

**ASSESSMENT OF QUALITY AND SHELF LIFE OF FRESH-CUT
PINEAPPLE (*Ananas comosus*) COATED WITH *Aloe vera* AND
HONEY AT REFRIGERATED CONDITION**

**A
THESIS
BY**

TANNY SAHA
Student ID.: 1405203
Session: 2014-2015
Semester: July-December, 2015

**MASTER OF SCIENCE
IN
FOOD PROCESSING AND PRESERVATION**



DEPARTMENT OF FOOD PROCESSING AND PRESERVATION

**HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY
UNIVERSITY DINAJPUR-5200**

DECEMBER, 2015

**ASSESSMENT OF QUALITY AND SHELF LIFE OF FRESH-CUT
PINEAPPLE (*Ananas comosus*) COATED WITH *Aloe vera* AND
HONEY AT REFRIGERATED CONDITION**

**A
THESIS
BY**

TANNY SAHA
Student ID.: 1405203
Session: 2014-2015
Semester: July-December, 2015

**Submitted to the
Department of Food Processing and Preservation**

Hajee Mohammad Danesh Science and Technology University, Dinajpur

In Partial Fulfillment of the Requirement for the Degree of

**MASTER OF SCIENCE (MS)
IN
FOOD PROCESSING AND PRESERVATION**

DEPARTMENT OF FOOD PROCESSING AND PRESERVATION

**HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY
UNIVERSITY, DINAJPUR-5200**

DECEMBER, 2015

**ASSESSMENT OF QUALITY AND SHELF LIFE OF FRESH-CUT
PINEAPPLE (*Ananas comosus*) COATED WITH *Aloe vera* AND
HONEY AT REFRIGERATED CONDITION**

**A
THESIS
BY**

TANNY SAHA
Student ID.: 1405203
Session: 2014-2015
Semester: July-December, 2015

Approved as to style and content by

.....
(Md. Mojaffor Hosain)
Supervisor

.....
(Dr. Maruf Ahmed)
Co-supervisor

.....
Chairman of the Examination Committee
and
Chairman, Department of Food Processing and Preservation

DEPARTMENT OF FOOD PROCESSING AND PRESERVATION

**HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY
UNIVERSITY, DINAJPUR-5200**

DECEMBER, 2015

DEDICATED
TO MY
BELOVED PARENTS



ABSTRACT

The effects of different concentrations of *Aloe vera gel* (50%, 75% and 100%) and honey (25%, 50% and 100%) on physico-chemical changes in fresh cut pineapple stored at 4⁰C were assessed in this research work. Pineapples were washed and cut into 2cm pieces. Then the pineapple pieces were coated by different concentration of *Aloe vera* and honey by dipping for 2 minutes and stored at 4°C for 12 days. The amount of juice leakage, firmness, pH, total soluble solids, vitamin C content, color and microbial load were observed during storage. Sensory testing was carried out to assess their acceptability. A significant variation was observed in weight loss, vitamin C, color, flavor and textural properties in all treated samples compared to control during entire storage period studied (12 days). Treatment with 100% *Aloe vera gel* was found most effective to prevent physico-chemical changes and to increase the shelf-life of fresh cut pineapple among the treatments. In addition, at the end of storage period, the highest amount of vitamin C (27.35 mg/100g) was resulted in sample coated with 100% *Aloe vera*. The results obtained in this study showed that *Aloe vera gel* could be used as effective coating material to prevent the physico-chemical and microbial changes in fresh cut pineapple during storage at refrigerated condition.

ACKNOWLEDGEMENT

Firstly, the author expresses her deepest sense of gratitude to almighty Creator whose blessings have enabled the author to carry out the whole research work and to complete this thesis successfully.

The author would like to express her deepest gratitude and profound regard to research Supervisor Md. Mojaffor Hosain, Assistant Professor Department of Food Processing and Preservation, for his guidance, invaluable advices, suggestion, encouragement, effort and moral support throughout the completion of this study.

The author would also like to express her sincere thanks and appreciations to her respected Co-supervisor Dr. Maruf Ahmed, Chairman and Associate Professor Department of Food Processing and Preservation, for his help, suggestions and assistance throughout the entire period of research work, as well as in the preparation of manuscript.

A special note of thanks to Shakti Chandra Mondal, Assistant Professor Department of Food Processing and Preservation, for his help, suggestion and thoughtful criticism throughout the entire period of research work.

The author would like to express her gratefulness to National Science and Technology (NST) under the ministry of Science and Technology, Bangladesh for financial support.

Cordial thanks are extended to all the staff member of the Department of Food Processing and Preservation and some of my younger brothers and sisters for their help and co-operation.

Specially, the author would like to express her heartfelt indebtedness to her beloved parents for their invaluable advices, patience, inspirations, sacrifices, blessing and encouragement for completing the research work successfully.

At last, the author is truly obliged to all those who have contributed to the completion of the research and improvements of this thesis.

The Author

CONTENTS

CHAPTER	TITLE	PAGE NO.
	ABSTRACT	I
	ACKNOWLEDGEMENT	II
	CONTENTS	III
	LIST OF ABBREVIATION	VI
	LIST OF TABLES	VII
	LIST OF FIGURES	VIII
	LIST OF APPENDICES	IX
CHAPTER I	INTRODUCTION	1-2
CHAPTER II	REVIEW OF LITERATURE	3-8
	2.1 Production of pineapple	3
	2.2 Health benefits of pineapple	3
	2.3 Fresh cut fruits and vegetables	4
	2.4 Natural methods to preserve fresh cut products	5
	2.5 Edible coating	5
	2.6 <i>Aloe vera</i>	7
	2.7 Health benefits of <i>Aloe vera</i>	7
	2.8 Use of <i>Aloe vera</i> in edible coating formulation	7
	2.9 Honey	8
	2.10 Insight from the review of literature	8
CHAPTER III	MATERIALS AND METHODS	9-16
	3.1 Experimental site	9
	3.2 Source of fruits and coating materials	
	3.3 Preparation of samples and edible coating solutions	9
	3.3.1 Preparation of pineapple	9
	3.3.2 Preparation of <i>Aloe vera</i> gel	9
	3.3.3 Process of making <i>Aloe vera</i> coating	9

CONTENTS (CONTINUED)

CHAPTER	TITLE	PAGE NO.
	3.3.4 Process of making honey coating	10
	3.3.5 Process of making <i>Aloe vera</i> - honey coating	10
	3.3.6 Pineapple coating process	10
	3.3.7 Treatments	10
3.4	Proximate composition of fresh pineapple	10
	3.4.1 Determination of moisture content	10
	3.4.2 Determination of protein	11
	3.4.3 Determination of fat	12
	3.4.4 Determination of total ash	12
	3.4.5 Determination of carbohydrate	12
3.5	Fruit physicochemical analysis	13
	3.5.1 Juice leakage	13
	3.5.2 Firmness	13
	3.5.3 Color	13
	3.5.4 Total soluble solids (TSS)	13
	3.5.5 pH	14
	3.5.6 Vitamin C	14
3.6	Microbial analysis	14
	3.6.1 Preparation of media	14
	3.6.2 Preparation of dilution blank	15
	3.6.3 Procedure of plating	15
	3.6.4 Incubation and colony count	16
3.7	Sensory analysis	16
3.8	Statistical analysis	16

CONTENTS (CONTINUED)

CHAPTER	TITLE	PAGE NO.
CHAPTER IV	RESULTS AND DISCUSSION	17-27
	4.1 Proximate analyses	17
	4.2 Physico-chemical properties	18
	4.2.1 Juice leakage	18
	4.2.2 Firmness	19
	4.2.3 Total soluble solids (TSS)	19
	4.2.4 pH	20
	4.2.5 Vitamin C	21
	4.2.6 Color	22
	4.3 Microbiological analysis	24
	4.4 Sensory evaluation	24
	4.4.1 Color	26
	4.4.2 Flavor	26
	4.4.3 Texture	26
	4.4.4 Taste	27
	4.4.5 Overall acceptability	27
CHAPTER V	SUMMARY AND CONCLUSION	28
	REFERENCES	29-36
	APPENDICES	37-41

LIST OF ABBREVIATION

AOAC	= Association of Analytical Chemists
CFU	= Colony Forming Unit
D	= Day
et.al.	= and others
FAO	= Food and Agriculture Organization
g	= Gram
GAE	= Gallic Acid Equivalent
LDPE	= Low Density Polyethylene
mg	= Milligram
Min	= Minute
ml	= Milliliter
N	= Normality
°C	= Degree Celsius
SD	= Standard Deviation
µg	= Microgram

LIST OF THE TABLES

SL. NO.	TITLE	PAGE NO.
Table 2.1	Nutritional composition of pineapple	4
Table 2.2	Nutritional composition of <i>Aloe vera</i>	7
Table 3.1	Dilution of sample	15
Table 4.1	Proximate composition of pineapple , <i>Aloe vera</i> and honey	17
Table 4.2	Effect of <i>Aloe vera</i> and honey coating on firmness	19
Table 4.3	Effect of <i>Aloe vera</i> and honey coating on pH	21
Table 4.4	Effect of <i>Aloe vera</i> and honey coating on color	23
Table 4.5	Effect of <i>Aloe vera</i> and honey coating on microbial growth during refrigerated storage	24
Table 4.6	Effect of <i>Aloe vera</i> and honey coating on sensory attributes at 12 th day of storage	26

LIST OF THE FIGURES

FIGURE NO.	TITLE	PAGE NO.
Figure 4.1	Effect of <i>Aloe vera</i> and honey coating on juice leakage	18
Figure 4.2	Effect of <i>Aloe vera</i> and honey coating on TSS	20
Figure 4.3	Effect of <i>Aloe vera</i> and honey coating on vitamin C	22
Figure 4.4	Sensory evaluation of pineapple	25

LIST OF THE APPENDICES

APPENDIX	TITLE	PAGE NO.
I	Effect of <i>Aloe vera</i> and honey coating on juice leakage of fresh cut pineapple at storage condition	37
II	Effect of <i>Aloe vera</i> and honey coating on firmness of fresh cut pineapple at storage condition	38
III	Effect of <i>Aloe vera</i> and honey coating on TSS of fresh cut pineapple at storage condition	39
IV	Effect of <i>Aloe vera</i> and honey coating on pH of fresh cut pineapple at storage condition	40
V	Effect of <i>Aloe vera</i> and honey coating on vitamin C content of fresh cut pineapple at storage condition	41



CHAPTER I

INTRODUCTION

CHAPTER I

INTRODUCTION

Pineapple (*Ananas comosus*) is a most popular tropical fruit in Bangladesh. The annual production of pineapple was nearly 180.9 thousand metric tons (BBS, 2012) and nearly 43% of its wasted every year (Hassan *et al.*, 2010). It is a rich source of vitamin C, fiber, minerals and antioxidant activity (Montero-Calderon *et al.*, 2010).

Demand of fresh cut fruits and vegetables are increasing day by day. It offers a convenient, time saving, ready to eat product. The shelf life of fresh cut fruit is very short because peeling and cutting of fruits increases metabolic activities such as respiration and delocalization of enzymes and substrates. These cause water loss, browning, softening, off-flavor and susceptible to microbial growth which give fresh-cut fruit a shorter shelf life (Di Egidio *et al.*, 2009).

