UNIVERSITI PUTRA MALAYSIA

*SHELF LIFE EXTENSION OF SABA BANANA*

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## ABSTRACT

The shelf life of banana is very much dependent on the processing, storage conditions and packaging design. Ordinary food packaging method traps air with the food product. Oxygen and moisture in the air cause the banana to degrade over time, lose flavor and nutrition as well as shorten the shelf life. Vacuum packaging is an effective solution that can be used to remove air in food package thus inhibit the growth of bacteria, mould, and yeast that contribute to food spoilage. Besides that, vacuum packaging can help to secure, seal, thus increasing the shelf life and keeping banana fresh longer. Therefore, this project is directed towards determining the effect of utilizing vacuum packaging and storing in various condition towards extending the shelf life of banana.

Peeled and unpeeled; and vacuum packed (VP) and non-vacuum packed (NVP) banana were kept at different storage conditions which include room temperature (25°C ± 3°C), cold temperature (9°C±2°C) and freezer temperature (-20°C ±2°C) for one month. The shelf life of banana was investigated in term of physical properties such as texture, moisture content and color changes and also in term of antimicrobial properties which was total plate count (TPC) of the colony. Based on the physical analysis, vacuum packaging and -20°C storage temperature was found to significantly delayed the ripening process in terms of fruit firmness, color changes and moisture content. This was due to the fact that the bananas were vacuum packed with good oxygen and moisture barrier properties of low density polyethylene (LDPE) bag hence reduction in oxygen content as well as low storage temperature had retarded the chemical reaction in the bananas. Antimicrobial analysis also revealed that, VP banana exhibited less colony forming unit (CFU/ml) than NVP banana.

# ABSTRAK

Jangka hayat pisang bergantung kepada pemprosesan, keadaan penyimpanan dan reka bentuk pembungkusan. Kaedah pembungkusan makanan biasa memerangkap udara dengan produk makanan. Oksigen dan kelembapan di udara menyebabkan kualiti pisang merosot dari masa ke masa, hilang rasa dan nutrisi pemakanan serta memendekkan jangka hayat. Pembungkusan vakum adalah penyelesaian berkesan yang boleh digunakan untuk mengeluarkan udara dalam pakej makanan yang menghalang pertumbuhan bakteria, mold, dan ragi yang menyumbang kepada kerosakan makanan. Selain itu, pembungkusan vakum boleh membantu untuk, mengelak, dengan itu meningkatkan jangka hayat dan mengekalkan kesegaran pisang. Oleh itu, projek ini diarahkan untuk menentukan kesan penggunaan pembungkusan vakum dan penyimpanan dalam pelbagai keadaan dalam menentu jangka hayat pisang.

Dikupas dan tidak dikupas; dan yang dibungkus vakum dan bukan vakum disimpan pada keadaan penyimpanan yang berbeza seperti suhu bilik (25°C ± 3°C), suhu penyejuk (9°C ± 2°C) dan suhu beku (-20°C ± 2°C) selama satu bulan. Jangka hayat pisang diselidik dari segi sifat fizikal seperti tekstur, kandungan lembapan dan perubahan warna dan juga dari segi sifat antimikrob yang menggunakan kaedah jumlah kiraan plat (TPC) koloni. Berdasarkan analisis fizikal, pembungkusan vakum dan suhu penyimpanan sebanyak -20°C didapati memperlahankan proses pematangan yang ketara dari segi tekstur, perubahan warna dan kandungan lembapan. Ini kerana pisang yang divakum dalam beg polietilena ketumpatan rendah (LDPE) mempunyai ciri-ciri halangan oksigen dan halangan kelembapan yang baik dengan pengurangan kandungan oksigen serta suhu penyimpanan yang rendah telah menghalang tindak balas kimia dalam pisang. Analisis antimikrob juga mendedahkan bahawa, pisang VP menunjukkan kurang membentuk jajahan koloni (CFU / ml) daripada pisang NVP.

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## LIST OF ABBREVIATION

| | |
|---|---|
| PE | Polyethylene |
| LDPE | Less Density Polyethylene |
| CFU | Colony Forming Unit |
| TPC | Total Plate Count |
| NVP | Non-Vacuum Packed |
| VP | Vacuum Packed |
| ΔE | Total Color Difference |
| L* | Lightness |
| a* | Redness |
| b* | Yellowness |
| CAP | Control Atmosphere Packaging |
| MAP | Modified Atmosphere Packaging |

# CHAPTER 1

# INTRODUCTION

## 1.1 Overview

Banana or the general scientific name, *Musa* from Musaceae family, is one of the most important fruit crops of the world and one of the popular tropical fruits in Malaysia. In the trade world ranks, banana is the fifth most important commodity after cereals, sugar, coffee, and cocoa [1]. The largest producers of banana are India, which grew 29 million tonnes per year on average between 2010 and 2015, and China at 11 million metric tons. Output in both countries mostly serves the domestic marketplace. Other large producers are the Philippines with an annual average of 9 million tonnes between 2010 and 2015, and Ecuador and Brazil both at an average of 7 million tonnes [1,2,3,4]. Based on Fruit Crops Statistic in Malaysia, banana ranks second after durian in term of hectarage plantation which is around 28,036.4 hectares after durian 66,037.5 hectares. In 2016, banana production was 309,507.6 metric tonne thus rank second after pineapple production, 391,714.4 metric tonne in terms of annual production [5].

In Malaysia, there is an abundance of Saba Banana in Sabah. Improper management such as handling, packaging, storage, and transportation can lead to wastage and the losses. This was the driving force for utilising a modified atmosphere setup such as vacuum packaging to enhance the shelf-life of perishable commodities and reduce the wastage[3]. Besides that, the storage temperature also one of the crucial factors that will influence the quality of banana thus prolong the shelf life of the banana. As these losses problems can affect the food industry[6], preservation method such as vacuum packaging can be one of the significant solution to this problem. Therefore, in this work, Saba Banana was chosen as sample. Saba banana is well-known as Pisang *Abu Nipah*. The scientific name is *Musa Acuminata Balbisiana (BBB)* where it is a hybrid of the seeded banana *Musa Balbisiana* and *Musa Acuminata*[7].

The color, texture, flavor (taste and aroma), and the nutritional value of fresh fruit and vegetable products are factors critical quality to consumer acceptance and the success of these products [8]. As a producer in the industry, these four attributes typically will affect the marketplace. In the order specified above, consumers will evaluate the visual appearance and color first, because the appearance of the product usually determines whether a product is accepted or declined. Therefore, this is one of the most critical quality attributes. While nutritional value is a hidden character that affects bodies in ways that consumers cannot perceive, but this quality attribute is becoming increasingly valued by consumers, scientists, and the medical profession[8].

In this work, the physical properties which include texture, color and moisture content as well as antimicrobial analysis were done to investigate the quality of vacuum packed (VP) and non-vacuum packed (NVP) banana stored at different storage temperature such as room (temperature 25°C ± 3 °C), cold (temperature 9 °C ± 2 °C) and freezer (temperature -20 °C ±2 °C) for one month .

## 1.2 Problem Statement

Presently, there is an abundance of Saba Banana in Sabah and improper management of the bananas can lead to wastage. Non-availability of adequate postharvest management including handling, packaging, storage, and transportation resulted to the losses, spoilage and placed a great menace to the commercial cultivation of banana. Inappropriate management has caused adverse physiological changes (loss of weight due to increased respiration and transpiration), softening of flesh and lack of resistance capacity against microbial attack.

