthocyanin content was quantified using a pH differential method described by Giusti and Wrolstad (2001). Samples were diluted in two buffer solutions: potassium chloride buffer 0.025 M (pH 1.0) and sodium acetate buffer 0.4 M (pH 4.5) and then the absorbance was measured simultaneously at 516 nm and 700 nm (to correct for haze) after 15 minutes of incubation at room temperature. Absorbance readings were made at room temperature against distilled water as blank. A Jasco V 530 UV-Vis spectrophotometer was used for measurements. The monomeric anthocyanin pigment concentration was calculated according to the following equation: C (mg/1) = $\frac{A\ XM\ WXD\ FX1000}{SXL}$

Where: A= (A516—A700) pH 1.0—(A516—A700) pH 4.5, MW is the molecular weight, *DF* is the dilution factor, c is the molar absorbance and / is the path length (1 cm). The total monomeric anthocyanins content was expressed as cyanidin-3-glucoside equivalents (MW=449.2 and = s 26900). Each sample was analyzed in triplicate and the results were expressed as the average of the two measurements.

3.5.5 Ascorbic acid

To evaluate the ascorbic acid, 2 g of sample was homogenized in 10 ml of (3% metaphosphoric acid for 10 min. It was determined by titration of 10 ml filtrated sample by 2, 6-dichlorophenolindophenol (DCPIP) and expressed as mg ascorbic acid/100 g sample as per FAO (1991).

3.5.6. Total Phenolic Content

The total phenolic content was determined by the Folin-Ciocalteau method **(Singleton and Rossi, 1965)** and **(Kaur and Kapoor, 2002).**Gallic acid was used for generating the standard curve having concentration 20-100mg/ml. 2.5 ml of 10 times dilute FC reagent was added to each test tube and mix well for 1 min and 2ml of 20 % sodium carbonate (Na_2CO_3) was added to it and allowed to incubate for 30 min at 37 ^{0}C and further the absorbance was measured at 725 nm and standard graph was plotted. The sample was also incubated at 37 ^{0}C for 30 mins and the absorbance was recorded at 725 nm. The total Phenolic content equivalent to gallic acid was determined from standard graph. 2 gm samples were homogenized in 15 ml of 80 % (v/v) aqueous ethanol at room temperature and centrifuged in cold condition at 10,000 rpm for 15 minutes at 4^{0}C and the supernatant was extracted. The residue obtained was re-extracted twice and the supernatant was poured into petridishes and evaporated to dryness at room temperature. Residue was dissolved in 5 ml of distilled water**.** 100 micro gram of this extract was diluted to 3 ml of water and 0.02 ml of folin-Ciocalteau reagent was added. after 3 minutes, 2ml of 20% sodium carbonate was added and contents were mixed thoroughly and blue was developed. The absorbance was measured at 725 nm in UV-spectrophotometer (Shizmadzu, Japan) using gallic acid as standard. The results were expressed as mg gallic acid/ 100 g fresh material.

3.5.7 Microbial analysis

Total yeast and mold, total plate and coliform counts were monitored at an interval 4, 8, 12 and 16th day to know the overall efficiency of the edible coating .Yeast and mold counts were done using potato dextrose agar as per the procedure given by ISO (1987), after incubating at 37 ^{0}C for 48 h. Total coliform count was analyzed by using violet red blue agar after incubating at 37 ^{0}C for 24 h as per Apha, (1995). Total plate counts were determined by plate count agar (PCA, Merck) followed by the pour plate method with an incubation temperature of 7 ^{0}C for 10 day and colonies were subsequently counted and results were analyzed.

3.5.8 Sensory Analysis of pomegranate arils

Determination of sensory qualities

Sensory quality attributes *viz.* colour and appearance, consistency, flavor, taste and overall acceptability of the samples were evaluated using 9 point Hedonic rating test method as recommended by **Ranganna (2001)**.This test measures the consumer's acceptability. The detailed procedure is given below. A semi trained panel consisting of 10 judges of different age groups having different eating habits were constituted to evaluate the quality. The judge were selected from the faculty members and staff of the Dairy Science and Food Technology, Department of Horticulture, B.H.U (Varanasi). The judgments were quantified by appropriate analysis for determing the significance of variation of average scores and the contribution of the individual quality characteristics to the overall quality. Samples were served to the panelists and they were asked to rate the acceptability of the product through sense organs. Different attributes viz. color and appearance, consistency, flavor and taste were rated on the basis of 9 point hedonic scale ranging from 1 to 9 **(Appendix I).**A score card was supplied to each panel member at the time of evaluation.

The treated and untreated pomegranate arils samples were subjected to sensory evaluation by nine semi-trained descriptive expert panel. The expert panel evaluated Color, Flavor and overall acceptability. The room temperature of the environment where sensory evaluation was performed was kept at 25^{0}C.The expert panel also evaluated the color of the pomegranate arils on a 9 point hedonic scale (where,1= Most dislike and 9=Most like).

Chapter-IV

RESULTS AND DISCUSSION

4.1 Total soluble solids (TSS, ^{0}Brix)

TSS is an index of soluble sugars present in fruits and vegetables. The TSS content of the samples were determined by using the refractometer (Bellingham and Stanley, UK) and the results was expressed as percentage by weight. Samples were thoroughly blended in a blender and were further filtered through a whatman filter paper. A few drops of the clear homogenate was taken on the prism of the refractometer and direct reading was observed on the screen as per the method of AOAC (1994)

The mean data pertaining influence of different treatments of edible coating on the changes in TSS of pomegranate arils was recorded from 4th to 16th day of storage and is presented in Table 4.1. The highest (15.63) TSS (^{0}Brix) on the 4th day of observation was found in T_6 (2 % Sodium alginate and 1 % Ascorbic Acid) while, on other days of observations i.e. 8th, 12th and 16th day (14.83, 14.27 and 7.93) the highest TSS was recorded in T_5 (2% Sodium alginate). Lowest TSS on the respective days 4th, 8th, 12th, and 16th day (14.43, 13.80, 14.27 and 6.10) were noted in T_3 (1.5%Sodium alginate), T_2 (1% Sodium alginate and 1% Ascorbic Acid), T_6 (2 % Sodium alginate and 1 % Ascorbic Acid) and T_4 (1.5 % Sodium alginate and 1 % Ascorbic Acid). TSS in the coated arils generally decreased and it was found that alginate treatments played a positive role in maintaining TSS of arils during storage period. Arils treated with 2 % Sodium alginate and 1 % Ascorbic acid recorded high TSS compared to other treatments on 7th day of storage whereas treating arils with 2 % Sodium alginate recorded significantly higher TSS during whole storage period except on 4th day. Initially the decrease in TSS during the storage was slow i.e. upto 12th day but after that it became fast after 12th day and on 16th day marked decrease was found in TSS of the arils. This may due to rapid utilization of reducing sugar and other organic metabolites.