A number of preservation techniques have been used to reduce deterioration, extend shelf life and preserve the nutritional quality of fresh fruits. Recently edible films and coatings become more popular for shelf life extension of fresh fruits and vegetables (Vargas *et al.*, 2008). Edible coating can be developed by using polysaccharides, proteins, and lipids or from a combination of these groups of material for fresh products, which may be improved by the addition of plasticizers, surfactants and emulsifiers (Arvanitoyannis and Gorris, 1999).

Consumers prefer the use of natural compounds instead of synthetic agents. In several studies it has been shown that *Aloe vera* can be used as coating material for fresh fruits and vegetables. It contains different types of nutrients such as vitamins, fatty acids, amino acids, sugars, minerals, and enzymes. Moreover, it has also anti-inflammatory (Esua and Rauwald 2006) and antimicrobial activity. *Aloe vera* gel provides a barrier to O₂ and CO₂ and acts as moisture barrier and thus reduces weight loss, browning, softening of fruits and growth of yeast and molds on fruits (Valverde *et al.*, 2005).

Honey can also be used as coating material because of its antibrowning (Jeon and Zhao, 2005) and antioxidant capacity (Bogdanov *et al.*, 2008). It also contains anti-bacterial and anti-fungal properties (Molan, 1992).

With all the important properties, *Aloe vera* and honey have selected as a coating material for fresh cut pineapple. Research with *Aloe vera* and honey for the purpose of using coating agents has not been carried out in Bangladesh so far literature and review ascertained. This study would help to generate new knowledge that could help to reduce the wastage of pineapple. Therefore, the objectives of this study were selected as below:

- a) to make suitable edible coating formulations based on *Aloe vera* and honey for fresh-cut pineapple.
- b) to examine the physico-chemical changes and microbial count of coated and uncoated fresh cut pineapple during refrigeration temperature.
- c) to assess the sensory of stored fresh cut and coated pineapple.



CHAPTER II

REVIEW OF LITERATURE

CHAPTER II

REVIEW OF LITERATURE

2.1 Production of pineapple

The pineapple (*Ananas comosus*) is a tropical fruit which grows in countries situated in the tropical and sub-tropical regions. Pineapple is the most economically significant plant in the Bromeliaceae family. It is edible, juicy fruit commonly known as “Anarash” in Bangladesh. It is widely cultivated in the districts of Tangail, Rangamati, Chittagong, Bandarban, Dhaka, Mymensingh, Khagrachari, Sylhet and Moulvibazar. Among all the fruits produced in the country, pineapple ranks 4th in terms of total cropping area and production (BBS, 2013). The annual production of pineapple was nearly 180.9 thousand metric ton (BBS, 2012). At least ninety varieties of pineapple are cultivated in the world but in our country only three varieties such as Giant Kew (locally kalendar), Honey Queen (jaldubi), and Red Spanish (ghorashal) are mostly grown (BBS, 2013).

2.2 Health benefits of pineapple

Fresh pineapple is rich in vitamin B complexes groups like folates, thiamin, pyridoxine, riboflavin and minerals like copper, manganese and potassium. It is low in calories and contains a proteolytic enzyme bromelain that helps to digest food by breaking down protein. Bromelain has anti-inflammatory, anti-clotting and anti-cancer properties. Fresh pineapple is an excellent source of antioxidant vitamin C (Hossain and Rahman, 2011) which is essential for the collagen synthesis in the body and helps to prevent scurvy; develop resistance against infectious agents and scavenge harmful, pro-inflammatory free radicals from the body. The nutritional composition of pineapple is shown in the Table 2.1.

Table 2.1 Nutritional composition of pineapple per 100 gm

Nutrients	Amount
Protein	0.54g
Carbohydrate	13.12g
Diatery fibre	1.4g
Energy	50 kcal
Vitamin A	130 I.U
Iron	0.29mg
Calcium	13mg
Magnesium	12mg
Potassium	109mg
Phosphorus	8mg
Vitamin B ₁	0.079mg
Vitamin B ₂	0.032mg
Vitamin B ₃	0.5mg
Vitamin B ₆	0.112mg
Vitamin C	47.8mg
Zinc	0.12mg

Source : Gazi, 2013

2.3 Fresh cut fruits and vegetables

The USDA and FDA definitions for “fresh” and “minimally processed” fruits and vegetables imply that fresh-cut (precut) products have been freshly cut, washed, packaged, and maintained with refrigeration. Fresh-cut products are raw, and even though processed (physically altered from the original form), they remain in a fresh state, ready to eat or cook, without freezing, thermal processing, or treatments with additives or preservatives (AMS 1998 and Anonymous 1998). Consumer demand for fresh cut products has been increased day by day. Since 1995, consumer demand for minimally processed, ready-to-eat fruits and vegetables has led to growth in the fresh-cut industry of 10% per year (Barth, 2000). Fresh fruits and vegetable contains not only vitamins, minerals, and dietary fiber, but it also offers other constituents, such as carotenoids and flavonoids which help to prevent degenerative diseases and lower the risk of cancer and heart disease (Grassmann *et al.*, 2002).

In the last two decades, food scientists have tried to develop new technologies that increase the shelf life of fresh cut product with good nutritive quality. In the same time consumer become more critical on the use of synthetic agents in foods. The growing demand for slightly processed products with good nutritive value insists researchers to focus most on developing new ways of extending the shelf life of fresh-cut produce.

2.4 Natural methods to preserve fresh cut products

Fresh-cut fruits processing techniques are still under development because of the difficulties in preserving their fresh-like quality for prolonged periods (Soliva-Fortuny and Martin-Belloso, 2003). Consumers and producers interest has encouraged the researchers to determine methods that can be maintained the antioxidant content of fruits and vegetables after processing (Chantanawarangoon and Kader, 2002). Several methods such as refrigeration, controlled atmosphere packaging, use of additives, and edible coatings have been used to minimize the deleterious effects of minimally processed fruits and vegetables (Wong *et al.*, 1994). Temperature, essential oils, modified atmospheres and packaging are more natural methods for keeping quality of fresh cut fruits and vegetables.

Temperature is one of the most important factors. It helps to control freshness and decay of the fresh cut products as well as increase the shelf life of the product. Essential oil is another natural method which helps to maintain the quality of fresh cut fruits and vegetables during storage. Modified and control atmosphere inhibits metabolic activity, decay, browning and thus helps to maintain quality and extend shelf life (Gunes and Lee, 1997) and especially by inhibiting ethylene biosynthesis and action (Mathooko *et al.*, 1995).

2.5 Edible coating

Edible coating is one of the novel methods that reduce the deleterious effect of minimal processing products. An edible coating is the application of thin layer of edible material on the surface of the food and can be eaten as part of the whole product. Edible coatings are capable of producing modified atmosphere on coated fruits (Nisperos *et al.*, 1996). Modified Atmosphere Packaging (MAP) has been used to extend the postharvest shelf life of fruits by reducing respiration rate and delaying senescence (Drake *et al.*, 1987). However, it causes anaerobiosis, and the fruit fails to ripen properly (El Ghaouth *et al.*,

1992b). It acts as a semipermeable barrier that helps to reduce loss of water and color changes, improve texture and mechanical integrity. It also helps to retain volatile flavor compounds and reduce microbial growth. Research has been conducted on the optimum storage atmosphere for fresh whole produce, but limited information is available on optimum atmosphere for fresh-cut produce (Gunes *et al.*, 2001).

In addition, the new generation coatings are being designed for controlled release of antioxidants, nutraceuticals, chemical additives and natural antimicrobial agents (Vargas *et al.*, 2008). Appropriate selection of edible coating is important for maintaining better quality of fresh cut fruits.

Azarakhsh (2012) showed the effect of alginate and gellan-based edible coating formulations for fresh-cut pineapples. They reported that after 10 days storage weight loss and respiration rate were significantly lower and firmness was well maintained.

Mantilla *et al.* (2013) worked on fresh cut pineapples and measured the effect of multilayered antimicrobial edible coating on quality and shelf-life of fresh-cut pineapple. In this study the moisture content of fresh pineapple was measured by using AOAC method 920.151. They found that the application of antimicrobial coating was effective in maintaining good texture, reduced the weight loss and increased the shelf life of fresh cut pineapples.

Oms- Oliu *et al.* (2008) studied the effect of alginate, pectin and gellan based edible coatings on the shelf life of fresh cut “Piel di Sapo” melon. They reported that all coatings prevented dehydration and ethylene production and did not improve microbial stability of fresh cut melon packaged under passive modified atmosphere. The application of edible coating retarded the microbial growth of fresh cut fruits.

Sayka *et al.* (2014) studied the effect of low molecular weight chitosan coating on physico-chemical properties and shelf life extension of pineapple. In this study they measure the moisture content, ash and total soluble solid content of fresh pineapple and chitosan coated pineapple at initial day and end of storage period. They reported that irradiated chitosan treated pineapple increased the shelf life of pineapple with minimum loss of moisture, shrivel, increased ascorbic acid content , protected pineapple fruits from visual fungal growth and able to conserve better sensory characteristics.

Brasil *et al.* (2012) researched on multilayered antimicrobial edible coating of fresh cut papaya. They observed that the multilayered antimicrobial coating improved the microbiological and physicochemical quality of fresh-cut papaya and extend the shelf life up to 15 days at 4°C. The coating reduced the losses of Vitamin C and total carotenoids content.

2.6 *Aloe vera*

Aloe vera is a tropical and subtropical plant. It has been well known for its medicinal and therapeutic properties. The two major liquid sources of *Aloe vera* are a yellow latex (exudates) and a clear gel (mucilage), which proceeds from the large leaf parenchymatic cells (Ni *et al.*, 2004). The raw pulp of *Aloe vera* contains about 98.5% water, while the mucilage or gel consists of about 99.5% water (Eshun and He, 2004).

2.7 Health benefits of *Aloe vera*

It consists of number of nutrients such as vitamins, fatty acids, amino acids, sugars, minerals, and enzymes. Moreover, other properties such as antiinflammatory and antibiotic activities against some diseases (diabetics, cancer, allergy, AIDS) have been reported (Eshun and He, 2004). It has also been reported that the *Aloe vera* extracts possessed antimicrobial activity against bacterial pathogens from gram positive and gram negative (Adetunji, 2008). So, it can be used as a functional ingredient for health benefits. The nutritional composition of *Aloe vera* as follows:

Table 2.2 Nutritional composition of *Aloe vera* per 100 gm

Nutrients	Amount
Moisture	98.93g
Protein	0.12g
Fat	0.01g
Carbohydrate	0.66g
Ash	0.16g

Source: Karina *et al.*, 2013

2.8 Use of *Aloe vera* in edible coating formulation

Recently, *Aloe vera* has been used in the formulation of various cosmetic and food products (Simal *et al.*, 2000). It is used in beverages and ice creams and also applied as an edible coating (Martinez-Romero *et al.*, 2003). *Aloe vera* based edible coatings have been shown to reduce loss of moisture and firmness, control respiration rate and

maturation development, delay oxidative browning, and reduce microorganism proliferation on in fruits such as sweet cherry, table grapes and recterones (Valverde *et al.*, 2005; Matinez-Romero *et al.*, 2005 and Ahmed *et al.*, 2009).The use of *Aloe Vera* for star fruit coating has been studied by Mardiana *et al.*, (2008). The aim of this work was to investigate the ability of edible coating from *Aloe vera* to protect the minimally processed fruit from deterioration.