Therefore, to prevent wastage, it is necessary to improve the postharvest management of the banana in order to increase the shelf life of banana. Banana with longer shelf life may be exported thus solving the abundance problem and contribute to economic growth. The shelf life of banana is very much dependent on the packaging design and storage condition. The oxygen and moisture in the air can cause the banana to degrade over time and shorten the shelf life of banana. Vacuum packaging method is capable to increase the shelf life of banana by reducing the oxygen content inside the package. Besides storing banana at the right condition such as temperature may enhance the shelf life of banana. Reduction in oxygen and temperature may improve the retardation of the respiratory metabolic activities and thereby enhances the longevity of the banana. Thus, this work was directed to investigate the effect of vacuum packaging and various storage temperatures, such as room (temperature 25°C ± 3 °C), cold (temperature 9 °C ± 2 °C) and freezer (temperature -20 °C ±2 °C) for one month towards shelf life of banana.

## 1.3 Research Objective

1. To investigate the effects of vacuum packaging and storage temperature towards the shelf life of Saba banana.
2. To study the antimicrobial properties of vacuum packed and non-vacuum packed Saba banana stored in freezer

## 1.4 Scope of Study

For the first objective, unpeeled and peeled banana was vacuum packed and stored at room (temperature 25°C ± 3 °C), cold (temperature 9 °C±2 °C) and freezer (temperature -20 °C ±2 °C) for one month. Non-vacuum packed banana was used as control. All the samples were kept in storage for one month and the shelf life of the banana was determined in term of physical properties such as moisture content, changes in color and texture/firmness for a designated period of time (day 7, day 14, day 21 and day 28) throughout the period of storage. Then, from the analyzed data of objective one, the best condition of banana was used to proceed with the second objective which involved with antimicrobial test using Total Plate Count (TPC) method. The unpeeled banana (non-vacuum packed) and vacuum packed banana was stored at most suitable temperature (-20 °C ±2 °C) for one month and the shelf life of the banana was determined by determining the number of bacteria colonies formed throughout designated period of time of storage (day 7, day 14, day 21 and day 28).

## 1.5 Significant Contribution

This study determined the reliable and right packaging method and storage condition that can increase the shelf life of banana thus reducing the wastage. Besides, longer shelf life of banana will open the windows towards exporting the banana thus increase the economy growth of the farmers and country.

## 1.6 Thesis Outline

This thesis is divided into five section which describe as below :

Chapter 1: This chapter introduces the overview of banana, problem statement, research objective, scope of study and significant contribution.

Chapter 2: This chapter elaborate the literature review on banana, importance of banana, shelf life and problem associated with banana, preservation method, ripening process of banana, quality of banana including color and texture, effect of temperature on food product, effect of relative humidity/moisture content on food product, effect of packaging on food product and overview of work analysis.

Chapter 3: This chapter explain the methodology involve in preparation of samples which unpeeled and peeled bananas and the shelf life study involve the texture analysis, color analysis, moisture analysis and Total Plate Count (TPC) analysis.

Chapter 4: This chapter discussed the result of unpeeled and peeled bananas based on the physical and antimicrobial analysis.

Chapter 5: This chapter will conclude everything and give recommendation on how to improve the research done before.

# CHAPTER 2

# LITERATURE REVIEW

## 2.1 Banana

There are two basic types of *Musa* genome groups which are, *Musa acuminate* (A genome, 2n = 22) and *Musa Balbisiana* (B genome, 2n = 22). Polyploidy and hybridization of A and B genomes have given rise to diploid (AA, AB, BB), triploid (AAA, AAB, ABB, BBB), and tetraploid (AAAA, AAAB, ABBB, AABB) banana [1].(Several other forms of banana also exist naturally or produced by crossbreeding of these genomes which have different nomenclature. Three common species that are widely grown in the world is (*M.cavandishii*, *M. paradisiaca* and *M. sapientum* [1]. Banana can be split into two groups, eaten fresh/dessert and cooking varieties/plantain. Commercially popular banana is usually triploids (2n = 33). Sweet bananas which are depleted as a dessert fruit are from the AAA genome and the plantains, which are usually cooked before consumption because of their higher starch percentage are from the AAB genome [1]. Cooking varieties or plantains are grown extensively as a staple food source in tropical areas. Plantain varieties account for about 85% of all banana cultivation worldwide [9]. In Malaysia, the most popular or common name of dessert bananas are *Pisang Mas* (AA), *Pisang Berangan* (AA), *Pisang Rastali* (AAB) and *Pisang Embun* (AAA) while the popular cooking types or plantain are *Pisang Raja*

(AAB), *Pisang Nangka* (AAB), *Pisang Tandok* (AAB), *Pisang Awak* (ABB) and *Pisang Abu Nipah* (BBB). Some of these bananas are produced for the export market[10].

Banana is possibly the world's oldest cultivated crop is one of the tallest of the herbaceous plants with a pseudostem [11]. The banana plant springs from an underground stem, or rhizome, to form a false trunk 3–6 meters high. This trunk is composed of the basal portions of leaf sheaths and is crowned with a rosette of 10 to 20 oblong to elliptic leaves that sometimes attain a length of 3–3.5 meters and a breadth of 65cm. A large flower spike, carrying numerous yellowish flowers protected by large purple-red bracts, emerges at the upper side of the false trunk and bends downward to become bunches of 50 to 150 individual fruits, or finger. The individual fruits, or bananas, are grouped in clusters, or hands, of 10 to 20. Desirable commercial bunches of bananas consist of nine hands or more and weigh 22–65 kg and the fruits become ready for harvesting 150 to 180 days after flowering [9].

## 2.2 Importance of Banana.

Banana is famous in view of its simple accessibility, minimal cost, various usage, high caloric and high nutritive content. It contains carbohydrate, crude fiber, protein, fat, ash, phosphorous, iron, $\beta$-carotene, riboflavin, niacin and ascorbic acid [11]. Banana are sometimes used as a treatment for gastric ulcer and diarrhea. Banana is also beneficial for preventing cancer and heart disease because of high vitamin A and B6 contents and also high potassium content that can reduce cardiovascular disease and thus controls blood pressure [12,13,1]. In addition, banana is fundamental component of the human eating routine [6] and there is extensive proof of the wellbeing and healthful advantages related with banana utilization and as suggested by numerous associations, such as, World Health Organization(WHO), Food and Agriculture Organization

(FAO), United States Department of Agriculture (USDA) and European Food Safety Authority (EFSA) to reduce the risk of cardiovascular diseases and cancer [6].

Furthermore, with expanding urbanization, bananas and plantains are becoming increasingly essential as cash crops and giving the sole source of earning to country populaces, hence playing an important role in poverty mitigation. Bananas and plantains can be an exceptionally shoddy sustenance to purchase and are thus a vital nourishment for low-salary families[14]. Other than have high nutritional values and help poverty mitigation, Figure 1 shows different usages of banana such as chips, raw ripened fruit, cooked green banana, fermented and unfermented beverages, juice, puree, dried flour for bakery and infant formula food. Banana is also used as a starch source for various chemicals and packaging materials. The shelf life and functional properties of these products can be altered by the application of various innovative food processing technologies[15].