Table 4.1-Effect of edible coating on changes in TSS (^{0}Brix) of pomegranate arils Treatments

Treatments	Treatment Details	Storage period			
		4th Day	8th Day	12th Day	16th Day
T_1	1% Sodium alginate	15.43	14.47	13.22	7.33
T_2	1% Sodium alginate + 1 % Ascorbic acid	14.57	13.80	13.20	7.17
T_3	1.5 Sodium alginate	14.43	13.97	13.27	7.33
T_4	1.5 Sodium alginate + 1 % Ascorbic acid	15.37	14.17	13.53	6.10
T_5	2 % Sodium alginate	15.13	14.83	14.27	7.93
T_6	2% Sodium alginate + 1 % Ascorbic acid	15.63	14.40	13.06	7.47
T_7	Control	15.47	14.70	13.83	6.40
CD		**0.30**	**0.21**	**0.24**	**0.18**
SEM (±)		**0.10**	**0.07**	**0.08**	**0.06**

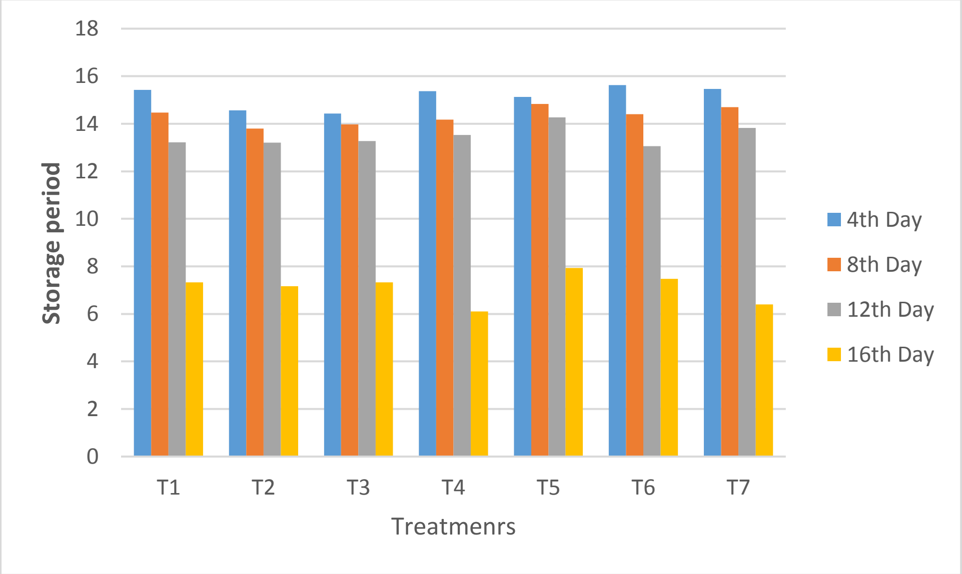


Figure 4.1 - Effect of edible coating on changes in TSS (^{0}Brix) of pomegranate arils Treatments

4.2 Total Sugars (%)

Perusal of the data on total sugars (%) is reported statistically significant and is demonstrated in Table 4.2. It was observed that treatment T_5 (2 % Sodium alginate) maintained higher amounts of total sugars (%) during the whole storage period i.e. 7.97, 7.43, 6.05 and 3.13 on 4th, 8th, 12th, and 16th day respectively. The minimum total sugar (%) (6.45) was recorded in T_4 (1.5 Sodium alginate and 1 % Ascorbic Acid) on 4th day. However, treatments T_4 (1.5 Sodium alginate and 1 % Ascorbic Acid) and T_7 (Control) were found with lowest total sugars (%) (6.24) on 8th day whereas on 12th and 16th day minimum total sugars (%) (3.92 and 1.77) were found in T_7 (Control) and T_6 (2 % Sodium alginate and 1 % Ascorbic Acid) respectively. The decrease in total sugars from initial to final storage period may be possible due to coating of sodium alginate; the conversion of starch into sugars as well as biosynthesis of sucrose slowed down as a result of modified gaseous exchange and reduced respiration rates and reflected as lower total sugars content.

Table 4.2 - Effect of edible coating on total sugar (%) of pomegranate arils Treatments

Treatments	**Treatment details**	**Storage period**			
		4th Day	**8th Day**	**12th Day**	**16th Day**
T_1	**1% Sodium alginate**	7.13	6.90	4.35	2.77
T_2	**1% Sodium alginate + 1 % Ascorbic acid**	6.65	6.45	4.55	2.05
T_3	**1.5 Sodium alginate**	7.43	7.14	5.55	2.56
T_4	**1.5 Sodium alginate + 1 % Ascorbic acid**	6.45	6.24	5.88	2.35
T_5	**2 %Sodium alginate**	7.97	7.43	6.05	3.13
T_6	**1 % Sodium alginate+ 1 % Ascorbic acid**	7.13	6.45	5.71	1.77
T_7	**Control**	6.88	6.24	3.92	2.08
CD		**0.03**	**0.03**	**0.02**	**0.08**
SEM (±)		**0.01**	**0.01**	**0.01**	**0.03**

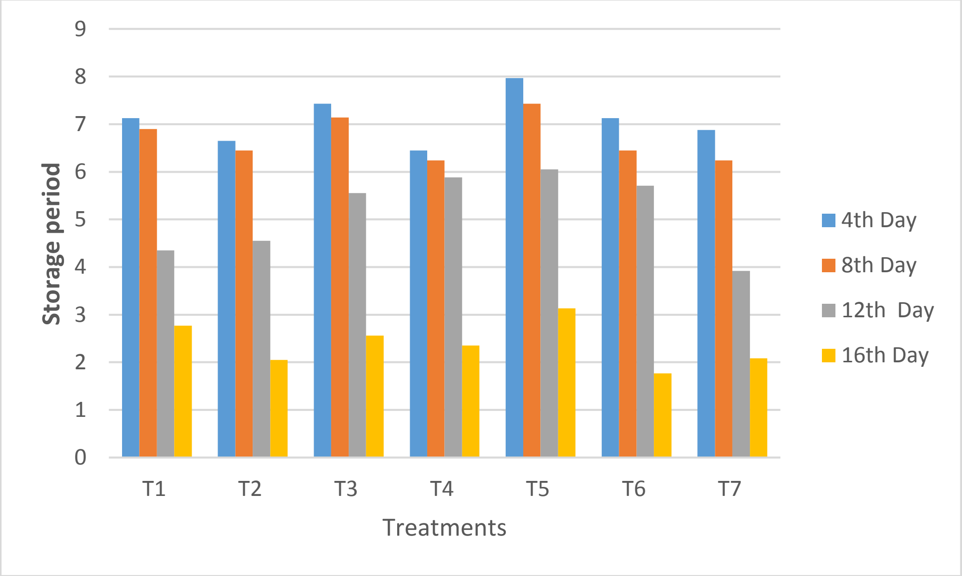


Figure 4.2 - Effect of edible coating on total sugar (%) of pomegranate arils Treatments

4.3 Reducing Sugars (%)

The statistical observation of data explaining the changes in reducing sugars of treated arils by different treatment has been observed significant which is showed in Table 4.3. Reducing sugar (%) which is a sweetness factor was recorded highest (6.88, 6.46, 5.54 and 2.93) in T_5 (2 % Sodium alginate) than all other treatments during storage on all observation days. Moreover, the lowest reducing sugars (%) were recorded as 6.05, 5.55 and 3.52 in treatments T_7 (Control) on 4th, 8th, and 12th day whereas it was found lowest in T_6 (2 % Sodium alginate) and 1 % Ascorbic Acid) on the 16th day.