Arowora *et al.* (2013) showed the effects of *Aloe vera* coatings on quality characteristics of oranges stored under cold storage. They reported that *Aloe vera* helps to reduce weight loss, gives better texture and increase the shelf life.

Aloe vera coating in oranges resulted in decrease in weight loss, increase in titrability of acids and higher TSS. *Aloe vera* gel effectively preserved total phenolics content, vitamin C, catalase enzyme activity and also reduced decay index of fruits (kumar, 2014).

2.9 Honey

Honey is considered as natural preservative for foods. It contains several vitamins and minerals such as ascorbic acid, pantothenic acid, niacin, riboflavin, calcium, copper, iron, magnesium, manganese, phosphorus, potassium and zinc. Since ancient times, honey has been used for its healing, nutritional and therapeutic properties. It has good anti-inflammatory and antioxidant capacities. It has low moisture content. Currently, many researchers have reported the anti-bacterial activity of honey and found that honey has some broad-spectrum antibacterial activity when tested against pathogenic bacteria and food spoilage bacteria. Bogdanov *et al.* (2008) reported that the phenolic content of honey has antioxidant capacity which influence on several gram positive and negative bacteria.

Jeon and Zhao. (2005) studied the effect of honey on fresh cut apples and found that Honey helps to prevent enzymatic browning of fresh-cut apples during storage.

2.10 Insight from the review of literature

From the review literature it is observed that a lot of research work has been carried out on edible coating of whole fruit or fresh cut fruit and many more are going on to increase the shelf life of whole or fresh cut fruit. Therefore, the present research work was done to find the suitable formulation of *Aloe vera* and honey coating that will increase the shelf life of fresh cut pineapple with good nutritive value.



CHAPTER III

MATERIALS AND METHODS

CHAPTER III

MATERIALS AND METHODS

3.1 Experimental site

The study was conducted in the laboratory of Food Engineering & Technology and Food Processing & Preservation department under the Faculty of Engineering of Hajee Mohammad Danesh Science and Technology University, Dinajpur. The study was also conducted in the chemistry laboratory of this university.

3.2 Source of fruits and Coating materials

Fresh pineapples were purchased from local market of Dinajpur, Bangladesh. Fruits were selected for their uniformity in size, color and free from defects. *Aloe vera*, honey and packaging material were collected from local market. All analytical grade chemicals such as glycerol, methanol, glacial acetic acid, meta-phosphoric acid were supplied from food processing laboratory of Hajee Mohammad Danesh Science and Technology University, Dinajpur.

3.3 Preparation of samples and edible coating solutions

3.3.1 Preparation of pineapple

At first all containers, cutting board, knives and other utensils were washed and sanitized with 2% methanol solution. Then pineapples were washed gradually with tap water by hand and dried naturally. Then they were peeled manually and cut into cubes of 2 cm.

3.3.2 Preparation of *Aloe vera* gel

Aloe vera leaves were washed with water and peeled the leave skins. The pulps were separated and ground in a blender. Then the mixture of *Aloe vera* pulp was filtered to remove the fibres.

3.3.3 Process of making *Aloe vera* coating

At first 100 mL *Aloe vera* juice was taken in a beaker. Then 1% glycerol, 1.5% CMC, 1% acetic acid and 2% methanol were added to the *Aloe vera* juice and stirred until distributed evenly. Then the *Aloe vera* gel solution was heated at a temperature of 70°C for 45min. The gel solution was then cooled immediately at ambient temperature.

3.3.4 Process of making honey coating

100 mL honey was taken in a beaker and 1% glycerol, 1.5% CMC, 1% acetic acid, and 2% methanol were added and stirred for better mixing.

3.3.5 Process of making *Aloe vera* -honey coating

Aloe vera gel solution and honey mixed together at different proportion. Then 1% glycerol, 1.5% CMC, 1% acetic acid, and 2% methanol were added to the solution and stirred continuously. After mixing the solution was heated at a temperature of 70°C for and cooled at ambient temperature.

3.3.6 Pineapple coating process

The coating solution was taken in a beaker. The cube cut pineapples then dipped into the *Aloe vera*-honey coating solution with different concentrations of *Aloe vera* and honey for 2 minutes and then dried for 20 minutes. After drying the coated pineapples were packed using LDPE and stored refrigerated at 4°C for 12days.

3.3.7 Treatments

Control (T₀): untreated pineapple

Treatment 1 (T₁) : pineapple was coated with 100% *Aloe vera*

Treatment 2 (T₂) : pineapple was coated with 100% honey

Treatment 3 (T₃) : pineapple was coated with 50% honey and 50% *Aloe vera*

Treatment 4 (T₄) : pineapple was coated with 25% honey and 75% *Aloe vera*

3.4 Proximate composition of fresh pineapple

3.4.1 Determination of moisture content

Moisture content was determined according to the method described by Mantilla *et al.* (2013). In a clean, dry and pre-weighted crucible 15g sample was taken. Then it was transferred to oven and dried at 60°C for 10 hours. Then the sample was placed in desiccator for cooling and weighed.

Moisture content was calculated using following formula:

$$\% \text{ Moisture} = (W_1 - W_2) / W \times 100$$

Here,

W_1 = weight of sample with crucible

W_2 = weight of dried sample with crucible

W = weight of sample

3.4.2 Determination of protein

AOAC method (2000) was used with some modification to determine the protein content. Usually three stages are used to determine protein content. These stages are given below:

A. Digestion:

Sample (1g), selenium powder (1g), CuSO_4 (0.1g), K_2SO_4 (10g) were taken into a volumetric flask. Then 25ml of H_2SO_4 (conc.) was added. After that the volumetric flask was heated at 100°C for 3 hr and cooled for 20 minute at room temperature.

B. Distillation:

After digestion 300 ml of distilled water and 125 ml of 40% NaOH were added to the volumetric flask. 25 ml of 4% boric acid solution and 2-3 drops mixed indicator were taken in a conical flask. The volumetric flask was connected with one end of the condenser and the conical flask was connected with other end. The volumetric flask was heated continuously until the conical flask was filled to 150 ml.

C. Titration:

The conical flask was disconnected and was taken for titration. Titrated against 0.2 N of H_2SO_4 solutions. The end point was indicated by orange color.

i) Calculation for N_2 content:

$$\% \text{ of } \text{N}_2 = \text{burette reading} \times \text{normality of } \text{H}_2\text{SO}_4 \times \text{ml equivalent of } \text{N}_2$$

Here;

$$\text{Normality of } \text{H}_2\text{SO}_4 = 0.2$$

$$\text{ml equivalent of } \text{N}_2 = 1.4$$

ii) Calculation for protein content:

$$\% \text{ Protein} = \% \text{ of } \text{N}_2 \times \text{protein factor}$$

Here;

$$\text{Protein factor} = 6.25$$

3.4.3 Determination of fat

Fat content was determined by AOAC method (2000). At first pineapple (5g) was taken into the thimble. Then the thimble was set into the soxhlet apparatus containing 200 ml petroleum ether. The fat was extracted for 6 hours. After that petroleum ether was evaporated at 80°C.

Fat content was calculated by following formula:

$$\% \text{ Fat} = (W_1 - W_2) / W \times 100$$

Here,

W_1 = weight of evaporated flask with fat

W_2 = weight of empty flask

W = weight of sample

3.4.4 Determination of total ash

Total ash content was determined by AOAC method (1984). 5g of sample was taken in a clean, dry and pre-weighted crucible. Then the crucible was placed into muffle furnace at 550°C for 5.5 hours. Then it was cooled at desiccator and weighed.

The ash content was calculated by the following formula:

$$\% \text{ Ash} = (W_1 - W_2) / W \times 100$$

Here,

W_1 = weight of ash with crucible

W_2 = weight of empty crucible

W = weight of sample

3.4.5 Determination of carbohydrate

Total carbohydrate content of samples was calculated by difference, that the percentage of moisture, protein, fat and ash was subtracted from 100 (Pearson, 1976).

$$\text{Carbohydrate} = 100 - (\% \text{ Protein} + \% \text{ Fat} + \% \text{ Ash} + \% \text{ Moisture})$$

3.5 Fruit physico-chemical analysis

3.5.1 Juice leakage

The percent of juice leakage was determined by measuring the weight loss of sample throughout storage period. The weight of each sample was recorded on day 0 and throughout storage using a digital balance. The formula of calculation is as follows:

$$\text{juice leakage (g /100g)} = (W_1 - W_2) / W_1 \times 100$$

Where,

W_1 = initial weight

W_2 = final weight

3.5.2 Firmness

Firmness of samples was determined by using a penetrometer. The cut pineapple pieces were placed at the centre of the platform and the force applied by the blade to cut the pieces was measured. The puncture test was performed using a stainless steel probe with 3.5 mm of diameter. The texture was measured with maximum force (expressed in Kilo-Newton) required to achieve probe penetration at three different location in each sample. The obtained value was used to determine the firmness of the pineapple.

3.5.3 Color

Changes in pineapple color were measured by using a spectrophotometer ((Minolta Camera, Tokyo, Japan). The unit was calibrated using standard white and black plates. Means of 3 replications were used to determine the color coordinates, L^* (lightness), a^* (greenness), and b^* (yellowness). The equation used for calculation as follows:

$$\text{Hue angle (H)} = \tan^{-1}(b/a)$$

Where,

b = yellowness and

a = redness

3.5.4 Total soluble solids (TSS)

The total soluble solids (TSS) were measured by AOAC method 932.14 (AOAC, 1990) with a digital refractometer. In this case at first the refractometer was calibrated with

distilled water. Then 1 drop of sample (pineapple juice) was dropped to the light prism cell and the value was recorded.

3.5.5 pH

pH of pineapples were determined by AOAC method 981.12 (AOAC,1990) with a digital pH meter. At first the pH meter was calibrated with standard buffer solution. After calibration the sensor was dipped into the sample (pineapple juice) for 10 min and the obtained value was recorded.

3.5.6 Vitamin C

Vitamin C content of pineapple was measured using AOAC method 985.33 (2,6-Dichloroindophenol titrimetric method, AOAC,1998). At first 20g of pineapples were blended with 50 mL of metaphosphoric acideacetic acid solution. Then the solution was filtered with qualitative filter paper. Then 10 mL of the filtered solution was taken in a conical flask and titrated with 2, 6-dichloroindophenol standard solution .The titration volume was recorded and used to calculate vitamin C content of the sample (milligrams of ascorbic acid/g of sample, wet basis). The indophenol solution was standardized by an ascorbic acid standard solution (1 mg/mL) and sample blanks.