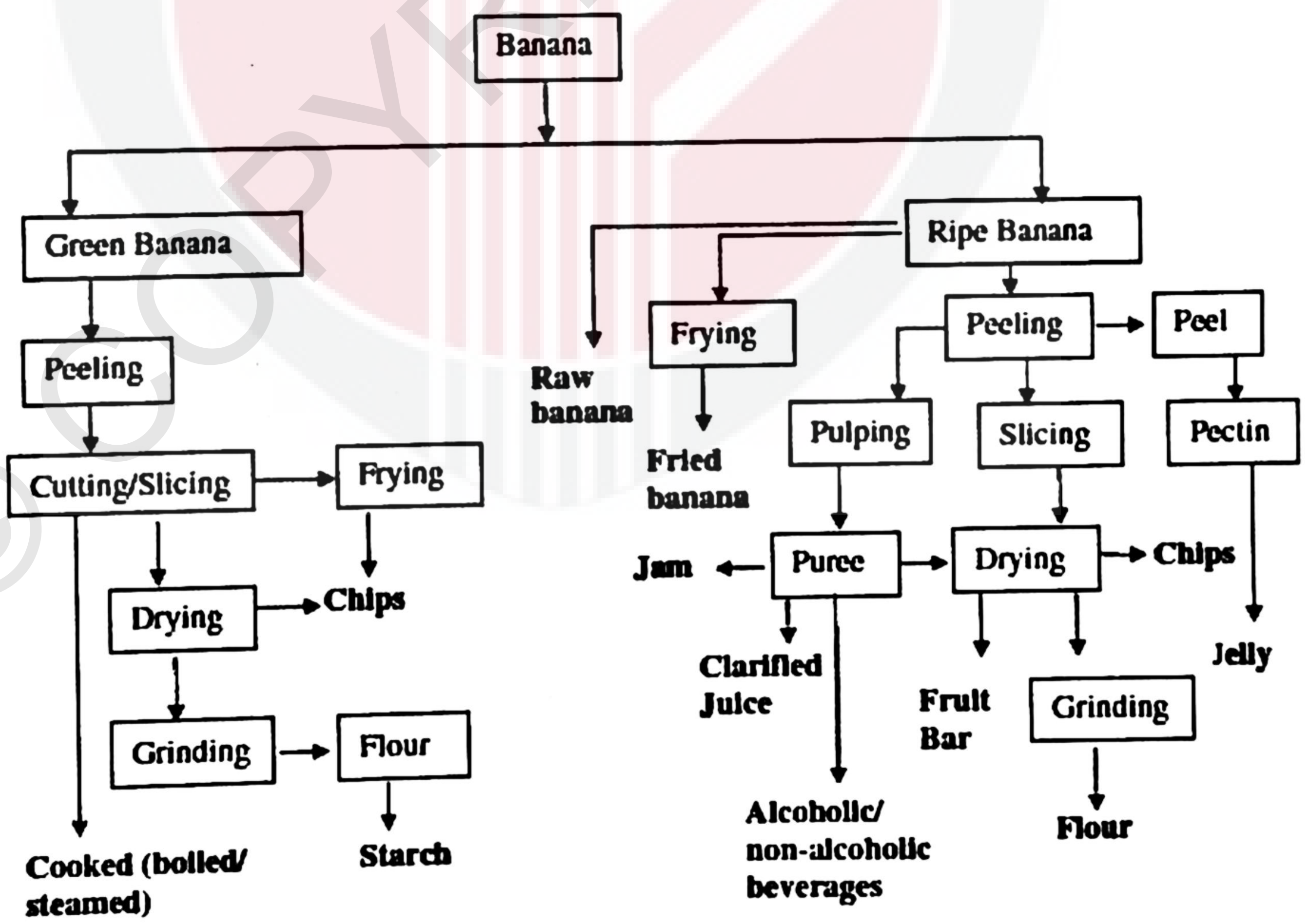



Figure 1: Various Usages of Banana

## 2.3 Shelf Life of Banana and Problems Associated with The Banana that Affect Shelf Life

Shelf-life is the timeframe for the foods to maintain its safety and/or quality under a reasonably predictable state of distribution, storage, and use. The shelf-life of a food starts from the time it is produced and/or packed. Various factors will influence the safety of food and lead to variation in shelf-life [16]. Four major factors that can affect the shelf life of banana are ethylene gas, temperature control, relative humidity control and adequate air circulation. The presence of ethylene which is naturally occurring plant hormone, produced in all plant tissues that is a colorless, odorless, and tasteless gas causes climacteric fruits to ripen and causes many effects in plant physiology[17]. Besides that, unsuitable temperature, humidity and air surrounding also will affect the banana quality such as growing of bacteria, increase the Maillard reaction and cause browning effect. Therefore, it is important to understand the biochemical basis of ripening in order to predict the ripening and ensure good quality [1]. Based on online article stilltasty.com and eatbydate.com, banana stored at room temperature can last up to 2-7 days while in refrigeration temperature banana can last for 2-9 days and if stored at frozen temperature banana can last for 2-3 month.

Banana with the advantages of health, high nutrition, and good flavor while still maintaining freshness, have gained great popularity among customers worldwide. This has contributed to a global trend of increased consumption and research investment on bananas in recent years. Nevertheless, bananas deteriorate faster due to the damages done by poor management (handling, packaging, storage, and transportation). These mismanagements usually shorten the shelf life by a series of typical symptoms, such as tissue softening, cut surface browning, decrease in the nutritional value, the presence of off-flavor and microbiological spoilage during storage. Moreover, the exposed surface also benefits the growth of some pathogenic microorganisms. The dramatically increase in cases recently reported foodborne disease outbreaks associated with this products have caused serious concerns towards public health [18]. Because of these problems, bananas must be put on the need of preservation to maintain and prolong the shelf life.

## 2.4 Fruits and Vegetable Preservation Method

In general, traditional preservation methods of fresh fruits and vegetables could be comprehensively arranged into three classes, namely physical-based preservation, chemical-based preservation and bio-preservation technology[18]. Physical-based preservation technology refers to the methods that modify environmental temperature, humidity, pressure and gas composition for shelf life expansion. Cold storage is a standout amongst the most regularly utilized physical-based techniques to enhance timeframe or shelf-life of fresh fruits and vegetables. It is exceedingly effective but also high in energy demand. However old storage can result to frosty harm/chilling damage because of an alternate temperature of various assortments of new fresh fruits and vegetables when stored under a similar condition.

Regarding chemical-based preservation technology, various of natural or engineered additive have been utilized to prolong the shelf life of fresh fruits and vegetables [19]. In any case, customers have likewise turned out to be more condemning of the utilization of manufactured added substances as their consciousness of well-being and food safety has expanded. This has called for the synthetic additive-free or natural additive-based preservation techniques in recent years. Based on Bico et al., (2009), Cavendish banana dipped with chemical such as 1% (w/v) calcium chloride, 0.75% (w/v) ascorbic acid and 0.75% (w/v) cysteine combined with controlled atmospheres presented the lowest rate of browning and firmness in banana after 5 days of storage at 5°C. While chemical dip combined with carrageenan edible coating plus storage under controlled atmosphere (3% O2 and 10% CO2) could be a good method to preserve bananas for 5 days at 5°C[20]. Besides that, Alum (Potassium Aluminum Sulfate) treatment combined with vacuum packaging-controlled crown rot disease of Cavendish banana completely up to two weeks in cold storage[13].

Concerning the traditional bio-preservation, with a long history of safe utilize, it refers to the rational utilization of the antimicrobial properties of natural microorganisms and their antibacterial products to extend the shelf life and enhance the safety of foods[18].

## 2.5 Ripening Process

Distinctive biochemical and physiological changes occur within a short span of time during ripening, so it is necessary to understand the ripening process of banana in order to extend its shelf life. Figure 2 shows the ripening stages of banana fruits from 1–7 which completely unripe to completely ripen [3]. Commercial standard color charts are available in which 7 stages of peel color were reproduced and translated to a numerical scale where stage 1=all green, 2= green with trace of yellow, 3= more green than yellow, 4= more yellow than green, 5= yellow with trace of green, 6= full yellow and 7= full yellow with brown spots. According to the color chart, there is significant difference in peel color occurs among the advanced stages of maturity, for example, stage 5, 6 and 7 as compared to initial stages [21].