Table 4.3 -Effect of edible coating on reducing sugar (%) of pomegranate arils Treatments

Treatments	Treatment details	Storage period			
		4th Day	8th Day	12th Day	16th Day
T_1	1% Sodium alginate	6.65	6.45	4.17	2.52
T_2	1% Sodium alginate + 1 % Ascorbic acid	6.24	6.06	4.25	1.65
T_3	1.5 Sodium alginate	5.73	6.45	5.13	2.25
T_4	1.5 Sodium alginate + 1 % Ascorbic acid	6.88	5.87	5.42	1.79
T_5	2 % Sodium alginate	6.88	6.46	5.54	2.93
T_6	2% Sodium alginate + 1 % Ascorbic acid	6.25	5.74	5.26	1.43
T_7	Control	6.05	5.55	3.52	1.86
CD		**0.02**	**0.03**	**0.02**	**0.04**
SEM (±)		**0.01**	**0.01**	**0.01**	**0.01**

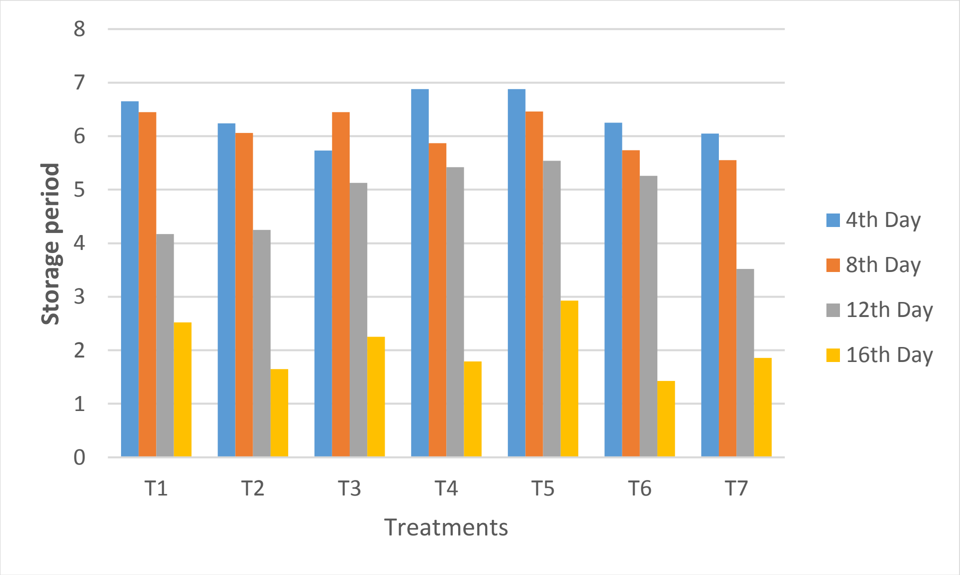


Figure 4.3 -Effect of edible coating on reducing sugar (%) of pomegranate arils Treatments

4.4 Non-reducing sugars (%)

It is evident from Table 4.4 that there were significant differences in non-reducing sugars of treated pomegranate arils due to different treatments. Non reducing Sugars (%) were found to high in T_5 (2 % Sodium alginate) during storage period except at 16th day on which it was recorded higher in T_2 (1 % Sodium alginate and 1% Ascorbic Acid).The minimum value varied as the storage period proceeded and treatment T_2 (1 % Sodium alginate and 1% Ascorbic Acid) recorded minimum value on 4th day, treatment T_4 (1.5 % Sodium alginate and 1 % Ascorbic Acid) recorded minimum value on 8th day whereas on 12th and 16th day it was noted in T_1 (1 % Sodium alginate), and T_2 (2 % Sodium alginate and 1% Ascorbic Acid). As proven that coating reduces respiration rates by modification in exchange of O_2 and CO_2 in fruits. The lower level of non-reducing sugar (sucrose) coated arils might be due to its specific capacity to altered respiration process resulted in slow hydrolysis of starch as well as inadequate biosynthesis of sucrose during ripening.

Table 4.4 - Effect of edible coating on non-reducing sugar (%) of pomegranate arils Treatments

Treatments	Treatment details	Storage period			
		4th Day	8th Day	12th Day	16th Day
T_1	1% Sodium alginate	0.48	0.44	0.18	0.25
T_2	1% Sodium alginate + 1 % Ascorbic acid	0.41	0.40	0.26	0.40
T_3	1.5 Sodium alginate	0.97	0.69	0.42	0.31
T_4	1.5 Sodium alginate + 1 % Ascorbic acid	0.72	0.38	0.46	0.57
T_5	2 % Sodium alginate	1.09	0.97	0.51	0.23
T_6	2% Sodium alginate + 1 % Ascorbic acid	0.88	0.72	0.45	0.33
T_7	Control	0.83	0.68	0.40	0.22
CD		**0.04**	**0.04**	**0.04**	**0.09**
SEM (±)		**0.01**	**0.01**	**0.01**	**0.03**

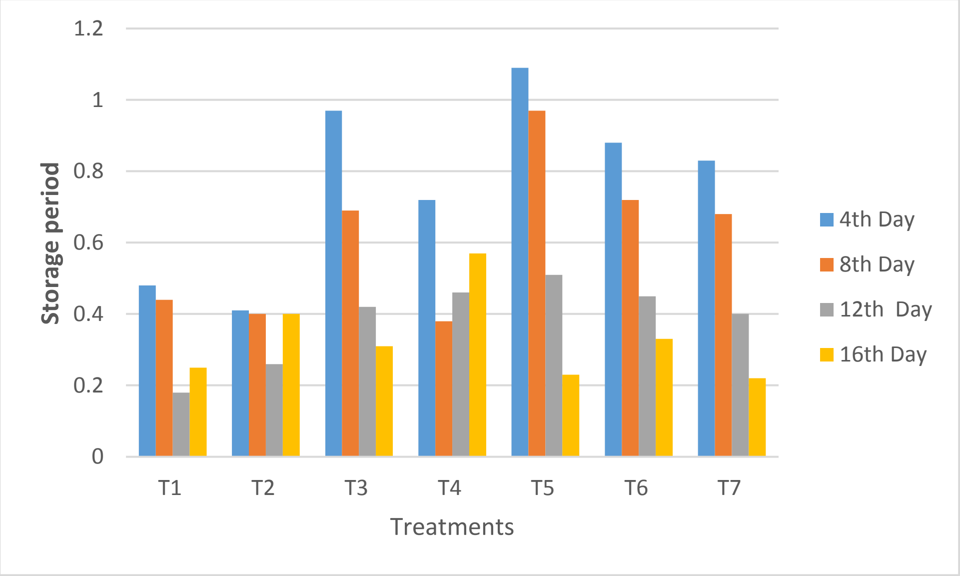


Figure 4.4 - Effect of edible coating on non-reducing sugar (%) of pomegranate arils Treatments

4.5 Titratable Acidity (%)

In the present research the data collected regarding the changes in titratable acidity (%) of treated arils of pomegranate has been found non-significant and is presented in Table 4.5. Least changes were noticed in titratable acidity (%) of treated arils. Treatments T_5 (2 % Sodium alginate), T_6 (2 % Sodium alginate and 1 % Ascorbic Acid) and T_7 (Control) were found with less acidity (%) upto 12th day as compared to other treatments. Moreover, acidity (%) of T_1 (1 % Sodium alginate), T_2 (1 % Sodium alginate and 1% Ascorbic Acid), T_3 (1.5 % Sodium alginate) and T_4 (1.5 % Sodium alginate and 1 % Ascorbic Acid) remained unchanged after 8th day and on 16th day these were found in line with remaining treatments. As revealed from the data slight variations in the titratable acidity were recorded in the treated arils during the storage.