The equation as follows:

Vitamin C (mg/100gm) = (Titre value×dye factor× 100)/ weight of sample

Dye factor = 0.5/ standard titre

3.6 Microbial analysis

For total viable count of microorganism present in the samples (pineapple), Standard pour plate method was followed according to the method described in “Recommended method for the microbiological examination of food” (Ali, 2008).

3.6.1 Preparation of media

Composition of Agar media:

Ingredients -0.5gm,	Peptone -0.5gm
Agar -1.8gm	Beef extract -0.3gm
Sodium chloride (NaCl) -0.2gm	Distilled water -100ml

All necessary ingredients were measured with the help of electric balance and taken them in a conical flask and mixed. The conical flask was heated for proper mixing. In the time of heating, the mixture was rotted with the glass rod. When the mixture was properly mixed, the mouth of the conical flask was blocked with cotton plug and covered with aluminium foil. Then the conical flask with media was placed in autoclave for sterilization (Temperature: 121°c, Pressure: 15 Ib/inch² and time: 15 mins.).

3.6.2 Preparation of dilution blank

In order to dilute the sample consecutively 1ml of the original sample was diluted stepwise through a series of tubes containing 9ml of distilled water. At first 9ml of the distilled water was taken in a sterile test tubes and then 1ml of the original sample was taken to the first test-tube with a sterile pipette. Water with the sample was vigorously shaken for homogenous distribution of the bacterial population in the solution. This tube was denoted as “A”. From the tube “F-1” another 1ml aliquot was transferred to the second tube and this tube was denoted as “F-2”. In this way “F-3”, “F-4”, “F-5”, “F-6” was prepared until the desired dilution is achieved. Now the tube “F-1” has got the dilution 10⁻¹, 10⁻², 10⁻³, 10⁻⁴, 10⁻⁵, 10⁻⁶ respectively.

The dilutions were as follows:

Table 3.1: Dilution of sample

Tube No.	Dilution	Volume of original fluid per ml
1	1/10	0.1 or 10 ⁻¹
2	1/100	0.01 or 10 ⁻²
3	1/1,000	0.001 or 10 ⁻³
4	1/10,000	0.0001 or 10 ⁻⁴
5	1/100,000	0.00001 or 10 ⁻⁵
6	1/1,000,000	0.000001 or 10 ⁻⁶

3.6.3 Procedure of plating

Now from the test-tube “F-1”, 1ml of the sample solution was taken in a sterile petridish containing 9ml of agar medium. The agar with bacterial sample was mixed by rotating the petridish. This petridish was marked as “A”. In this way “B”, “C”, “D”, “E”, “F” marked petridishes were prepared from the tubes “F-2”, “F-3”, “F-4”, “F-5” and “F-6” respectively. Then these petridishes were placed on a level surface for few minutes for solidifying the agar medium.

3.6.4 Incubation and colony count

After solidification petridishes were placed in the incubator at 37⁰C for 48 hours, the over loaded petridishes were avoided and the petridishes containing countable colony were selected. Colonies were counted with the aid of a magnifying glass and finally the total number of bacteria per gram of sample was calculated by the following equation:

Colony count (per ml) = Number of colonies (per plate) × Reciprocal of the dilution.

Moreover, it also can be calculated as

$$\text{Mean count of sample (cfu/g)} = \frac{\text{No of colony} \times \text{Reciprocal of dilution}}{\text{sample taken}}$$

The bacteria count was determined by diluting the samples serially and plating 0.1 ml aliquot on nutrient agar. Plate count agar was used and incubated at 37°C for 48 hours using pour plate count methods after which colonies were counted.

3.7 Sensory analysis

The sensory evaluation was performed to evaluate the taste, flavor, color and overall acceptance of coated fresh cut pineapples in comparison with uncoated by using the Hedonic scale suggested by Krum (1955). Ten panelists were selected for this purpose. Panelists were asked to rate the difference between samples by giving the numbers from 0-9, where 0-2 represent dislike extremely, 3-5 for dislike, 6-8 for good and 9 for excellent aroma, taste and flavor.

3.8 Statistical analysis

Statistical analysis was performed by using SPSS 11.5 version to test the significant difference between the quality of coated and uncoated pineapple during storage. Significant differences were measured by one-way ANOVA while Duncan Multiple Range Test (DMRT) was used to compare differences between treatments with ($P < 0.05$).



CHAPTER IV

RESULTS AND DISCUSSION

CHAPTER IV

RESULTS AND DISCUSSION

4.1 Proximate analyses

Table 4.1 shows the proximate composition of fresh pineapple, *Aloe vera* and honey. In this study, it was observed that the moisture, protein, fat, ash, carbohydrate and total soluble solid (TSS) content of fresh pineapple were 86.80%, 0.3%, 0.16%, 0.97%, 11.73% and 11.56 °Brix respectively which was nearly similar as reported by Sayka *et al.* (2014). They found 86.2% moisture, 0.92% ash and 11°Brix total soluble solid (TSS) in fresh pineapple. The carbohydrate content of fresh pineapple was similar to Jahan *et al.* (2011), who found 10.74% carbohydrate in fresh pineapple. However, in case of *Aloe vera* the moisture content, protein, carbohydrate and ash were 96.7%, 0.14%, 0.5% and 0.12% respectively. This result is in agreement with Karina *et al.* (2012) who found the moisture content, protein, carbohydrate and ash were 98.23%, 0.20%, 0.63% and 0.15% respectively. On the other hand for honey the moisture content, protein and carbohydrate were 26.13%, 0.56% and 72.76% respectively which was nearly similar to Buba *et al.* (2013). The variation in result might due to instrumental error and difference in variety of *Aloe vera* and pineapple.

Table 4.1: Proximate composition of pineapple, *Aloe vera* and honey

Sample	Proximate composition (%)					
	Moisture	Protein	Fat	Carbohydrate	Ash	TSS
Pineapple	86.80±0.74	0.3±0.07	0.16±0.04	11.73±1.4	0.97±0.03	11.56±0.28
<i>Aloe vera</i>	96.7±0.5	0.14±0.03	-	0.5±0.02	0.12±0.04	1.2±0.01
Honey	26.13±0.02	0.56±0.04	0.20±0.01	72.76±0.74	0.35±0.02	71±0.34

*Mean ± standard deviation

4.2 Physico-chemical properties

4.2.1 Juice leakage

Figure 4.1 shows the effect of coating on juice leakage during storage period of fresh cut pineapple. In this study it was found that the amount of juice leakage was higher in uncoated sample than coated. The percentage of juice leakage of uncoated sample increased from 5.02% to 28.46%. Among all coating formulations the minimum juice leakage was observed in 100% *Aloe vera* coating and the value varies from 2.95% to 14.13%. This reduction in percentage of juice leakage was probably for coating which act as a semi-permeable barrier against oxygen, carbon-dioxide, moisture and solute movement, thereby reducing respiration, water loss and oxidation reaction rates (Baldwin *et al.*, 1999 and Park, 1999). Similar result was found by Arowora *et al.* (2013) who worked on orange coated with *Aloe vera*. The reduction in percentage of juice leakage may also be due to lower temperature (Kasim and Kasim, 2009).

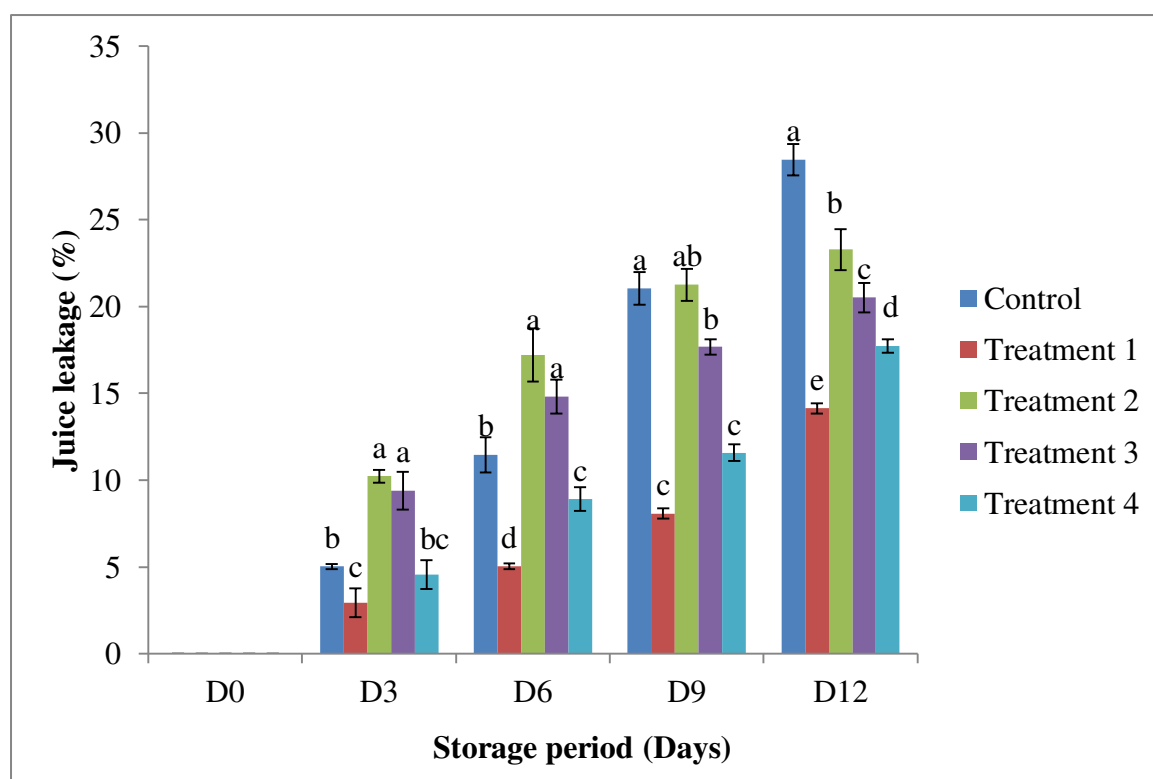


Figure 4.1: Effect of *Aloe vera* and honey coating on juice leakage during refrigerated storage

* Abbreviation of Control, Treatment 1, Treatment 2, Treatment 3, Treatment 4 are given in 3.3.7

4.2.2 Firmness

Firmness is an important quality parameter for the consumer acceptability of fresh fruit and vegetables and it is related to water content and metabolic changes. Fruits become softer during storage for prolonged times. In this study the firmness of coated and uncoated fresh cut pineapples was observed and found that the firmness was better in coated than uncoated fresh cut pineapple (Table 4.2). This experiment showed that edible coating of 100% *Aloe vera* and combination of (75% *Aloe vera* + 25% honey) coating formulation provided better firmness among other coating formulation. In case of 100% honey and combination of (50% *Aloe vera* + 50% honey), the firmness was not good because of high amount of moisture removal. Similar result was given by Rojas-Grau *et al.* (2008) and Oms-Oliu *et al.* (2008). They reported that gellan and alginate-based edible coatings had beneficial effect in maintaining the firmness of fresh cut apples and melons. In addition Martinez–Romero *et al.* (2006) found that *Aloe vera* gel has a power to lower the weight loss which also influences the retarding of firmness.