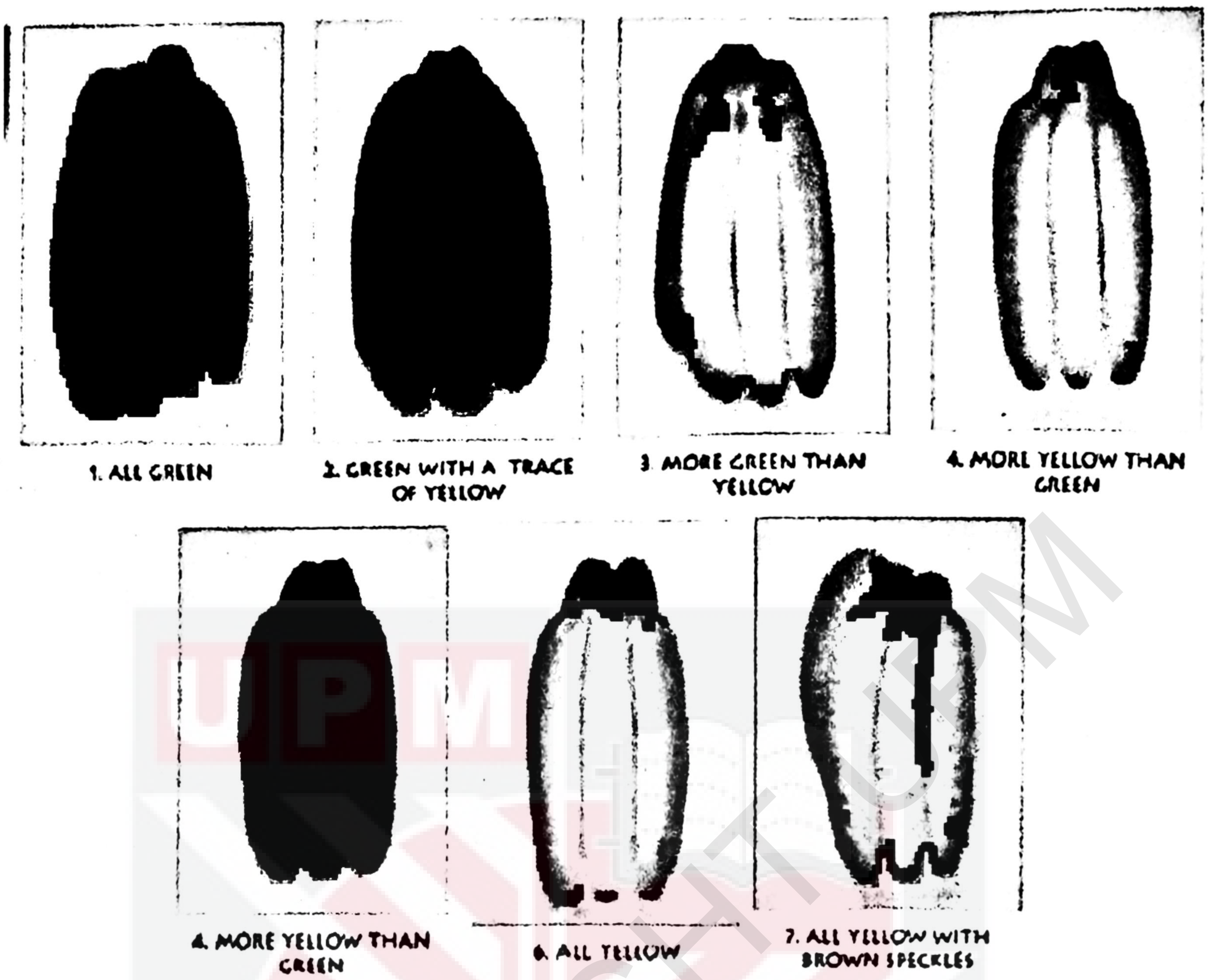



Figure 2: Ripening Stages of Banana Fruit

### 2.5.1 Nature of Respiration

Respiration is the oxidative breakdown of complex substrates like carbohydrates to simpler molecules like $CO_2$ and $H_2O$ [1]. The glycolytic pathway which occurs in the cytoplasm of the cell is the first step of respiration followed by tricarboxylic acid cycle, pentose phosphate pathway and electron transport system [1]. Fruits can be characterized into two general classifications, climacteric and non-climacteric according to their ripening behavior. Climacteric fruits are defined as fruits that enter 'climacteric phase' after harvest for example, they continue to ripen off the tree / plant [22]. Usually, most ripened climacteric fruits are too soft and delicate to withstand rigours of transport and repeated handling. These climacteric fruits are harvested hard and green, but fully mature and ripening is done near the consumption areas. Example of climacteric fruits is mango, guava, banana, apple, papaya and many more. For unripe banana (harvested hard and green) shows a low level of ethylene, a natural plant hormone that regulates every facets of plant growth. During ripening stage, higher ethylene production induces higher metabolic rates of starch to sugar conversion; chlorophyll degradation and unmasking of carotenoids, thus the green banana turns progressively yellow and dark coloured. This also brings about decrease in astringency, decrease in polyphenol content, increase in polyphenol oxidase activity, attributing to browning of peels, increased respiration rate, loss in firmness and increase in moisture content in the pulp[23].

While for non-climacteric fruits, these fruits are once harvested, do not ripen further [22]. For example, orange, grapefruit, strawberry, watermelon and many more.

## 2.6 Quality Defined

A degree of excellence or a high standard is what a quality means. In term of foods quality, it may be defined as the composite of those attributes that differentiate individual units of a product and have criticalness in deciding the degree of acceptability of that product to the consumer [8]. Food quality is always related to color, flavor, and texture of the products.

### 2.6.1 Color In Term of Fruits and Vegetables Quality

Color is derived from the natural pigments in fruits and vegetables which change as the plant maturation and ripening process occurs. The main pigments imparting color quality are the fat-soluble chlorophylls (green) and carotenoids (yellow, orange, and red) and the water-soluble anthocyanins (red, blue), flavonoids (yellow), and betalains (red) [8]. In addition, the enzymes involved in browning reactions such as polyphenol oxidase, which catalyzes the oxidation of polyphenolic compounds, and phenylalanine ammonia lyase, which catalyzes the synthesis of precursors to phenolic substrates can cause enzymatic browning reactions that may result in the formation of brown or black colored pigments. The chlorophylls, carotenoids, anthocyanins, flavonoids, and betalains have its own sensitivity toward heat, acid, alkali, light, oxidation and pH [8].

For banana, changes in color might be due to the the metabolic processes linked to biosynthesis that had caused changes in chlorophyll (green) pigments to carotenoid (yellow) pigment in the fruit which demonstrated that ripening process occurred from time to time in which the rate of the ripening process was dependent on the storage temperature and packaging design[4]. Besides ripening process that had change the color pigment enzymatic browning is also one of the most limiting factors on the shelf-life of fresh-cut products including peeled banana [24]. Cells are broken causing enzymes to be liberated from tissues and put in contact with their substrates. Enzymatic browning is the discoloration which results from the action of a group of enzymes called polyphenol oxidases (PPO), which have been reported to occur in all plants including banana. That why PPO has been considered one of the most damaging enzymes to quality maintenance[24]. Usually, brown pigments are formed, but in addition, reddish-brown, blue-gray and even black discolorations can be produced on some bruised plant tissues.

Therefore, color and appearance are one of the important characteristics that indicate the quality of freshness and flavor for consumer attraction either to purchase the product or not. Consumer still use the external appearance of a whole fruit as an indicator although it can be a misleading indicator. For example, a preferred color for a specific item in consumer's eye is yellow with no brown spots for bananas, tomatoes are supposed to be red and not orange, while cherries are red not yellow, and kiwifruit green-fleshed should not be yellow. Colors that are not appropriate for the item may indicate the loss of freshness or lack of ripeness and can turn away the willingness of consumers to buy the fruits or vegetables [8].

### 2.6.2 Flavor, Aroma, and Taste in Term of Food Quality

Flavor has been specified as a unitary experience which includes sensations of taste, aroma, smell, and cutaneous sensations such as warmth, or mild pain [8]. The flavor also may have the biggest effect on acceptability and desire to consume it once more [8]. Fruits and vegetables can be separated into two major groups upon their flavor attributes [8]. The main group of fruits and vegetables has a strong flavor, for example, bananas with isoamyl-acetate, onions with characteristic of sulfide compounds, and celery, with distinctive phthalides. While the second group of fruits and vegetables has less flavor such as snap beans, muskmelons, and tomatoes. None of these second group conveys the specific characteristic of flavor. Aroma are significantly more diverse and hard to classify, but an attempt includes the following, spicy, flowery, fruity, resinous or balsamic, burnt, and foul. Aroma compounds are unpredictable, and they are perceived primarily by the nose, while taste receptors exist in the mouth and are affected when the food is chewed. Taste has been classified into five primary tastes which are sweet, sour, salty, bitter, and umami. Umami can be portrayed as a taste related to salts of amino acids and nucleotides [8].