Table 4.5 - Effect of edible coating on titratable acidity (%) of pomegranate arils Treatments

Treatments	Treatment details	Storage period			
		4th Day	8th Day	12th Day	16th Day
T_1	1% Sodium alginate	0.02	0.03	0.03	0.03
T_2	1% Sodium alginate + 1 % Ascorbic acid	0.02	0.03	0.03	0.03
T_3	1.5 Sodium alginate	0.02	0.03	0.03	0.03
T_4	1.5 Sodium alginate + 1 % Ascorbic acid	0.02	0.03	0.03	0.03
T_5	2 % Sodium alginate	0.02	0.02	0.02	0.03
T_6	2% Sodium alginate + 1 % Ascorbic acid	0.02	0.02	0.02	0.03
T_7	Control	0.02	0.02	0.02	0.03
CD		NS	NS	NS	NS
SEM (±)		NS	NS	NS	NS

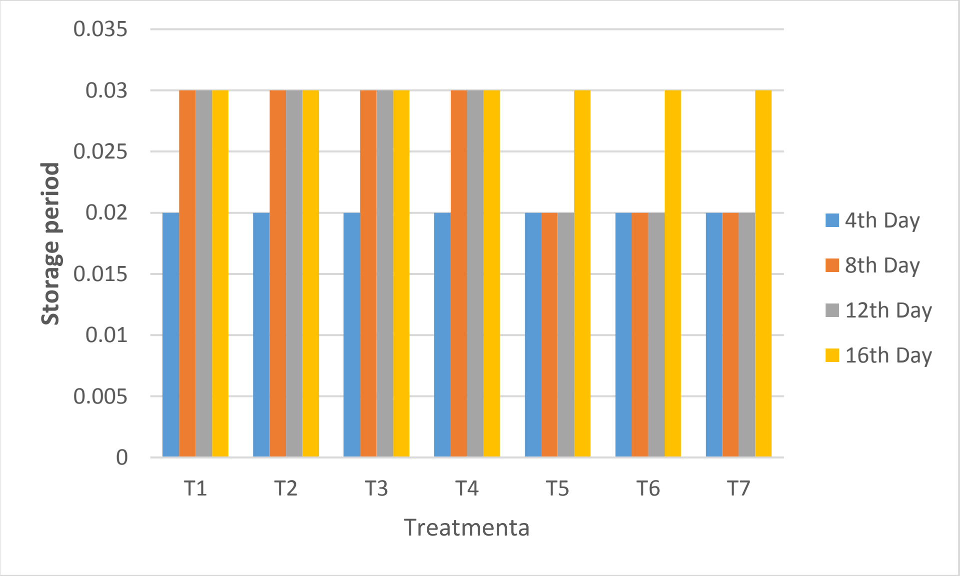


Figure 4.5 - Effect of edible coating on titratable acidity (%) of pomegranate arils Treatments

4.6 Anthocyanin content (mg/100 g)

The perusal of the observations on changes in anthocyanin content (mg/ 100) of the treated arils is found statistically significant and is demonstrated in Table 4.6. Anthocyanin content (mg/100g) was found high (12.92, 11.38, 9.29 and 5.21) in treatment T_5 (2 % Sodium alginate) during storage period on all observation days however, variations were recorded in lowest anthocyanin content (mg/100g) and it was recorded minimum (11.33 and 9.62) in T_7 (Control) on 4th and 8th day while at 12th day lowest (6.68) anthocyanin content (mg/100g) was recorded in T_3 (1.5 % Sodium alginate) and on 16th day T_7 (Control) was found with less anthocyanin content (mg/100g) (4.01) compared to other treatments. The arils treated with two per cent sodium alginate were found better in anthocyanin keeping properties compared to other treatments.

Table 4.6 - Effect of edible coating on anthocyanin content (mg/100 g) of pomegranate arils Treatments

Treatments	Treatment details	Storage period			
		4th Day	8th Day	12th Day	16th Day
T_1	**1% Sodium alginate**	12.01	10.32	8.32	4.52
T_2	**1% Sodium alginate + 1 % Ascorbic acid**	12.13	10.27	8.10	4.34
T_3	**1.5 Sodium alginate**	11.85	10.02	6.68	4.44
T_4	**1.5 Sodium alginate + 1 % Ascorbic acid**	12.24	10.46	6.96	4.99
T_5	**2 % Sodium alginate**	12.92	11.38	9.29	5.21
T_6	**2% Sodium alginate + 1 % Ascorbic acid**	11.67	10.11	9.29	4.60
T_7	**Control**	11.33	9.62	7.11	4.01
CD		**0.21**	**0.13**	**0.22**	**0.11**
SEM (±)		**0.06**	**0.05**	**0.07**	**0.04**

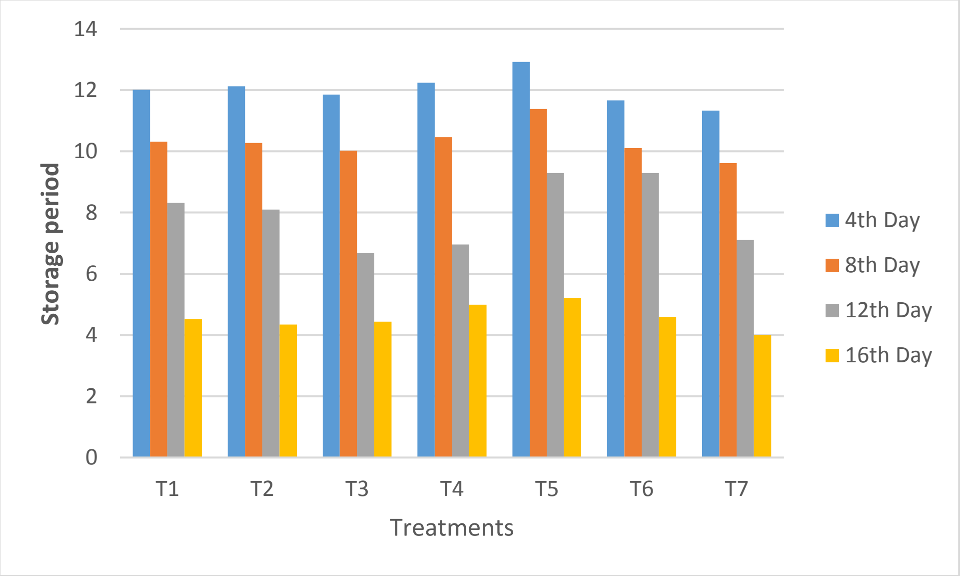


Figure 4.6 - Effect of edible coating on anthocyanin content (mg/100 g) of pomegranate arils Treatments

4.7 Ascorbic acid (mg/ 100g)

The perusal of the observations on changes in ascorbic acid content (mg/ 100g) of the treated arils is found statistically significant and is demonstrated in Table 4.7. Ascorbic acid content (mg/100g) was found high (8.54,8.45,8.09 and 7.99) in treatment T_5 (2 % Sodium alginate) during period of observation days however, variations were recorded in lowest ascorbic content (mg/100g) and it was recorded minimum (8.50 and 8.19) in T_7 (Control) on 4th and 8th day while at 12th day lowest (7.61) ascorbic acid content (mg/100g) was recorded in T_3 (1.5 % Sodium alginate) and on 16th day T_7 (Control) was found with less ascorbic acid content (mg/100g) (7.31) compared to other treatments. The arils treated with two per cent sodium alginate were found better in ascorbic acid keeping properties compared to other treatments.