Table 4.2: Effect of *Aloe vera* and honey coating on firmness during refrigerated storage

Treatment (N)	Day				
	D ₀	D ₃	D ₆	D ₉	D ₁₂
T ₀	6.15±0.35 ^a	4.75±0.77 ^a	3.6±0.70 ^a	2.54±0.36 ^a	2.65±0.63 ^c
T ₁	6. ±0.28 ^a	5.9±0.42 ^a	5.8±0.42 ^a	5.55±0.49 ^a	5.1±0.84 ^a
T ₂	5.8±0.42 ^a	4.95±0.21 ^a	4.35±1.62 ^a	4.1±2.54 ^a	3.85±0.35 ^{abc}
T ₃	5.75±1.06 ^a	5.2±0.42 ^a	4.2±1.27 ^a	4.75±0.77 ^a	3.5±0.28 ^{bc}
T ₄	6.25±0.35 ^a	5.05±0.91 ^a	4.9±0.28 ^a	5.1±0.42 ^a	4.85±0.21 ^{ab}

*Mean ± standard deviation values (n = 3) followed by a different lower-case letter within the same column are significantly different ($P < 0.05$) by Duncan's multiple range test.

* Abbreviation of T₀, T₁, T₂, T₃, T₄ are given in 3.3.7

4.2.3 Total soluble solids (TSS)

Figure 4.2 shows the total soluble solid (TSS) content of coated and uncoated fresh cut pineapples. It was observed that the total soluble solid content was increased gradually with time in coated and uncoated pineapple. However, coating with 100% *Aloe vera* and combination of (25% *Aloe vera* + 75% honey) formulation was more effective in reducing TSS compared to other coating formulation. In case of 100% honey and combination of (50% *Aloe vera* + 50% honey) formulation, there was no significant difference with uncoated pineapple. This result was found because pineapple is a non-climacteric fruit and its sugar content does not change drastically during its shelf-life

(Paull and Chen, 2003). Same results were reported by Brasil *et al.* (2012) who studied on fresh-cut papaya with a chitosan-based coating. In addition Mali *et al.* (2003) reported that *Aloe vera* coating could provide similar effects as modified atmosphere packaging (MAP).

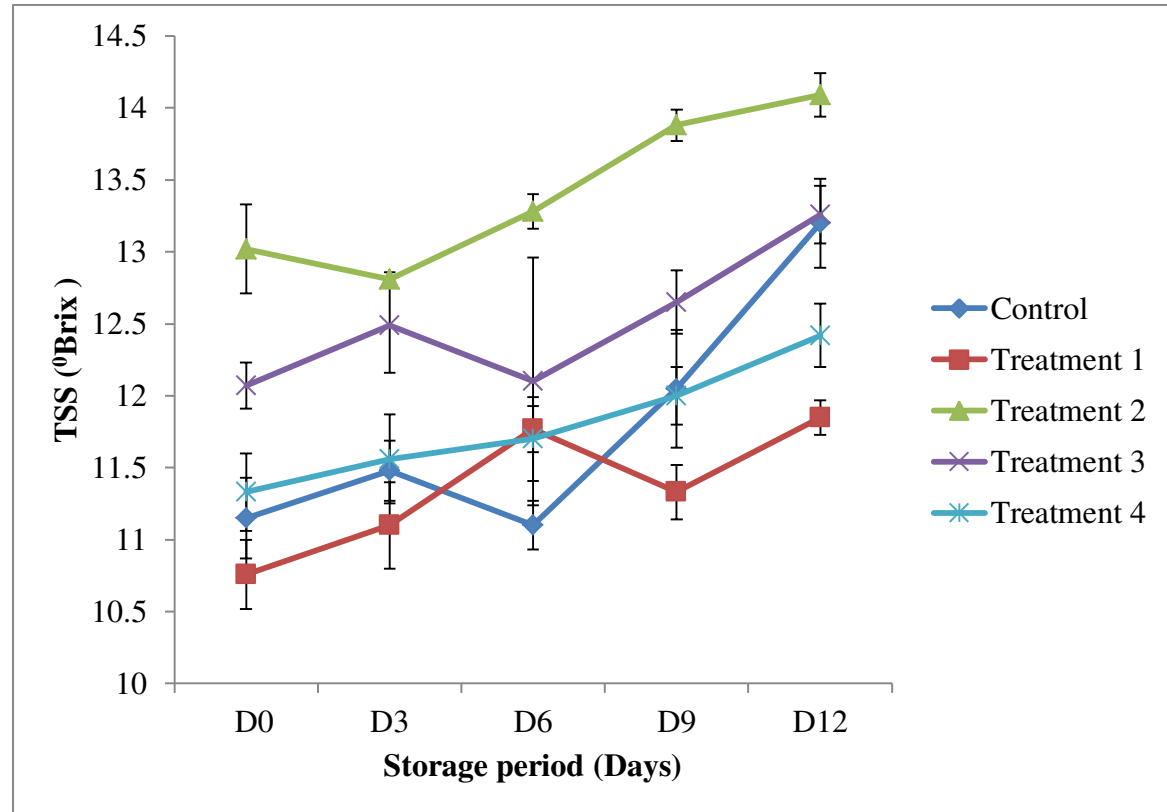


Figure 4.2: Effect of *Aloe vera* and honey coating on TSS during refrigerated storage

* Abbreviation of Control, Treatment 1, Treatment 2, Treatment 3, Treatment 4 are given in 3.3.7

4.2.4 pH

The pH value of fresh cut pineapple increased significantly in both coated and uncoated pineapple during storage at 4°C. The value is higher in uncoated sample while lower in coated sample. The result in the Table 4.3 showed that at the end of storage period the pH of coated pineapples were 3.60, 3.58, 3.54, and 3.62 respectively while in uncoated it was 3.71. Similar results were reported by Mantilla *et al.* (2013) who studied the effect of multilayered antimicrobial edible coating on fresh cut pineapples storage. Coating was effective in delaying compositional changes of fresh cut pineapple. It was also reported by El-Ghaouth *et al.* (1991) and Garcia *et al.* (1998) that the decrease of acidity during storage resulted in fruit senescence. Biochemical reaction between fruits and coating material, slower rate of respiration and metabolic activity are common reasons behind the

change in pH (Jitareerat *et al.*, 2007). In addition, the increase in pH may be due to the breakup of acids with respiration during storage (Pesis *et al.*, 1999).

Table 4.3: Effect of *Aloe vera* and honey coating on pH during refrigerated storage

Treatment	Day				
	D ₀	D ₃	D ₆	D ₉	D ₁₂
T ₀	3.54±0.04 ^a	3.6±0.01 ^{ab}	3.65±0.05 ^a	3.5±0.02 ^b	3.71±0.03 ^a
T ₁	3.56±0.05 ^a	3.71 ±0.03 ^a	3.66 ±0.06 ^a	3.77±0.05 ^a	3.60±0.04 ^{ab}
T ₂	3.53±0.04 ^a	3.47±0.06 ^b	3.57±0.09 ^a	3.79±0.01 ^a	3.58±0.06 ^{ab}
T ₃	3.49±0.02 ^a	3.51±0.05 ^b	3.55±0.04 ^a	3.53±0.08 ^b	3.54±0.02 ^b
T ₄	3.51±0.01 ^a	3.53±0.04 ^b	3.68±0.05 ^a	3.75±0.03 ^a	3.62±0.07 ^{ab}

*Mean ± standard deviation values (n = 3) followed by a different lower-case letter within the same column are significantly different ($P < 0.05$) by Duncan's multiple range test.

* Abbreviation of T₀, T₁, T₂, T₃, T₄ are given in 3.3.7

4.2.5 Vitamin C

Figure 4.3 shows the effect of *Aloe vera* and honey coating on vitamin C content of fresh cut pineapple during storage. In this study, the vitamin C content of fresh pineapple was found 37.46 mg/100gm which was nearly similar to Masamba *et al.* (2013), who found 38.7mg/100gm vitamin C in fresh pineapple. It was observed that the loss of vitamin C was 21.92 and 10.92 mg/100gm in control sample and 100% *Aloe vera* coated sample respectively. It was also found that all coating formulation helps to retain vitamin C but among them higher concentration of *Aloe vera* gel solution was more effective to reduce the loss of vitamin C content of fresh cut pineapple. This finding was supported by Adetunji *et al.* (2014) who reported that *Aloe vera* gel coating possessed low oxygen permeability which lowered the activity of the enzymes and prevented oxidation of ascorbic acid. Similar results were found by Bierhals *et al.* (2011) who studied with fresh-cut pineapple where the vitamin C content decreased significantly upon storage.

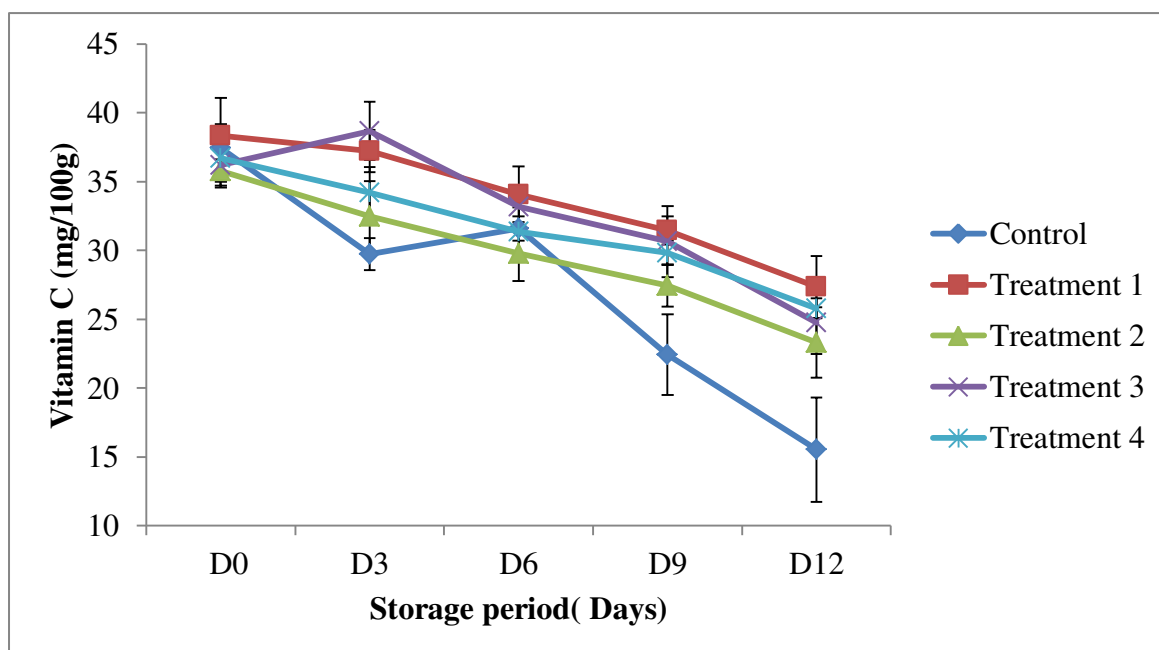


Figure 4.3: Effect of *Aloe vera* and honey coating on vitamin C during refrigerated storage

* Abbreviation of Control, Treatment 1, Treatment 2, Treatment 3, Treatment 4 are given in 3.3.7

4.2.6 Color

Color is an important quality parameter for fresh cut pineapple. In this study the color of coated and uncoated fresh cut pineapples were observed by measuring the lightness (L^*), yellowness (a^*), redness (b^*) and hue angle (H) during 12 days of storage at 4°C. Table 4.4 shows the changes of color of fresh cut pineapples during storage. Lightness of all sample both coated and uncoated decreased with the increasing of storage time. At the end of storage period, it was shown that the lightness of fresh cut pineapple coated with 100% *Aloe vera* had significantly higher L value compared to other coated and uncoated pineapples. From Table 4.3, it was observed that a^* values of uncoated pineapple increased upon storage while in all coated pineapple it decreased. The b^* values of coated sample varied less than uncoated at 12th day. It was observed that at the end of storage period the b^* value of 100% *Aloe vera* coating was nearly similar to that of initial day. This observed result was similar to Mantilla *et al.* (2013). However, small changes in a^* and b^* values are good indication of absence of oxidative browning of fresh pulp (Rocha & Moraes, 2000). In case of hue angle it was shown that, in control sample it decreased with time while in coated sample it increased. This increasing tend of hue angle in coated sample indicated less redness and more yellowness. Chauhan *et al.* (2006) also reported the same trend in increasing of hue angle.