### 2.6.3 Texture in Term of Food Quality

The sense of touch, either when the product is picked up by hand or placed in the mouth and chewed is the textural parameters of fruits and vegetables. These qualities can be effectively estimated using instrumental techniques compared to flavor attributes. The texture of fruits and vegetables is derived from their turgor pressure and the composition of individual plant cell walls and the middle lamella "glue" that holds singular cells together and the cell walls are mainly composed of cellulose, hemicellulose, pectic substances, proteins, and lignin[8].

The texture is also a critical quality attribute that helps both the industry and consumer to determine the acceptability of foods. Fruits and vegetables that have firmness, crispness and crunchy texture are profoundly desired by consumers because of their relationship with tissue deterioration [18]. Consumers or panelists are generally more sensitive to small differences in texture than flavor [18]. Loss of firmness is primarily connected with enzymatic degradation of pectins catalyzed by pectin methylesterase (PME) and polygalacturonase (PG)[18]. Besides that, bananas that exhibited lower value in penetration force may be due to ripening process of the banana. The main reasons were due to the breakdown of starch into sugars, the breakdown of cell walls or reduction in cohesion of the middle lamella due to solubilization of pectic substances and also because of migration of water from skin to flesh due to osmosis process[25].

Previous research reported by Kharthiani et al., (2013) whom found that the firmness of Robusta and Poovan banana to decrease as the storage period increased. While reasearch done by Soltani et al., (2011) also showed the same result where firmness and hardness of *Cavendish* banana decreased from 7647.87g to 2753.23g as fruit ripened with the same reason as before.

## 2.7 Effect of Temperature on Food Product

The safety and shelf-life of most foods are influenced by the temperature. Therefore, during all stages of food manufacture, store, distribution and use, the supervision of temperature should be carefully dealt with, measured and documented. Besides that, temperature abuse such as during storage, distribution, and use should also be prevented for maintaining the quality and reduce the losses[16]. This is because all types bacteria have an optimum temperature in which they can growth from time to time. The relationship between temperature and growth rate constant varies significantly across groups of microorganisms. Four major groups of microorganisms based on their temperature ranges for growth are thermophiles, mesophiles, psychrophiles, and psychrotrophs. The optimum temperature for bacteria growth for each group are shown in Table 1. Based on previous research made the lowest recorded temperature of growth for a microorganism of concern in foods is −34°C[26]. Referring to the temperature range as shown in Table 1, the bacteria in bacteria in this research can be classified into mesophile, psychrotrophs and psychrophile.

Table 1: Temperature range for microorganism growth

| Groups | Temperature range °C (°F) | | |
|---|---|---|---|
| | Minimum | Optimum | Maximum |
| **Thermophiles** | 40 - 45 (104 - 113) | 55 - 75 (131 - 167) | 60 - 90 (140 - 194) |
| **Mesophiles** | 5 - 15 (41 - 59) | 30 - 45 (86 - 113) | 35 - 47 (95 - 117) |
| **Psychrotrophs** | -5 - +5 (23 - 41) | 25 - 30 (77 - 86) | 30 - 35 (86 - 95) |
| **Psychrophiles** | <-5 (<23) | 12 - 15 (54 - 59) | 15 - 20 (59 - 68) |

Source : Factor that influence microbial growth[27].

## 2.8 Effect of Relative Humidity to Food Product

Relative humidity is the concentration of moisture in the air surrounding a food[16]. Typically, there is a trade of moisture between a food and its atmosphere which proceed until the moisture in food is equilibrium with the surrounding air [16]. As such, the relative humidity can influence the water activity of foods and this factor must be taken into consideration during food handling. The relative humidity is related to the storage and distribution temperature of foods. Generally, a higher relative humidity is required for a lower storage temperature, to ensure that the food product characteristics are maintained [16].

Lower RH is the driving force for water loss from the fruit. Low RH reduces the green life and can produce peel symptoms that are similar to the damage of chilling injury.[14] Besides that, the storage conditions that have high relative humidity might also favor the moisture migration from the environment to the banana. This led to increase in moisture content and also known to lead to increase of reactant mobility[28].

## 2.9 Effect of Packaging to Food Product

A good packaging system will protect or reduce the damage to the package contents. Packaging will also help to control both the gas atmosphere and relative humidity of foods [16]. However, packaging material will exhibit different properties such as gas and water vapor permeability, which will affect food safety and shelf-life. For foods packaged in impermeable packaging materials, the relative humidity of the storage environment is unlikely to influence the shelf-life of food. However, if the shelf-life of the food is limited by moisture gain/loss or if the food is packaged in moisture sensitive packaging, control of relative humidity should be a consideration in setting and validating shelf-life of food[16]. There are many types of packaging materials used for food packaging in the food industry and one of it is modified atmosphere packaging such as vacuum packaging. The packaging design also related to the materials used and the barrier properties of food packaging in determining the quality of food.

### 2.9.1 Control Atmosphere Packaging (CAP)

The gas atmosphere and its composition surrounding a food will affect the shelf-life of the food. In order to extend the shelf life of food, the gas atmosphere and its composition can be toilered accordingly by utilizing a modified atmosphere packaging (MAP) instrument, such as gas flushing or vacuum packaging (VP) [18]. The extension of shelf-life using MAP or VP requires the control of temperature and other attributes of the food such as pH. Moreover, the concentrations of gases, the packaging, and equipment used would also be able to affect the food safety. For example, the permeability of packaging and actively respiring fruit and vegetables can affect the composition of gases in the pack during ripening process which in turn, can affect microbial growth and product safety [16].

Vacuum packaging (VP) is able to increase the shelf life of food products by removing air from the product pouch and sealing it. The absence of air in the food package create a vacuum condition of food surrounding, thus inhibit the growth of microorganisms and food oxidation. The growth of aerobic microorganisms is supported by oxygen, so removal of oxygen from the modified atmosphere has been shown as a good attempt to extend the microbiological shelf life[29]. A vacuum pressure will affect the percentage of oxygen remains in food package[29]. For example, a vacuum of 91.75 kPa resulted in 2.09% residual oxygen while 97.929 kPa vacuum leaves 0.6% residual oxygen [29]. In order to get a residual oxygen content of lower than 1%, a vacuum pressure of better than 95 kPa is needed. Under good vacuum condition, the oxygen level is brought down to less than 1% and depending to the barrier properties of the film used, entry of oxygen from outside will be limited. Therefore, vacuum packaging is one of the recent technology that can be used to delay the ripening process, decrease disease incidence and retard biochemical decay in order to extend the shelf-life of fruits [3]. Research done by Garcia and Barret (2002) shown that, using a moderate vacuum packaging with polyethylene (80μm) for the storage of shredded Iceberg lettuce at 5°C, browning was inhibited over a 10 day period[24].

Although the effectiveness of vacuum packaging in prolonging shelf life has been proven, vacuum packaging has its own advantages and disadvantages, and some is tabulated in Table 2.

Table 2:Advantages and disadvantages of vacuum packaging

| Advantages | Disadvantages |
|---|---|
| • A simple solution to packaging goods requiring protection from oxygen.<br>• Positive control of the moisture content of the product.<br>• Inhibits the growth of aerobic spoilage bacteria.<br>• Lower costs than those of rigid containers.<br>• Longer shelf life for goods | • It is virtually impossible to remove all the oxygen since small quantities will surely be trapped within food cells.<br>• Microorganisms which are not affected by oxygen are not advantageously affected by vacuum packaging. |

Source : Vacuum Packaging Technology : a Novel Approach for Extending the Storability and Quality of Agricultural Produce

### 2.9.2 Gas and Vapor Permeability

Optimization of packaging permeability is very crucial to extend the shelf-life of food and to gain the good quality of food. Hence, to find the most suitable material for the food product, it is important to characterize the permeability of different packaging material to the types of packaging gases and its effect in different environmental condition [30]. Barrier properties of packaging materials are necessarily associated with their inherent ability to permit the exchange of gas and water vapor to a higher or lower extent through mass transport processes like permeation.