Table 4.7 - Effect of edible coating on ascorbic acid (mg/ 100g) of pomegranate arils Treatments

Treatments	Treatment details	Storage period			
		4th Day	8th Day	12th Day	16th Day
T_1	1% Sodium alginate	8.53	8.28	7.63	7.32
T_2	1% Sodium alginate + 1 % Ascorbic acid	8.51	8.38	8.02	7.89
T_3	1.5 Sodium alginate	8.53	8.35	7.61	7.35
T_4	1.5 Sodium alginate + 1 % Ascorbic acid	8.52	8.33	7.97	7.55
T_5	2 % Sodium alginate	8.54	8.45	8.09	7.99
T_6	2% Sodium alginate + 1 % Ascorbic acid	8.51	8.41	8.05	7.54
T_7	Control	8.50	8.19	7.89	7.31

Standard deviation ±0.05

n=10

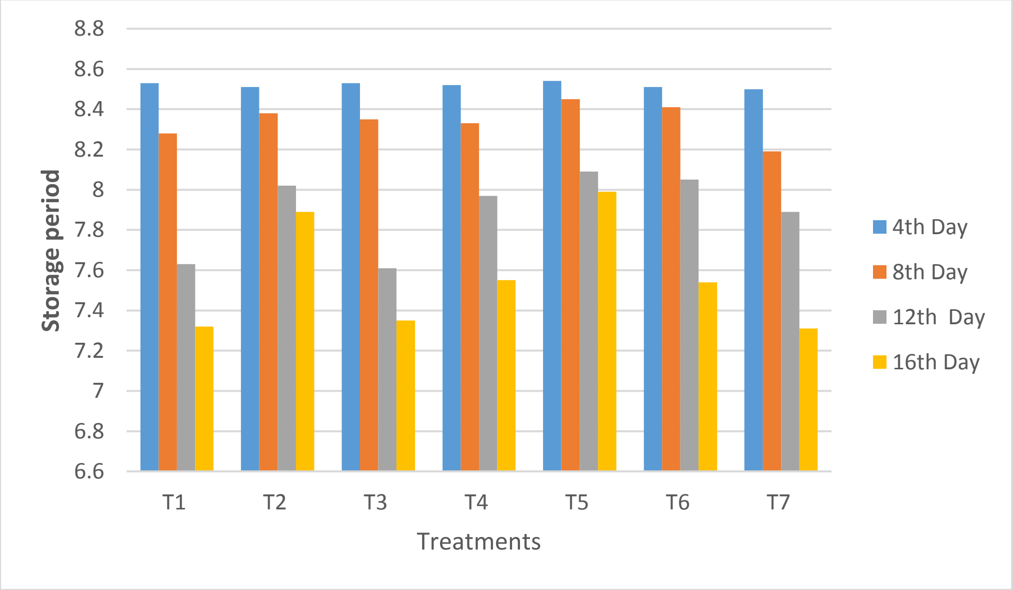


Figure 4.7 - Effect of edible coating on ascorbic acid (mg/ 100g) of pomegranate arils Treatments

4.8 Total Phenolic Content (mg gallic acid/100gm)

In the present research the data collected regarding the changes in total phenolic content of treated arils of pomegranate has been found non-significant and is presented in Table 4.8. Least changes were noticed in total phenolic content (mg gallic acid/100gm) of treated arils. Treatments T_4(1.5 % sodium alginate and 1 % ascorbic acid) was (46.75,36.50,31.28,18.25) on 4th,8th,12th and 16th day respectively recorded maximum and T_7(Control) was (46.75,21.08,18.50,4.70)) on 4th,8th,12th and 16th day recorded minimum.

Table 4.8 -Effect of edible coating on total phenolic content (mg gallic acid/100gm) of pomegranate arils Treatments

Treatments	Treatment details	Storage period			
		4th Day	8th Day	12th Day	16th Day
T_1	1% Sodium alginate	46.75	31.25	23.75	9.19
T_2	1% Sodium alginate + 1 % Ascorbic acid	46.75	22.02	21.75	11.75
T_3	1.5 Sodium alginate	46.75	25.25	24.50	7.75
T_4	1.5 Sodium alginate + 1 % Ascorbic acid	46.75	36.50	31.28	18.25
T_5	2 % Sodium alginate	46.75	22.75	16.75	6.75
T_6	2% Sodium alginate + 1 % Ascorbic acid	46.75	21.50	19.25	9.50
T_7	Control	46.75	21.08	18.50	4.70

Standard deviation ±0.05
n=10

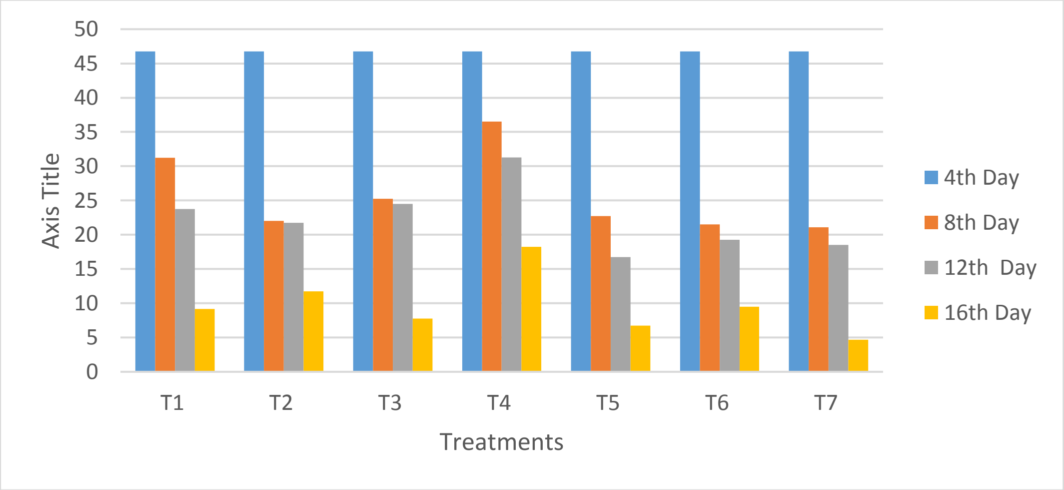


Figure 4.8 - Effect of edible coating on total phenolic content (mg gallic acid/100gm) of pomegranate arils Treatments

4.9 Microbial analysis (log_{10}cfu/gm)

Table 4.9 shows the changes in the level of total aerobic bacteria counts of control and chemical treated pomegranate arils during storage. The total level of aerobic bacteria counts in the control and treated samples stored at 10^0C reached 6.95log_{10}cfu/gm and 6.35log_{10}cfu/g, respectively at the end of 16th days of storage. Control samples exhibit maximum amount of bacterial growth in comparison to treated samples during its storage period. Due to increase in pH, bacterial count was found to be increased during its storage period of 16th days. Hence the viable counts of aerobic bacteria counts (log_{10}cfu/gm) in control and coated pomegranate arils were found significantly different ($p‹0.05$) during 16th days of storage at 10 ^{0}C.