Table 4.4: Effect of *Aloe vera* and honey coating on color during refrigerated storage

Treatment	Day				
	D ₀	D ₃	D ₆	D ₉	D ₁₂
L value					
T ₀	53.96±3.59 ^a	48.16±6.71 ^a	49.76±5.15 ^a	50.30±1.7 ^a	43.19±8.02 ^b
T ₁	59.92±1.06 ^a	55.82±2.14 ^a	45.79±1.56 ^a	52.91±7.61 ^a	57.77±6.47 ^a
T ₂	55.01±2.26 ^a	53.87±4.9 ^a	51.59±6.46 ^a	47.67±0.77 ^a	54.71±2.89 ^{ab}
T ₃	59.99±3.49 ^a	47.3±3.44 ^a	54.83±5.11 ^a	51.39±2.89 ^a	52.32±1.55 ^{ab}
T ₄	57.31±0.79 ^a	48.27±3.13 ^a	53.82±1.89 ^a	54.62±5.16 ^a	53.65±2.48 ^{ab}
a value					
T ₀	1.76±0.15 ^a	1.91±0.13 ^a	1.66±0.24 ^a	2.05±0.09 ^a	2.20±0.34 ^a
T ₁	1.43±0.31 ^a	1.19±0.17 ^a	1.25±0.47 ^{ab}	0.64±0.12 ^b	0.70±0.16 ^b
T ₂	1.54±0.55 ^a	1±0.62 ^a	1.13±0.29 ^{ab}	0.83±0.37 ^b	0.91±0.12 ^b
T ₃	1.48±0.62 ^a	1.15±0.30 ^a	0.74±0.24 ^b	0.93±0.29 ^b	0.63±0.34 ^b
T ₄	1.29±0.12 ^a	1.51±0.45 ^a	0.81±0.28 ^{ab}	0.59±0.39 ^b	0.78±0.19 ^b
b value					
T ₀	23.27±0.96 ^a	22.72±1.44 ^a	23.65±5.31 ^a	24.89±3.96 ^a	15.51±5.19 ^b
T ₁	24.37±0.88 ^a	20.91±2.75 ^a	20.35±0.79 ^a	25.44±0.82 ^a	25.54±1.89 ^a
T ₂	25.92±2.10 ^a	19.22±5.14 ^a	18.15±0.50 ^a	21.54±5.28 ^a	21.34±0.83 ^{ab}
T ₃	24.61±2.26 ^a	21.07±0.13 ^a	20.86±1.48 ^a	21.5±4.86 ^a	22.48±3.71 ^{ab}
T ₄	24.26±0.43 ^a	27.19±2.85 ^a	23.5±1.13 ^a	21.41±2.36 ^a	21.94±1.72 ^{ab}
Hue angle					
T ₀	85.65±0.55 ^a	85.15±0.64 ^a	85.93±0.30 ^a	85.24±0.53 ^b	81.23±4.13 ^b
T ₁	86.64±0.62 ^a	86.73±0.05 ^a	86.49±1.18 ^a	88.54±0.33 ^a	88.39±0.48 ^a
T ₂	86.62±0.95 ^a	86.63±2.76 ^a	86.44±0.84 ^a	87.83±0.46 ^a	87.54±0.43 ^a
T ₃	86.6±1.14 ^a	86.86±0.80 ^a	87.93±.81 ^a	87.37±1.38 ^{ab}	88.28±1.16 ^a
T ₄	86.94±0.22 ^a	86.84±0.63 ^a	88.04±0.59 ^a	88.34±1.21 ^a	87.92±0.66 ^a

*Mean ± standard deviation values (n = 3) followed by a different lower-case letter within the same column are significantly different ($P < 0.05$) by Duncan's multiple range test.

* Abbreviation of T₀, T₁, T₂, T₃, T₄ are given in 3.3.7

4.3 Microbiological analysis

The effect of *Aloe vera* and honey based coating on microbial growth during storage of fresh cut pineapple is shown in Table 4.5. The total microbial count of uncoated pineapple increased from 1.84×10^3 to 5.38×10^6 CFU/g on day 12th of storage. All the coating formulation with different concentration of *Aloe vera* and honey effectively inhibited the microbial growth during storage. The results were similar with the results of Campaniello *et al.* (2008) and Simoes *et al.* (2009), who reported that the chitosan coating on strawberries and carrot sticks, respectively, could inhibit the growth of microorganisms. In addition, Antonioli *et al.* (2006) and Sperber and Doyle (2009) reported that the lower temperature could retard microbial growth and thus lower the respiration rate and provides a longer shelf-life of fresh cut products.

Table 4.5: Effect of *Aloe vera* and honey coating on microbial growth during refrigerated storage

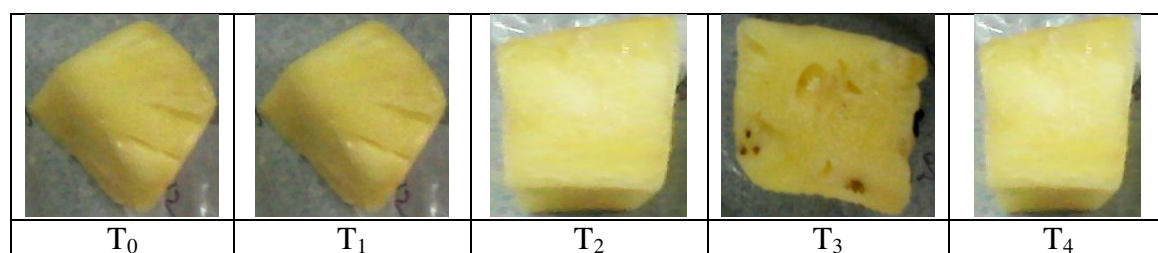
Treatment	Day				
	D ₀	D ₃	D ₆	D ₉	D ₁₂
T ₀	1.84×10^3	3.28×10^3	4.21×10^5	4.74×10^4	5.38×10^6
T ₁	2.13×10^4	1.6×10^4	2.21×10^3	2.52×10^4	2.77×10^4
T ₂	2.09×10^3	2.12×10^3	2.82×10^4	2.68×10^4	2.82×10^5
T ₃	1.9×10^3	2.79×10^2	2.1×10^5	2.73×10^3	3.53×10^4
T ₄	2.24×10^4	1.74×10^5	2.41×10^3	2.51×10^2	2.64×10^3

* Abbreviation of T₀, T₁, T₂, T₃, T₄ are given in 3.3.7

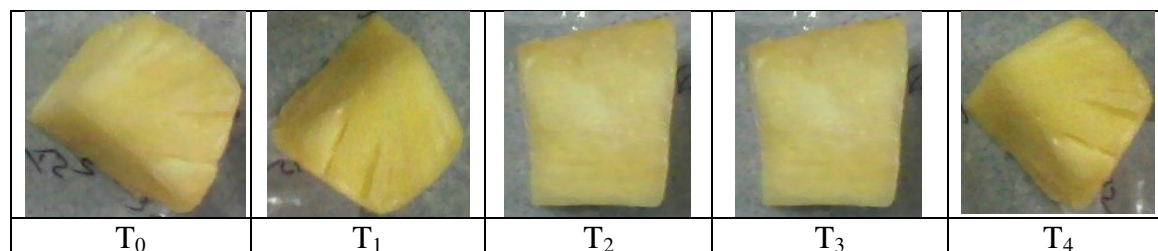
4.4 Sensory evaluation

As shown in Table 4.6, data indicated the changes in sensory attributes such as color, flavor, firmness, taste and overall acceptability of coated pineapple during 12 days of storage.

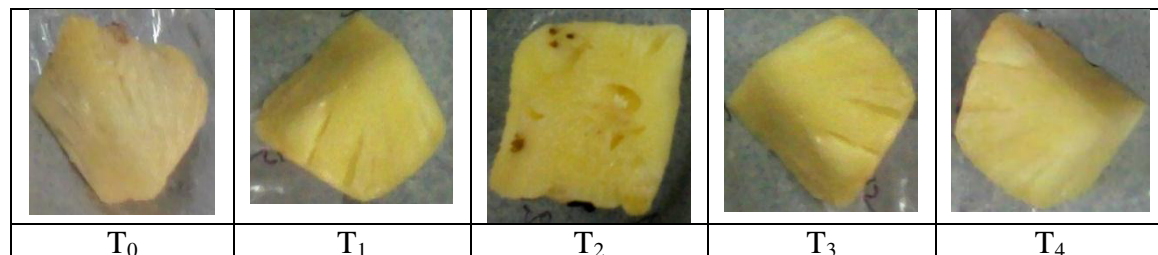
Sensory Evaluation of Pineapple



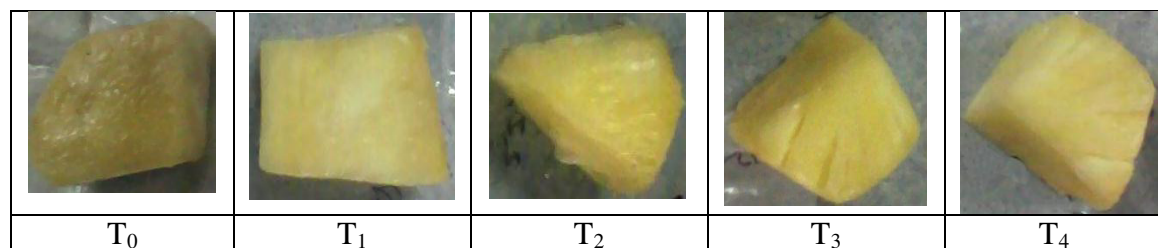
Day 0



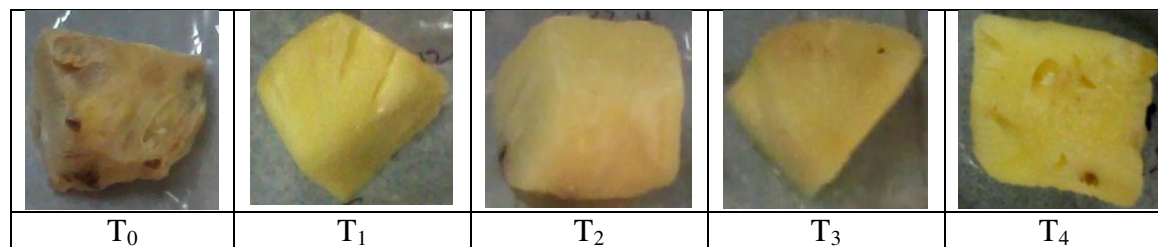
Day 3



Day 6



Day 9



Day 12

Figure 4.4 Sensory Evaluation of Pineapple

* Abbreviation of T₀, T₁, T₂, T₃, T₄ are given in 3.3.7

Table 4.6: Effect of *Aloe vera* and honey coating on sensory attributes at 12th day of storage