Oxygen Transmission Rate (OTR) is the oxygen barrier property of a food packaging materials for food product and this property plays important role in the preservation process. The oxygen barrier can be quantified by the oxygen permeability coefficients (OPC) which shows the quantity of oxygen that permeates per unit of area and time in a packaging material (kg·m/(m2·s·Pa)). A polymer film packaging that has low oxygen permeability coefficients indicates that the oxygen pressure inside the package drops to the point where the oxidation is retarded thus, extending the shelf-life of the product [30].

### 2.9.3 Low Density Polyethylene (LDPE)

There are many types of packaging materials that can be used as food packaging. For instance, polyethylene (PE), polypropylene (PP), polystyrene (PS), polyvinyl alcohol (PVOH) /ethylene vinyl alcohol (EVOH), polyamides (PA or nylon) and many more. All packaging material exhibit different properties and benefit, but in this projects, LDPE is used as packaging material, this is because LDPE polymerized from ethylene, is the most commonly material applied for food packaging[31]. Besides that, LDPE exhibit flexibility, better moisture control, oil and chemical resistance, and good impact strength. LDPE is also an inexpensive material, so for applications where its performance is suitable, this material is usually the best option. LDPE has low amorphous content and thus higher crystallinity. Therefore, LDPE has high barrier properties because permeation occurs almost exclusively through amorphous areas of the material. LDPE is therefore, a good water vapor, oxygen and carbon dioxide barrier[31].

## 2.10 Overview of Work Analysis in This Research

### 2.10.1 Texture Analysis

Texture evaluation can be separated into three classes which are fundamental, empirical, and imitative tests. Fundamental tests measure properties that are familiar to engineers like strength, Poisson's ratio, and various moduli such as Young's modulus, Shear modulus, and Bulk modulus. While, empirical tests cover a wide range of simple and rapid tests, including puncture, compression, extrusion, shear, and others, which measure one or more textural properties and are commonly employed in quality control applications [8]. Most methods used for the evaluation of the textural properties of fruits and vegetables are empirical and commonly practiced methods for the evaluation of textural properties are via puncture or compression. Therefore, in this project, empirical method is used to determine the firmness of banana. It consists of quantifying the force and/or deformation required to force a probe or punch into a food to a depth that causes irreversible harm or failure. The use of a texture analyzer has illustrated consistent firmness loss in stored fresh fruits with both a puncture test and a compression test [8].

Bananas that exhibited lower value in penetration force may be due to ripening process of the banana. The main reasons were due to the breakdown of starch into sugars, the breakdown of cell walls or reduction in cohesion of the middle lamella due to solubilization of pectic substances and also because of migration of water from skin to flesh due to osmosis process[25]. Previous research reported by Kharthiani et al., (2013) whom found that the firmness of Robusta and Poovan banana to decrease as the storage period increased. While reasearch done by Soltani et al., (2011) also showed the same result where firmness and hardness of *Cavendish* banana decreased from 7647.87g to 2753.23g as fruit ripened with the same reason as before.

### 2.10.2 Color Analysis

The color of fruit can be described by several colors coordinate systems. Some of the most popular systems are CIELAB and RGB. The CIELAB color space is shown in Figure 3 may be divided into a three-dimensional (L, a and b) rectangular area such that L (lightness) axis goes vertically from 0 (perfect black) to 100 (perfect white) in reflectance or perfect clear in transmission. The a-axis (red to green) considers the positive values as red and negative values as green; 0 is neutral. The b axis (blue to yellow) expresses positive values as yellow and negative values as blue; 0 is neutral (Soltani, Alimardani, & Omid, 2011; Barrett et al., 2010). Fruits and vegetables are often described in terms of their L, a, and b values. In this project the color of banana sample was measured by a spectrophotometer and ΔE* (Total Color Difference) is calculated based on delta L*, a*,b* color differences and represents the distance of a line between the sample and standard [33]. The ΔE can also be an indicator to the changes in banana colors as the lower the total color difference or ΔE value, the closer the sample is to the standard[28,33]

In term of banana, higher changes in ΔE* and b* this increment could be due to the metabolic processes linked to biosynthesis that had caused changes in chlorophyll pigments to carotenoid pigment in the fruit which demonstrated that ripening process occurred from time to time in which the rate of the ripening process was dependent on the storage temperature and packaging design[4]. Research done by Kudachikar et al., (2011) also reported that Robusta and Poovan banana exhibited increment in a* value of the peels of packed green banana across storage period. Besides that, similar result by Soltani et al., (2013) also reported that, there were color changes in the Cavendish banana with the same reason[32].

For L* that indicate to lightness from perfect white to perfect black can be used to measure the ligness of peeled banana or fresh cut banana. If the L* value was decreased it shown that the white color of bananas at the beginning turned to blackish slowly. These changes might be because of enzymatic browning reaction during storage period. Enzymatic browning is one of the most limiting factors on the shelf-life of fresh-cut products including peeled banana [24]. Cells are broken causing enzymes to be liberated from tissues and put in contact with their substrates. Enzymatic browning is the discoloration which results from the action of a group of enzymes called polyphenol oxidases (PPO), which have been reported to occur in all plants including banana. That why PPO has been considered one of the most damaging enzymes to quality maintenance[24]. Usually, brown pigments are formed, but in addition, reddish-brown, blue-gray and even black discolorations can be produced on some bruised plant tissues.

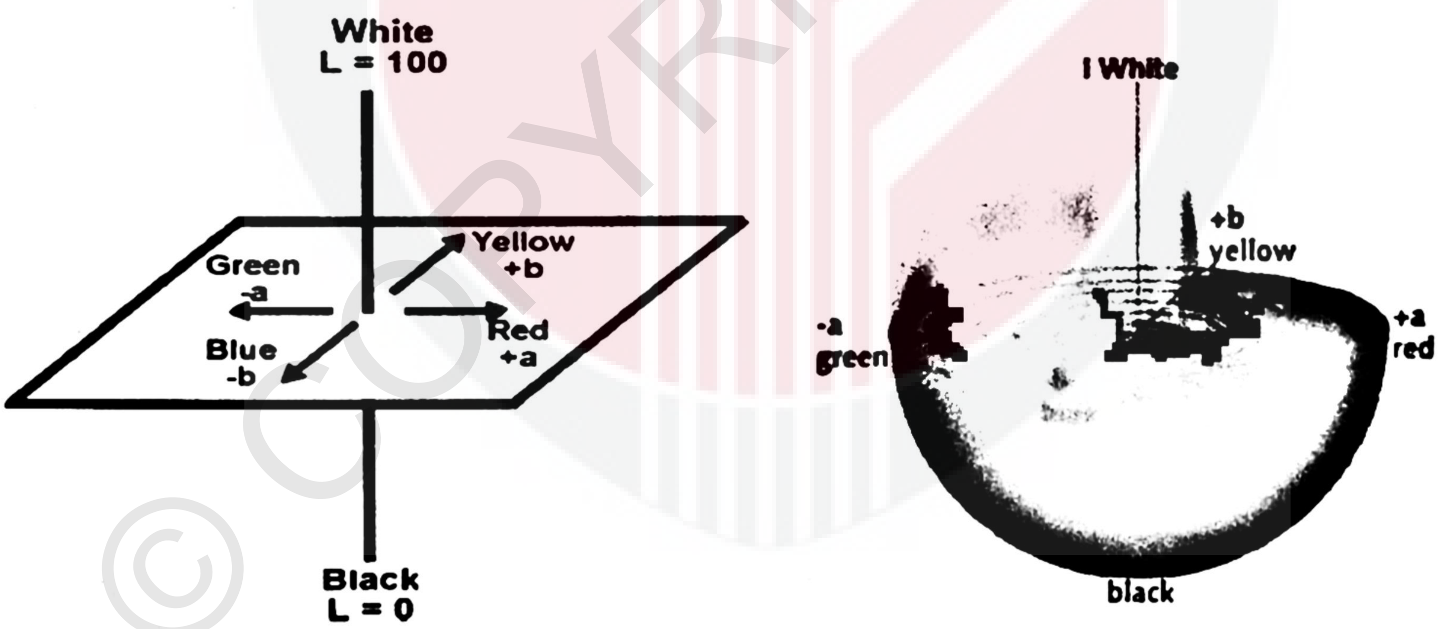



Figure 3: Diagram depicting three-dimensional L, a and b color space.