Table 4.9 - Effect of edible coating on in total aerobic bacteria counts (log_{10} cfu/gm) of pomegranate arils Treatments

Treatments	Treatment details	Storage period			
		4th Day	8th Day	12th Day	16th Day
T_1	**1% Sodium alginate**	4.48	5.02	5.93	6.35
T_2	**1% Sodium alginate + 1 % Ascorbic acid**	4.48	4.92	5.84	6.39
T_3	**1.5 Sodium alginate**	4.48	4.71	5.61	5.75
T_4	**1.5 Sodium alginate + 1 % Ascorbic acid**	4.48	4.67	5.56	5.98
T_5	**2 % Sodium alginate**	4.48	4.89	5.78	6.15
T_6	**2% Sodium alginate + 1 % Ascorbic acid**	4.48	4.80	5.71	6.30
T_7	**Control**	4.48	5.11	6.44	6.95

Standard deviation ±0.05

n=10

Table 4.10 depicts the viable counts of yeast and mould ($\log_{10}$cfu/gm) of contro and treated sample during storage. The total level of yeast and mold counts in the control and treated samples stored at 10^0C reached 7.99 $\log_{10}$cfu/gm and 7.32 $\log_{10}$cfu/gm respectively at the end of 16th days of storage. Hence the viable counts of aerobic bacteria counts ($\log_{10}$cfu/gm)in control and coated pomegranate arils were found significantly different ($p‹0.05$) during 16th days of storage at 10 ^{0}C.

Table 4.10 - Effect of edible coating in yeast and mold counts ($\log_{10}$ cfu/gm) of pomegranate arils Treatments

Treatments	**Treatment details**	**Storage period**			
		4th Day	**8th Day**	**12th Day**	**16th Day**
T_1	**1% Sodium alginate**	5.47	6.26	7.03	7.32
T_2	**1% Sodium alginate + 1 % Ascorbic acid**	5.74	6.01	6.90	7.89
T_3	**1.5 Sodium alginate**	5.47	5.82	6.50	7.35
T_4	**1.5 Sodium alginate + 1 % Ascorbic acid**	5.47	5.74	6.38	7.55
T_5	**2 % Sodium alginate**	5.47	5.93	6.77	7.35
T_6	**2% Sodium alginate + 1 % Ascorbic acid**	5.47	5.89	6.61	7.54
T_7	**Control**	5.47	6.44	7.61	7.99

Standard deviation ±0.05

n=10

4.11 Sensory Analysis

The mean data pertaining to sensory analysis of treated arils is presented in Table 4.11.Overall acceptability of sensory analysis of pomegranate arils analysis is highest in T_3 (1.5 % Sodium alginate) and lowest in T_1 (1 % Sodium alginate). The highest rank found in T_3 and lowest in T_1.

Table 4.11 Sensory Analysis of pomegranate arils

Treatments	Colour	Flavour	Taste	Texture	Overall Acceptability	Average	Rank
T_1	6.92	6.64	6.77	6.69	6.85	6.77	7
T_2	7.31	6.96	6.92	6.81	7.13	7.03	4
T_3	7.54	7.19	7.04	7.23	7.21	7.24	1
T_4	7.54	6.77	7.00	6.77	7.15	7.05	3
T_5	6.46	6.85	6.92	6.77	6.96	6.99	5
T_6	7.54	6.81	6.77	7.23	7.12	6.89	6
T_7	7.62	6.58	7.04	7.15	7.19	7.12	2

n=10

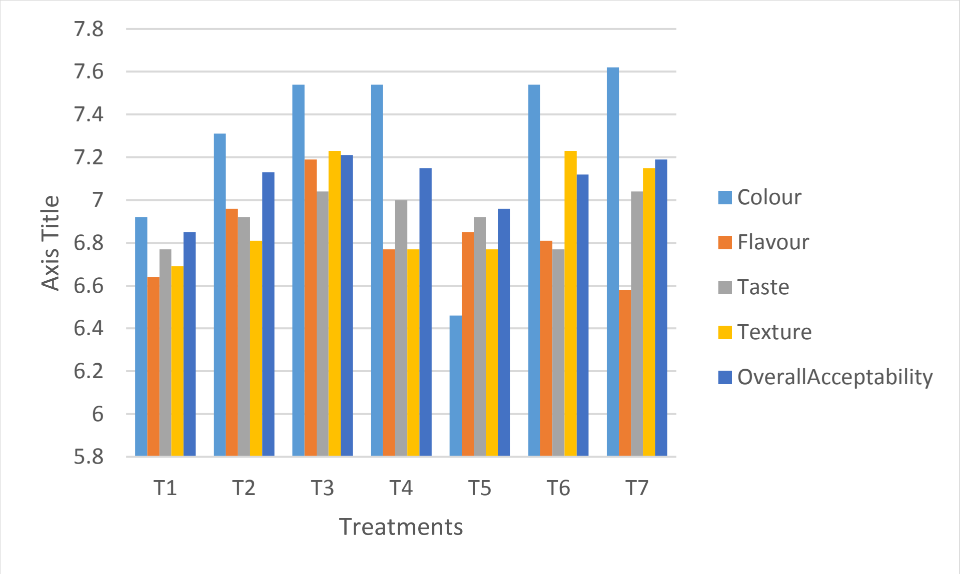


Figure 4.11 Sensory Analysis of pomegranate arils

Chapter-V

SUMMARY AND CONCLUSION

In the modern era, the fresh cut fruits and vegetables represent the rapidly growing segment of the produce industry due to increase in consumers demand for fresh, convenient and nutritious foods .This is due to the lifestyle of modern consumers that prefer a fresh product that is easier and faster to eat, and desire natural products that can promote health benefits. Perceiving the potential of shelf life enhanced pomegranate arils in the market, the present project was carried out for developing an eco-friendly and economically viable technique for preservation of pomegranate arils. Combinational use of suitable and simple preservation technologies such as edible coating, refrigeration and plastic packaging have been used to effectively enhance the shelf life of pomegranate arils.

Application of such preservative technologies can be very effective in significantly reducing post-harvest losses of fresh fruits and vegetables like pomegranate .Such hurdle preservative technologies an also come very handy in a developing country like India.

CONCLUSION

Based on the results obtained from the study, it can be inferred that edible coating of pomegranate arils with 2 % sodium alginate proved to be beneficial in maintaining the quality of arils during storage period. Application of 2% sodium alginate as edible coating on arils was found promising in maintenance of quality parameters such as total soluble solids, titratable acidity, total sugars, anthocyanin content etc. Overall acceptability of sensory analysis of pomegranate arils analysis is highest in T_3 (1.5 % Sodium alginate) and lowest in T_1 (1 % Sodium alginate).The highest rank found in T_3 and lowest in $T_{1.}$

Therefore, it is concluded that coating of 1 % sodium alginate can successfully be used to extend shelf life of pomegranate arils upto 12 days of storage.

Recommendation

The combinational use of preservation and hurdle technologies such as Edible coat, refrigeration, packaging in plastic films can significantly increase the shelf life of fresh or minimally processed pomegranate. Before developing any preservation technique, the combined effects and safety of various technologies and concepts used on the quality of products must be thoroughly studied.

Future Aspects

Although subsequent amount of research works relating the Edible coating has been done for the preservation of fresh fruits and vegetables. But still a path-breaking success in the development of efficient preservation technique is still awaited.

BIBLIOGRAPHY

Agar, I. T., Streif, J., & Bangerth, F. (1997). Effect of high CO2 and controlled atmosphere (CA) on the ascorbic and dehydroascorbic acid content of some berry fruits. Postharvest Biology and Technology, **11**(1), 47–55.