Treatment	Physiological character				
	Color	Flavor	Texture	Taste	Overall acceptability
T ₁	7.2±0.78 ^a	6.8±1.03 ^{ab}	7.3±0.82 ^a	6.5±1.08 ^a	6.1±1.10 ^{ab}
T ₂	6.3±0.94 ^b	5.9±0.99 ^c	5±0.81 ^b	5.2±0.91 ^b	5.3±0.94 ^b
T ₃	6.9±0.73 ^{ab}	6±0.94 ^{bc}	5.7±0.82 ^b	5.6±0.96 ^b	5.5±0.84 ^b
T ₄	7.5±0.84 ^a	6.9±0.56 ^a	6.7±0.94 ^a	7±0.81 ^a	6.5±1.08 ^a

*Mean ± standard deviation values (n = 3) followed by a different lower-case letter within the same column are significantly different ($P < 0.05$) by Duncan's multiple range test.

* Abbreviation of T₀, T₁, T₂, T₃, T₄ are given in 3.3.7

4.4.1 Color

For color preference a one way of analysis of variance ANOVA was carried out and it was found that at 12th day of storage there was significant difference in color of coated pineapple ratings by the panelists (Table 4.6) coated with different concentration of *Aloe vera* and honey. Color is an important parameter for perception. Low color ratings of sample can decrease the consumer acceptability. The color rating was higher in 100% *Aloe vera* and combination of (75% *Aloe vera* + 25% honey) coating of fresh cut pineapples.

4.4.2 Flavor

Flavor is also an important attribute for consumer acceptability. For flavor preference analysis of variance (ANOVA) showed that there was significant difference among all coating formulations. At 12th day of storage the control sample was given off flavor while in coated sample there was no off-flavor. From table 4.6 it was found that 100% *Aloe vera* and combination of (75% *Aloe vera* + 25% honey) were more accepted by panelists because of its good flavor.

4.4.3 Texture

Texture is another important parameter for consumer acceptance. From Table 4.5 significant difference was found among all coated sample. Panelists gave higher ratings for 100% *Aloe vera* coated pineapple and combination of (75% *Aloe vera* +25% honey)

coating. In case of 100% honey and combination of (50% *Aloe vera* +50% honey) coating the texture of pineapple was not so good.

4.4.4 Taste

The taste acceptability of coated pineapple ranged from 5.2 to 7.0. There was significant difference in taste of pineapple coated with different concentration of *Aloe vera* and honey (Table 4.6). Panelist preferred the 100% *Aloe vera* coated pineapples most than other coating formulations.

4.4.5 Overall acceptability

It was apparent from the results of analysis of variance ANOVA showed that there was significant ($p < 0.05$) difference in overall acceptability among the coated pineapple. Table 4.6 shows that the ratings of honey coated pineapples were lower because of its color, texture, flavor was not good. On the other hand, *Aloe vera* coated pineapple liked most by the panelists at 12th day of storage because of its good color, flavor, texture and taste.



CHAPTER V

SUMMARY AND CONCLUSION

CHAPTER V

SUMMARY AND CONCLUSION

The experiment was studied on fresh cut pineapple coated with *Aloe vera* and honey with the objectives to document the physical and chemical properties, proximate composition, nutritional quality and shelf life. Though the annual yield of pineapple production is high but due to post-harvest loss, a large portion of pineapple damaged. To minimize post-harvest loss the experiment was done carefully.

The prepared fresh cut pineapples were very good in appearance at first day. During storage period the quality was deteriorated at increasing rate in control sample. The edible coatings based on *Aloe vera* and honey was effectively used for maintaining the quality and extending the shelf life of fresh cut pineapples in this study. This coating seemed to have a beneficial impact on quality parameters of fresh cut pineapple by reducing juice leakage, pH, total soluble solids, microbial growth and color changes. In addition, it was effective in retaining firmness and vitamin C during storage. At the end of the storage period sensory perception indicated the good quality of the treated product compared with the non-treated product. However significant differences were observed among the coating formulation. The result showed that among all coating formulations, 100% *Aloe vera* provided better texture, flavor, and color. Finally, it can be concluded that among all the formulation 100% *Aloe vera* coating was more effective in increasing the shelf life of fresh cut pineapple with good nutritive value.



REFERENCES

REFERENCES

- Adetunji C.O., Fadiji A.E. and Aboyeji O.O. (2014). Effect of chitosan coating combined *Aloe vera* gel on cucumber (*Cucumis Sativa* L.) post-harvest quality during ambient storage. *Journal of Emerging Trends in Engineering and Applied Sciences*, 5(6): 391-397.
- Adetunji C.O., Fawole O.B., Arowora K.A., Nwaubani S.I., Ajayi E.S., Oloke J.K., Majolagbe O.M., Ogundele B.A., Aina J.A., and Adetunji J.B. (2012). Effects of edible coatings from *Aloe vera* gel on quality and postharvest physiology of *Ananas Comosus* (L.) fruit during ambient storage. *Global Journal of Science Frontier Research Bio-Tech & Genetics*, 12(5): 39-43.
- Adetunji C.O. (2008). The antibacterial activities and preliminary phytochemical screening of *vernonia amygdalina* and *Aloe vera* against some selected bacteria. MS Thesis. University of Ilorin, 40-43.
- Ahmed M.J., Singh Z. and Khan A.S. (2009). Postharvest *Aloe vera* gel-coating modulates fruit ripening and quality of 'Arctic Snow' nectarine kept in ambient and cold storage. *International Journal of Food Science & Technology*, 44(5): 1024-1033.
- Ali M.A. (2008). Study on the shelf-life of parboiled rice, MS Thesis, Department of Food Technology and Rural Industries, Bangladesh Agricultural University, 36-42.
- AMS [U.S. Department of Agriculture, Agricultural Marketing Service]. (1998). Quality through verification program for the fresh-cut produce industry. Federal Register 63:47220-47224.
- Anonymous. (1998). Food and Drugs. Food labeling. Code of Federal Regulations, 21CFR101.95.
- Antoniolli L.R., Benedetti B.C., Sigrist J.M.M., Filho M. and Alves R.E. (2006). Metabolic activity of fresh-cut Perola pineapple as affected by cut shape and temperature. *Brazilian Journal of Plant Physiology*, 18(3):413-417.

- AOAC. (1984). Official Methods of Analysis, Association of Official Analytical Chemists. (Fourteenth edition). Washington DC: AOAC International.
- AOAC. (1990). Official method 981.12. pH of acidified foods. Official methods of analysis (Thirteenth edition). Washington DC: AOAC International.
- AOAC. (1998). Official Method 985.33. Vitamin C (reduced ascorbic acid) in ready-to-feed milk-based infant formula- 2,6-dichloroindophenol titrimetric method. Official methods of analysis (Sixteenth Edition). Washington DC: AOAC International.
- AOAC. (2000). Official Methods of Analysis. Association of Official Analytical Chemists. Washington DC: AOAC International.
- Arowora K.A., Williams J.O., Adetunji C.O., Fawole O.B., Afolayan S.S., Olaleye O.O., Adetunji J.B. and Ogundele B.A. (2013). Effects of *Aloe vera* coatings on quality characteristics of oranges stored under cold storage. *Greener Journal of Agricultural Sciences*, 3(1): 039-047.
- Arvanitoyannis I. and Gorris L.G.M. (1999). Edible and biodegradable polymeric materials for food packaging or coating in processing foods: Quality optimization and process assessment. *CRC Press, Boca Raton, FL.*, 357-371.
- Azarakhsh N., Osman A., Ghazali H.M., Tan C.P. and Mohd Adzahan N. (2012). Optimization of alginate and gellan-based edible coating formulations for fresh-cut pineapples. *International Food Research Journal*, 19(1): 279-285.
- Baldwin E.A., Burns J.K., Kazokas W., Brecht J.K., Hagenmaier R.D., Bender R.J. and Pesis E. (1999). Effect of two edible coatings on mango (*Mangifera indica* L.) ripening during storage. *Postharvest Biology and Technology*, 17: 215-226.
- Barth M. (2000). Current status of the fresh-cut produce industry and future directions. *IFT Annual Meeting, New Orleans, LA. Abstract*, 71–6.
- BBS. (2012). Yearbook of Agricultural Statistics of Bangladesh, Bureau of Statistic, Ministry of Planning, Govt. of Peoples Republic of Bangladesh, Dhaka.
- BBS. (2013). Report on The Productivity Survey of Pineapple Crop. Bureau of Statistic, Ministry of Planning, Govt. of Peoples Republic of Bangladesh, Dhaka.

- Bierhals V. S., Chiumarelli M. and Hubinger M. D. (2011). Effect of Cassava starch coating on quality and shelf-life of fresh-cut pineapple (*Ananas comosus* L. Merrill cv “Perola”). *Journal of Food Science*, 76: 62-71.
- Bogdanov S., Jurendic T., Sieber R. and Gallmann P. (2008). Honey for Nutrition and Health: a Review. *American Journal of the College of Nutrition*, 27: 677-689.
- Brasil I.M., Gomes C., Puerta-Gomez A., Castell-Perez M.E. and Moreira R.G. (2012). Polysaccharide-based multilayered antimicrobial edible coating enhances quality of fresh-cut papaya, *LWT - Food Science and Technology*, 47:39-45.
- Buba F., Gidado A. and Shugaba A. (2013). Analysis of biochemical composition of honey samples from North-East Nigeria. *Biochemistry & Analytical Biochemistry*.
- Campaniello D., Bevilacqua A., Sinigaglia M. and Corbo M.R. (2008). Chitosan antimicrobial activity and potential applications for preserving minimally processed strawberries. *Food Microbiology*, 25(8): 992-1000.
- Chantanawarangoon S. and Kader A.A. (2002). Calcium treatments and modified atmospheres extended post-cutting life of mango cubes. *Presented at the Institute of Food Technologists "Annual Meeting and Food Exposium" Anaheim, CA*.
- Chauhan O.P., Raju P.S., Shylaja R., Dasgupta D.K. and Bawa A.S. (2006). Synergistic effects of modified atmosphere and minimal processing on the keeping quality of pre-cut papaya (*Carica papaya* L.). *The Journal of Horticultural Science and Biotechnology*, 81:903–909.
- Di Egidio V., Sinelli N., Limbo S., Torri L., Franzetti L. and Casiraghi E. (2009). Evaluation of shelf-life of fresh-cut pineapple using FT-NIR and FT-IR spectroscopy. *Postharvest Biology and Technology*, 54: 87-92.
- Drake S.R., Fellman J.K. and Nelson J.W. (1987). Postharvest use of sucrose polyesters for extending the shelf- life of stored ‘Golden Delicious’ apples. *Journal of Food Science*, 52(5): 1283-1285.
- El Ghaouth A., Arul J., Ponnampalam R. and Boulet M. (1991). Use of chitosan coating to reduce water-loss and maintain quality of cucumber and bell pepper fruits. *Journal of Food Processing and Preservation*, 15: 359-368.