### 2.10.3 Moisture Test

Moisture content analysis is crucial in food samples, because moisture content influences the taste, texture, weight, appearance, and shelf life of foodstuffs. Even a slight deviation from a defined standard can adversely impact the physical properties of a food material. Also, the rate of microbial growth increases with total water content, possibly resulting in spoiled foods[16].

The increase in moisture content of banana was basically due to the ripening process that occurred throughout the storage period and this occurrence was because of osmotic transfer of moisture from peel to pulp during the storage[34,4]. Besides that, the storage conditions that have high relative humidity might also favor the moisture migration from the environment to the banana. This led to increase in moisture content and also known to lead to increase of reactant mobility[28]. In addition, the increase in moisture content was also due to the breakdown of starch into sugars hence high moisture content[25]. Similar result was obtained in the research done by Kudachikar et al. (2011), where they found that there was an increase value in moisture content for Robusta and Poovan banana as the storage period increased. Research conducted by Bassey at al., (2013) also gave the similar result pattern when they analyzed on other type of cooking banana.

### 2.10.4 Total Plate Count (TPC)

The microbial population of fresh fruits and vegetables is determined to a large extent by the origin of fruits and vegetables, agricultural practices, conditions of harvesting, processing and storage. During distribution and storage, high humidity in packaged products and cut surfaces provide favourable conditions for the growth of spoilage microorganisms [35]. Therefore, microbial quality assessment should be taken into consideration in evaluating the impact of handling, processing and storage condition on maintaining the quality of fruits and vegetables, ensuring the safety and determining their shelf-life. According to French regulations, an aerobic plate count (APC) also known as total plate count (TPC) of $5x10^7$ colony forming units (CFU)/ml is the maximum acceptable value at the end of the microbiological shelf-life of numerous for fruits and vegetables [35].

Aerobic plate count (APC), also known as Total Plate Count (TPC) is used as an indicator of the number of bacteria in a food product. APC only measures those microorganisms capable of growing at 30-37°C in the presence of oxygen. Aerobic plate counts are typically incubated at 36±1°C for 24±3 hours, but other temperatures (e.g. 25°C) may be used [36]. The APC quantifies bacterial densities using growth media that contains no selective or differential additives. The media most commonly used for this purpose is plate count agar (PCA). APC can be performed using pour plate, spread plate or spiral plating techniques and each dilution should be plated in duplicate or triplicate. Besides that, Peptone water (0.1% w/v peptone) is often used as the dilutant in order to maintain the osmotic stability of diluted cells [35]. For microbial quality analyses, a ratio of 1:9 sample:peptone is used for example, a 10g sample of fruit or vegetable is usually weighed into a sterile blender cup or a stomacher bag. Based on the sample, either sterile bags or sterile filter bags are used. Then, peptone water

(90mL) of 0.1% is added and the sample is either blended or pummelled in laboratory stomacher. The bacteria counts are usually expressed as log10 CFU/g of the fruit or vegetable and the logarithmic growth curve is plotted [35].

Bacterial growth curve is a curve on a graph that shows the changes in size of a bacterial population over time in a culture. Based on Figure 4, there are various phase for bacterial growth. When a microorganism is introduced into the fresh medium, it takes some time to adjust with the new environment. This phase is termed as Lag phase, in which cellular metabolism is accelerated, cells are increasing in size, but the bacteria are not able to replicate and therefore no increase in cell mass [37]. The length of the lag phase depends directly on the previous growth condition of the organism. When the organism can easily adapt to the environment, it can start the cell division without any delay, and therefore will have less lag phase and Exponential or Logarithmic (log) phase may starts. During this phase, the microorganisms are in a rapidly growing and dividing state. Their metabolic activity increases and the organism begin the DNA replication by binary fission at a constant rate. The growth medium is exploited at the maximal rate, the culture reaches the maximum growth rate and the number of bacteria increases logarithmically (exponentially) and finally the single cell divide into two, which replicate into four, eight, sixteen, thirty two and so, this will result in a balanced growth. The next phase is Stationary phase. As the bacterial population continues to grow, all the nutrients in the growth medium are used up by the microorganism for their rapid multiplication. This result in the accumulation of waste materials, toxic metabolites and inhibitory compounds such as antibiotics in the medium. This shifts the conditions of the medium such as pH and temperature, thereby creating an unfavourable environment for the bacterial growth. The reproduction rate will slow down, the cells undergoing division is equal to the number of cell death, and finally bacterium stops its division completely. The cell number is not increased and thus the growth rate is stabilised. Then, Decline or Death phase will take place where the depletion of nutrients and the subsequent accumulation of metabolic waste products and other toxic materials in the media will facilitates the bacterium to move on to the Death phase. During this, the bacterium completely loses its ability to reproduce. Individual bacteria begin to die due to the unfavourable conditions and the death is rapid and at uniform rate. The number of dead cells exceeds the number of live cells.