Ali Sabir, Ferhan, K, Sabir and Zeki, K. 2011. Effects of modified atmosphere packing and honey dip treatments on quality maintenance of minimally processed grape cv. Razaki during cold storage. *Journal of Food Science and Technology*. 48:312–318.

Andrady, A. L. and Xu, P. 1997. Elastic behaviour of chitosan films. *Journal of Polymer Science*. 5:307-521.

AOAC (2000). Vitamin C (ascorbic acid) in vitamin preparations and juices: 2,6 Titrimetric method final action. Association of Official Analytical Chemists Official Method, **4**(967), 21.

Aviram, M.; B. Fuhrman; M. Rosenblat; N. Volkova; M.Kaplan; T. Hayek; D. Presser and L. Dornfeld (2002). Pomegranate juice polyphenols decreases oxidative stress, low-density lipoprotein atherogenic modificationsand atherosclerosis. Free Radical Res., 36: 72–73.

Aviram, M.; L. Dornfeld; M. Rosenblat; N. Volkova; M. Kaplan and R. Coleman (2000). Pomegranate juice consumption reduces oxidative stress, atherogenic modifications to LDL, and platelet aggregation: studies in humans and in atherosclerotic apolipoprotein Eedeficient mice. *American J. Clinical Nutrition*, **71** (5): 1062-1076.

Ayhan, Z. and E. O. Türk (2009). Overall quality and shelf life of minimally processed and modified atmosphere packaged “ready-to-eat” pomegranate arils. *Journal of Food Science*; 74: 399-405.

Ayhan, Z. and Esturk, O. 2009. Overall quality and shelf life of minimally processed and modified atmosphere packaged ready-to-eat pomegranate arils. *Journal of Food Science*. **74**(5): 399-405.

Babich, H., & Stotzky, G. (1978). Toxicity of zinc to fungi, bacteria, and coliphages: Influence of chloride ions. Applied and Environmental Microbiology, **36**(6), 906–914.

Bondet, V.; W. Brand-Williams; and C Berset (1997). Kinetics and mechanism of antioxidant activity using the DPPH free radical method. LWT-Food Sci. Technol; 30: 609– 615.

Bourtoom, T. (2008). Edible films and coatings: Characteristics and properties. *International Food Research Journal*, **15**(3), 237–248.

Braga, L.C.; J.W. Shupp; C. Cummings; M. Jett; J.A. Takahashi and L.S. Carmo (2005). Pomegranate extract inhibits *Staphylococcus aureus* growth and subsequent enterotoxin production. *J. Ethnopharmacology*, **96**(1-2): 335-339.

Brayner, R., Ferrari-Iliou, R., Brivois, N., Djediat, S., Benedetti, M. F., & Fiévet, F. (2006). Toxicological impact studies based on Escherichia coli bacteria inultrafine ZnO nanoparticles colloidal medium. Nano Letters, **6**(4), 866–870.

Cadogan, E. I.; C. H. Lee; S. R. Popuri and H. Y. Lin (2014). Effect of solvent on physicochemical properties and antibacterial activity of chitosan membranes. *International Journal of Polymeric Materials and Polymeric Biomaterials*; **63**(14): 708-715.

Caleb, O. J., Opara, U. L., & Witthuhn, C. R. (2012). Modified atmosphere packaging of pomegranate fruit and arils: A review. Food and Bioprocess Technology, **5**(1), 15–30.

Caleb, O. J., Opara, U. L., Mahajan, P. V., Manley, M., Mokwena, L., & Tredoux, A. G. J. (2013). Effect of modified atmosphere packaging and storage temperature on volatile composition and postharvest life of minimally-processed pomegranate arils (cvs. ‘‘Acco” and ‘‘Herskawitz”). Postharvest Biology and Technology, 79, 54–61.

Caleb, O. J.; U. L. Opara and C. R. Witthuhn (2012). Modified atmosphere packaging of pomegranate fruit and arils: a review. Food and Bioprocess Technology; 5: 15-30.

Caleb, O. J.; U. l. Opara; P. V. Mahajan; M. Manley; L. Mokwena and A.G.J. Tredoux (2013). Effect of modified atmosphere packaging and storage temperature on volatile composition and postharvest life of minimally-processed pomegranate arils (cvs. Acco and erskawitz). Postharvest Biology and Technology; 79: 54-61.

Caleb, O.J, Opara, U.L, Mahajan, P.V, Manley, M, Mokwena, L, Tredoux, A.G.J, 2013.Effect of modified atmosphere packaging and storage temperature on volatile composition and postharvest life of minimally-processed pomegranate arils (cvs. ‘Acco’ and ‘Herskawitz’). Postharvest Biol. Tech. 79, 54-61.

Chiabrando, V. and G. Giacalone (2015). Anthocyanins, phenolics and antioxidant capacity after fresh storage of blueberry treated with edible coatings. *International Journal of Food Sciences and Nutrition*; http://dx.doi.org/10.3109/09637486.2014.986075

Chung Y.C. and C.Y. Chen (2008). Antibacterial characteristics and activity of acid-soluble chitosan. Bioresource Technology, 99: 2806–2814.

Dahiya, N.; R. Tewari; R. Tiwari and G.S. Hoondal (2006). Biochemical aspects of chitinolytic enzymes: A review. Applied Microbiology and Biotechnology, 71: 773–782.

Das, D.K, Dutta, H. and Mahanta, C.L. 2013. Development of a rice starch-based coating with antioxidant and microbe-barrier properties and study of its effect on tomatoes stored at room temperature. LWT-Food Science and Technology **50**(1): 272-278.

De Reuck, K.; D. Sivakumar and L. Korsten (2009). Effect of integrated application of chitosan coating and modified atmosphere packaging on overall quality retention in litchi cultivars. *Journal of the Science of Food and Agriculture*, 89: 915–920.

Dhall, R. K. (2013). Advances in edible coatings for fresh fruits and vegetables: a review. Critical Reviews in Food Science and Nutrition; **53**(5): 435-450.

Dhinesh K.V and Ramasamy D. 2016. Pomegranate Processing and Value Addition: Review. J Food Process Technol. 7:565.

Dong, H, Cheng, L, Tan, J, Zheng, K, Jiang, Y, 2004. Effect of chitosan coating on quality and shelf life of Effect of chitosan coating on quality and shelf life of peeled litchi fruit. *J. Food Eng*. **64**(3), 355-358.

Drosionon, E. H.; N. P. Skandamis and M. Mataragas (2009). Antimicrobials treatments. In: Toldrá, F. (Ed.) Safety of meat and processed meat. Springer Science and Business Media, LLC, New York, USA. Chapter, 10: 255 – 274.

Emamifar, A., Kadivar, M., Shahedi, M., & Soleimanian-Zad, S. (2010). Evaluation of nanocomposite packaging containing Ag and ZnO on shelf life of fresh orange juice. Innovative Food Science & Emerging Technologies, **11**(4), 742–748.

Fisher, R.A. 1950. Statistical methods for research workers. Oliver and Boyd, Edinburgh.

Garcia, L.C., Pereira, L.M., de Luca Sarantópoulos, C.I., Hubinger, M.D. (2010). Selection of an edible starch coating for minimally processed strawberry. Food and Bioprocess Technology, **3**(6), 834-842.

Ghasemnezhad, M., Zareh, S., Rassa, M., & Sajedi, R. H. (2012). Effect of chitosan coating on maintenance of aril quality, microbial population and PPO activity of pomegranate (Punica granatum L. cv. Tarom) at cold storage temperature. *Journal of the Science of Food and Agriculture*, **93**(2), 368–374.