- El Ghaouth A., Arul J., Ponnampalam R., and Boulet M. (1992b). Chitosan coating to extend the storage life of tomatoes. *Horticulture Science*, 27(9): 1016-1018.
- Esua M.F. and Rauwald J.W. (2006). Novel bioactive maloylglucans from *Aloe vera* gel: isolation, structure elucidation and invitro bioassays. *Carbohydrate Research*, 341: 355–364.
- Eshun K. and He Q. (2004). *Aloe vera*: A valuable ingredient for the food, pharmaceutical and cosmetic industries—A review. *Critical Reviews in Food Science and Nutrition*, 44: 91–6.
- Garcia M.A., Martino M.N. and Zaritzky N.E. (1998). Plasticized starch-based coatings to improve strawberry quality and stability. *Journal of Agricultural and Food Chemistry*, 46: 3758- 3767.
- Gazi T.S.M. (2013). A comparative economic analysis of pineapple mono crop and pineapple intercrops of Madhupur area in Tangail district of Bangladesh. MS Thesis, Department of Agricultural economics, Bangladesh Agricultural University, Mymensingh.
- Grassmann J., Hippeli S. and Elstner E.F. (2002). Plant's defence and its benefits for animals and medicine: role of phenolics and terpenoids in avoiding oxygen stress. *Plant Physiology and Biochemistry*, 40: 471 – 478.
- Grau M. (2008). Using polysaccharide-based edible coatings to maintain quality of fresh-cut Fuji apples. *LWT*, 41: 139-147.
- Gunes G. and Lee C.Y. (1997). Color of minimally processed potatoes as affected by modified atmosphere packaging and antibrowning agents. *Journal of Food Science*, 62: 572-575.
- Gunes G., Watkins C.B., and Hotchkiss J.H. (2001). Physiological responses of fresh cut apples slice under high CO₂ and low O₂ partial pressures. *Postharvest Biology and Technology*, 22: 197-204.
- Hassan M.K., Chowdhury B.L.D. and Akhter N. (2010). National Food Policy Capacity Strengthening Programme, 118-126.

- Hossain M.A. and Rahman S.M.M. (2011). Total phenolics, flavonoids and antioxidant activity of tropical fruit pineapple. *Food Research International*, 44(3): 672-676.
- Jahan S., Gosh T., Begum M. and Saha B.K. (2011). Nutritional profile of some tropical fruits in Bangladesh: specially anti-oxidant vitamins and minerals. *Bangladesh Journal of Medical Science*, 10(2).
- Jeon M. and Zhao Y. (2005). Honey in combination with vacuum impregnation to prevent enzymatic browning of fresh-cut apples. *International Journal of Food Science and Nutrition* , 56(3):165-76.
- Jitareerat P., Paumchai S. and Kanlayanarat S. (2007). Effect of chitosan on ripening enzymatic activity and disease development in mango (*Mangifera indica* L.) fruit. *New Zealand Journal of Crop and Horticultural Science*, 35: 211-218.
- Karina Di Scala, Antonio Vega-Gálvez, Kong Ah-Hen, Yissleen Nuñez-Mancilla, Gipsy Tabilo-Munizaga, Mario Pérez-Won and Claudia Giovagnoli (2013). Chemical and physical properties of *Aloe vera* (*Aloe barbadensis* Miller) gel stored after high hydrostatic pressure processing, *Food Science and Technology, Campinas*, 33(1): 52-59.
- Kasim M. U. and Kasim R. (2009). Vapor heat treatment increase quality and prevent chilling injury of cucumbers (*Cucumis Melo* L. Cv. *Silor*). *American-Eurasian Journal & Environmental Sciences*, 11: 269 – 279.
- Krum J.K. (1955). Truest evaluation in sensory panel testing. *Journal of Food Engineering*, 27: 74-78.
- Kumar S. and Bhatnagar T. (2014). Studies to Enhance the Shelf Life of Fruits Using *Aloe Vera* Based Herbal Coatings: A Review. *Journal of Agriculture and Food Science Technology*, 5(3): 211 218.
- Mali S. and Grossmann M.V. (2003). Effects of yam starch on storability and quality of fresh strawberries (*Fragaria ananassa*). *Journal of Agricultural and Food Chemistry*, 21: 7005-7011.

- Mantilla N., Castell-Perez M.E., Gomes C. and Moreira R.G. (2013). Multilayered antimicrobial edible coating and its effect on quality and shelf-life of fresh-cut pineapple (*Ananas comosus*), *LWT - Food Science and Technology*, 51:37-43.
- Mardiana K., Sugiarto E. and Savitri (2008). Pemanfaatan gel lidah buaya sebagai edible coating buah belimbing manis (*Averrhoa carambola* L.), *Fakultas Teknologi Pertanian, Institut Pertanian Bogor*.
- Martinez-Romero D., Guillen F., Castillo S., Valero D. and Serrano M. (2003). Modified atmosphere packaging maintains quality of table grapes. *Journal of Food Science*, 68:1838-1843.
- Martinez-Romero D., Serrano M., Valera D. and Castillo S. (2005). Application de *Aloe vera* comorecubricetnto stobre frutas Yhor taliza, Spain patent-200302937.
- Martinez-Romero D., Alburquerque N., Valverde J.M., Guillen F., Castillo S. and Valero D. (2006). Postharvest sweet cherry quality and safety maintenance by *Aloe vera* treatment: a new edible coating. *Postharvest Biology and Technology*, 39(1): 93–100.
- Masamba K.G. and Mndalira K. (2013). Vitamin C stability in pineapple, guava and baobab juices under different storage condition using different levels of sodium benzoate and metabisulphite. *African Journal of Biotechnology*, 12(2): 186-191.
- Mathooko F.M. (1995). Regulation of ethylene biosynthesis in higher plants by carbon dioxide. *Postharvest Biology and Technology*, 7: 1-26.
- Molan P.C. (1992). The antibacterial activity of honey: 1. The nature of the antibacterial activity. *International Bee Research Association*, 73(1): 5-28.
- Montero-Calderon M., Rojas-Grau M. and Martn-Belloso O. (2010). Mechanical and chemical properties of Gold cultivar pineapple flesh (*Ananas comosus*). *European Food Research and Technology*, 230(4):675-686.
- Ni Y., Turner D., Yates K. M. and Tizard I. (2004). Isolation characterization of structural components of *Aloe vera* L. leaf pulp. *Internatonal Immunopharmacology*, 4: 1745-1755.

- Nisperos M.O. and Baldwin E.A. (1996). Edible coatings for whole and minimally processed fruits and vegetables. *Food Australia*, 48(1): 27-31.
- Oms-Oliu G., Soliva-Fortuny R. and Martin-Belloso O. (2008). Edible coatings with antibrowning agents to maintain sensory quality and antioxidant properties of fresh-cut pears. *Postharvest Biology and Technology*, 50:87–94.
- Park H.J. (1999). Development of advanced edible coatings for fruits. *Trends in Food Science and Technology*, 10: 250-260.
- Paull R. E. and Chen C.C. (2003). Postharvest physiology, handling and storage of pineapple. Wallingford, UK: CAB International, 253-279.
- Pearson D. (1976). Chemical Analysis of Foods. 7th Edn., Church Hill Livingstone, London, UK., 72-73,138-143, 488-496.
- Pesis E., Dvir O., Feygenberg O., Arie R.B., Ackerman M. and Lichter (1999). Production of acetaldehyde and ethanol during maturation and modified atmosphere storage of litchi fruit. *Postharvest Biology and Technology*, 26: 157-165.
- Rocha A. M., and Moraes A. M. (2000). Effects of controlled atmosphere on quality of minimally processed apple (cv. Jonagored). *Journal of Food Processing and Preservation*, 24: 435-451.
- Rojas-Grau M.A., Tapia M.S. and Martin-Belloso O. (2008). Using polysaccharide-based edible coatings to maintain quality of fresh-cut Fuji apples. *Food Science and Technology*, 41: 139-147.
- Sayka M. I., Nahar S., Islam M. M. J., Islam M., Hoque M. M., Huque R., and Khan M.A. (2014). Effect of low molecular weight chitosan coating on physico-chemical properties and shelf life extension of pineapple (*Ananas sativus*). *Journal of Forest Products & Industries*, 3(3): 161-166.
- Simal S., Femenia A., Llull P. and Rossello C. (2000). Dehydration of *Aloe vera*: simulation of drying curves and evaluation of functional properties. *Journal of Food Engineering*, 43: 109–114.

- Simoes A.D.N., Tudela J.A., Allende A., Puschmann R. and Gil M.I. (2009). Edible coatings containing chitosan and moderate modified atmospheres maintain quality and enhance phytochemicals of carrot sticks. *Postharvest Biology and Technology*, 51(3): 364-370.
- Soliva-Fortuny R.C. and Martin-Belloso O. (2003). New advances in extending the shelf life of fresh-cut fruits: a review. *Trends in Food Science & Technology*, 14: 341–353.
- Sperber W.H. and Doyle M.P. (2009). Compendium of the microbiological spoilage of foods and beverages. *Springer*, New York.
- Sritananan S., Uthairatanakij A., Jitareerat P., Photchanachai S. and Vongcheeree S. (2005). Effects of irradiation and chitosan coating on physiological changes of mangosteen fruit stored at room temperature. *Int. Symposium New Frontier of Food and Non-Food Products*, KMUTT, Bangkok, Thailand.
- Valverde J.M., Valero A., Martinez-Romero D., Guileen F. and Castillo S. (2005). Novel edible coating based on *Aloe vera* gel to maintain table grape quality and safety. *Journal of Agricultural and Food Chemistry*, 53(20): 7807-7813.
- Vargas M., Pastor C., Chiralt A., McClements D.J. and González-Martínez C. (2008). Recent Advances in edible coatings for fresh and minimally processed fruits. *Critical Reviews in Food Science and Nutrition*, 48: 496-511.
- Veltman R.H., Kho R.M., Van Schaik A.C.R, Sanders M.G. and Oosterhaven J. (2000). Ascorbic acid and tissue browning in pears (*Pyrus communis* L. cvs Rocha and conference) under controlled atmosphere conditions. *Postharvest Biology and Technology*, 19(2): 129-137.
- Wong W.S., Tillin S.J., Hudson J.S. and Pavlath A.E. (1994). Gas exchange in cut apples with bilayer coatings. *Journal of Agricultural and Food Chemistry*, 42: 2278–2285.