Ghasemnezhad, M., Zareh, S., Rassa, M., Sajedi, R. H. (2013). Effect of chitosan coating on maintenance of aril quality, microbial population and PPO activity of pomegranate (Punica granatum L. cv. Tarom) at cold storage temperature. *Journal of the Science of Food and Agriculture*, **93**(2), 368-374.

Gil, M. I., Martínez, J. A., & Artés, F. (1996). Minimally processed pomegranate seeds. LWT – Food Science and Technology, 29, 708–713.

Gil, M.I., Martınez, J.A., Artés, F. (1996). Minimally processed pomegranate seeds. LWT-Food Science and Technology, **29**(8), 708-713.

Meighani, H., Ghasemnezhad, M. (2015). Effect of different coatings on post-harvest quality and bioactive compounds of pomegranate (Punica granatum L.) fruits. *Journal of Food Science and Technology*, **52**(7):4507–4514.

Holland, D, Hatib, K, and Bar-Y I. 2009. Pomegranate: botany, horticulture, breeding. Horticultural Review, 35: 127-191.

Jiang, Y, Li, Y, 2001. Effects of chitosan coating on postharvest life and quality of longan fruit. Food Chem. **73**(2), 139-143.

Kester J, Fennema O (1986) Edible films and coatings: a review. Food Technol. 40:47 59.

Mırdehghan S.H., Rahemi, Serrano, M., Guillen, F., Martıänez-Romero, D., Valero, D. (2007). The application of polyamines by pressure or ımmersion as a tool to maintain functional properties in stored pomegranate arils. *Journal of Agriculture and Food Chemistry*, 55,755−76.

Nabigol, A., Asghari, A. (2013). Antifungal activity of *Aloe vera* gel on quality of minimally processed pomegranate arils, *International Journal of Agronomy and Plant Production*, **4**(4), 833–838.

Ojeda, G.A., Sgroppo, S.C., Zaritzky, N.E. (2014). Application of edible coatings in minimally processed sweet potatoes (Ipomoea batatas L.) to prevent enzymatic browning. *International Journal of Food Science & Technology*, **49**(3), 876-883.

Olivas, G.I., Mattinson, D.S., Barbosa-Cánovas, G.V. (2007). Alginate coatings for preservation of minimally processed ‘Gala’apples. Postharvest biology and Technology, **45**(1), 89-96.

Opara, U.L., Atukuri, J., Fawole, O.A. (2015). Application of physical and chemical postharvest treatments to enhance storage and shelf life of pomegranate fruit—A review. Scientia Horticulture, 197, 41-49.

Öz, A.T., Hiçyilmaz, S., Yarsi, T. (2017). Effects of the Alternative Postharvest Treatments on ‘Hicaznar’ Pomegranate Fruit Phytochemicals, Organic Acids and Sugar Content. *Indian Journal of Pharmaceutical Education and Research*, **51**(3)-49.

Oz, A.T., Ulukanli, Z. (2012). Application of edible starch-based coating including glycerol plus oleum nigella on arils from long-stored whole pomegranate fruits. *Journal of Food Processing and Preservation*, **36**(1), 81-95.

Özdemir, K.S., Gökmen, V. (2017). Extending the shelf-life of pomegranate arils with chitosan-ascorbic acid coating. LWT-Food Science and Technology, 76, 172-180.

Pen, L.T., Jiang, Y.M. (2003) Effect of chitosan coating on shelf life and quality of fresh-cut Chinese water chestnut. Lebensmittel-Wissenschaft & Technologie, 36,359–364.

Quirós-Sauceda AE, Ayala-Zavala JF, Olivas GI, González-Aguilar GA. 2014. Edible coatings as encapsulating matrices for bioactive compounds: a review. *J Food Sci Technol*. **51**(9):1674–1685

Salama, M.E.I, Ayaad, H.M, Aboul-Anean, H.E, Fahmy, H.M, 2012. Effect of edible coating as a carrier of essential oils and ultraviolet light (UV-C) on improving quality of minimally processed Manfalouty pomegranate. Res. *J. Agric. Biol. Sci*. **8**(2), 315-324.

Shaarawi, S.A., Salem, A.S., Elmaghraby, I. M., & Eman, A.A. (2016). Effect of salicylic acid, calcium chloride and calcium lactate applications on quality attributes of minimally-processed 'W*onderful*' pomegranate arils. Notulae Botanicae Horti Agrobotanici Cluj-Napoca, **44**(2):508-517.

Zahran, A.A., Hassanein, R.A., Abdelwahab, A. T. (2015). Effect of chitosan on biochemical composition and antioxidant activity of minimally processed " Wonderful " pomegranate arils during cold storage, Journal of Applied Botany and Food Quality 88, 241–248.

Zhang, D.L. and Quantick, P.C. 1998. Antifungal effects of chitosan coating on fresh strawberries and raspberries during storage. *Journal of Horticulture Science and Biotechnology*. 73: 763–67.

* * *

APPENDICES

Appendix-A

Sensory score card for sensory analyses of pomegranate arils

SENSORY SCORE CARD

Product   Pomegranate arils

Date

Time

Name of Panelist

Introduction

The given samples are of pomegranate arils; you are requested to judge the sample in terms of general acceptability with special reference to Color, Flavor and overall acceptability on the scale described below:

Rate the color of skin of pomegranate arils on the basis of 1-9 color scale:

Dislike Extremely	1
Dislike Very Much	2
Dislike Moderately	3
Dislike Slightly	4
Neither like nor dislike	5
Like Slightly	6
Like Moderately	7
Like Very Much	8
Like Extremely	9

Table A Score the given pomegranate for colour parameter:

S. No.	**Sample Code**	**Colour Score**
1.	Sample A	
2.	Sample B	

9 Pont Hedonic scale Rating and their Numerical Values

Rating	**Numerical value**
Dislike Extremely	**1**
Dislike Very Much	**2 (1.5-2.5)**
Dislike Moderately	**3 (2.6-3.5)**
Dislike Slightly	**4 (3.6-4.5)**
Neither like nor dislike	**5 (4.6-5.5)**
Like Slightly	**6 (5.6-6.5)**
Like Moderately	**7 (6.6-7.5)**
Like Very Much	**8 (7.5-8.5)**
Like Extremely	**9 (8.6-9.0)**

Table B Score the giver pomegranate arils for the following sensory features:

S. No.	**Characteristics**	**Sample A**	**Sample B**
1.	Crunchiness		
2.	Stickiness of the skin		
3.	Toughness		
4.	Juiciness		
5.	Sourness		
6.	Aroma intensity		
7.	Grassiness		
8.	Overall acceptability		

Remarks:

Signature:

Table C Description of sensory terms used in the score card:

S. No.	Characteristics	Description
1.	**Crunchiness**	Making a crunching or cracking sound , as when chewed
2.	**Stickiness of the skin**	Degree to which skin of fruit adheres the teeth
3.	**Toughness**	Hard to cut or chew
4.	**Juiciness**	Yielding juice when pressed/chewed with mouths
5.	**Sourness**	Having a taste characteristic of that produced by acid;sharp.tart.or tangy
6.	**Aroma intensity**	Intensity of pleasant characteristic odour of the food
7.	**Grassiness**	Having the scent of glass, including freshly grown grass and hay

◆