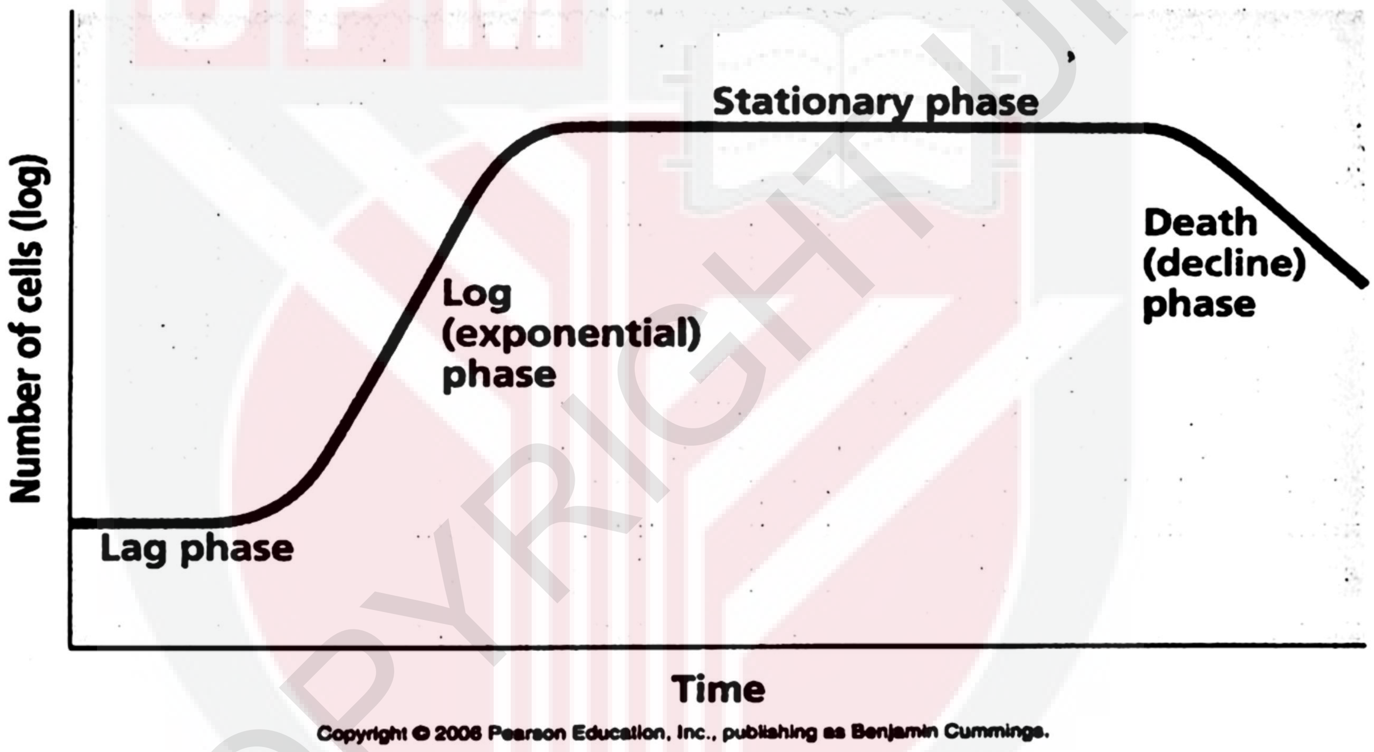



Figure 4: Standard Bacterial Growth Curve

# CHAPTER 3

# METHODOLOGY

In this chapter, preparation and the handling of samples is discussed. This include the analysis done to investigate the shelf life of Saba banana such as texture analysis, color analysis, percentage moisture content analysis and antimicrobial analysis. All method/procedure and equipment used are slso elaborated in this chapter.

## 3.1 Preparation of Samples

A firm fruit is desired to improve handling, transporting and shipping process. This is because the firm fruit can withstand mechanical damage during postharvest handling and therefore may show excellent visual quality and acceptance to retailers and consumers [8]. In this project, mature green bananas or stage one Saba banana that have uniform size, shape and color were selected according to the ripeness index. The bananas were harvested from the same plantation located at Hutan Lipur Lentang, Bentong, Pahang to avoid variation in experimental fruits [11].

### 3.1.1 Banana Preparation for Physical Analysis

The samples were grouped into 2 categories which were unpeeled samples (vacuum packed (VP), non-vacuum packed (NVP)) and peeled samples (vacuum packed (VP), non-vacuum packed (NVP)). Firstly, bunches of banana were `de-handed' and 3 fingers with total weight of approximately 200g-250g were selected to be packed. All fingers were washed under running water to remove dirt. Then the samples were allowed to dry naturally before being vacuum packed. The samples were placed in Low-Density Polyethylene (LDPE) bags with a dimension of 20cm x 25cm. After that, vacuum was applied for about 30s using vacuum sealer machine (GWP, Malaysia) as shown in Figure 5 to evacuate the air inside the LDPE bag and the machine sealed the bag automatically. The samples were stored in three different storage conditions which were room (temperature 25°C ± 3 °C), cold (temperature 9 °C±2 °C) and freezer (temperature -20 °C ±2 °C) for one month. Analysis on the banana was done every week (day 7, day 14, day 21 and day 28). The NVP banana (control sample) was also stored at the same 3 conditions and analyze accordingly. For peeled banana, the peel was removed after washing process and the preparation proceeded as unpeeled banana preparation. Summary of banana samples preparation is shown in Figure 6.

Figure 5: vacuum sealer machine

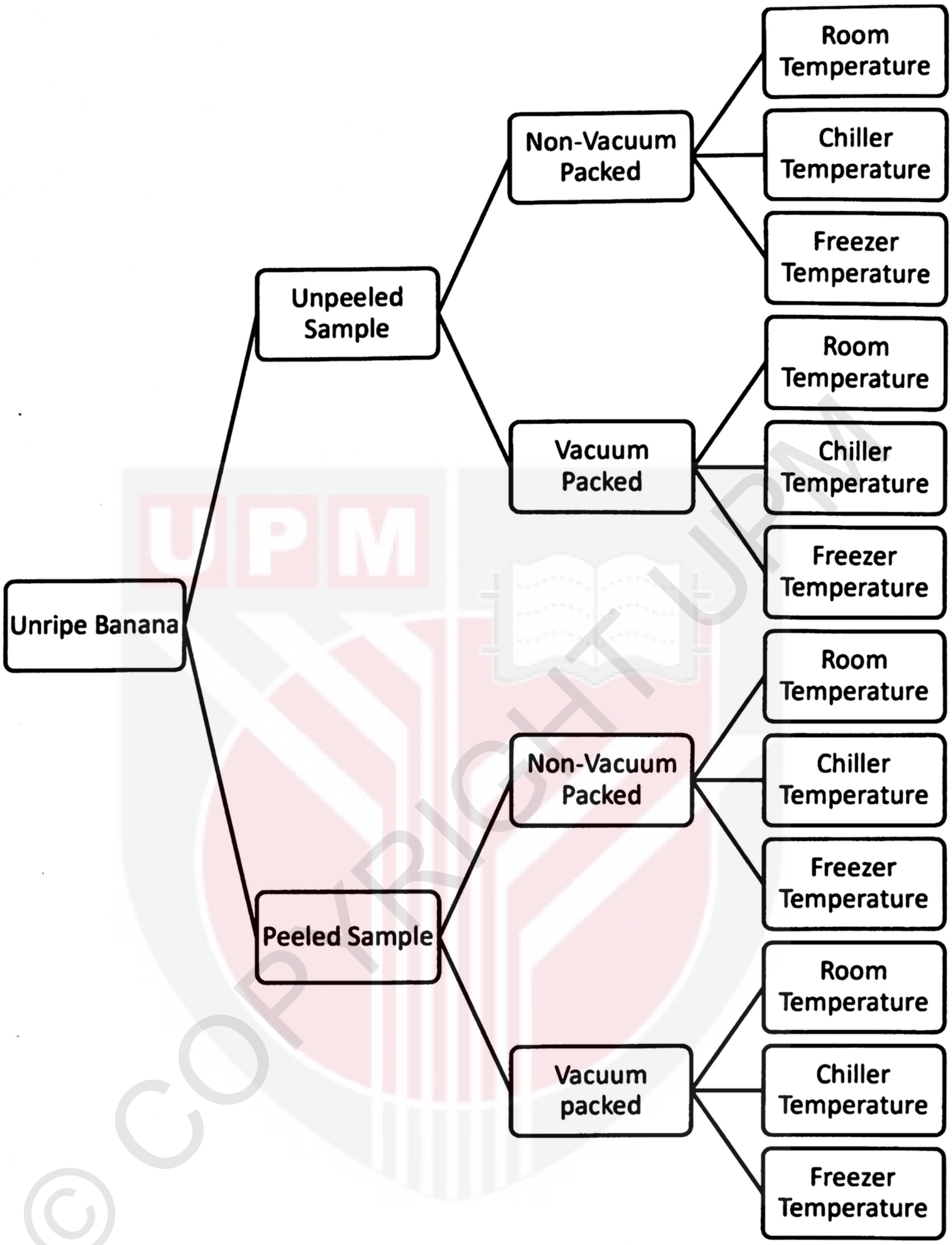



Figure 6: Summary of banana preparation for physical analysis

### 3.1.2 Banana Sample Preparation for Antimicrobial Analysis.

The microbial population of bacteria was investigated using TPC method. Based on the physical analysis made, the chosen banana condition to be used in this analysis was unpeeled banana. The preparation of banana samples are same as the physical analysis and then the unpeeled banana was packed in vacuum packaging and the control banana sample was without packaging. The banana samples were then stored in freezer (temperature-20 °C ±2 oC)(Elba, Fiamma Sdn Bhd, Malaysia) as shown in Figure 7 for one month of storage period and the analysis was made every week (week (day 7, day 14, day 21 and day 28). The summary of banana sample preparation is showed in Figure 8.

Figure 7: Freezer used to keep the banana samples

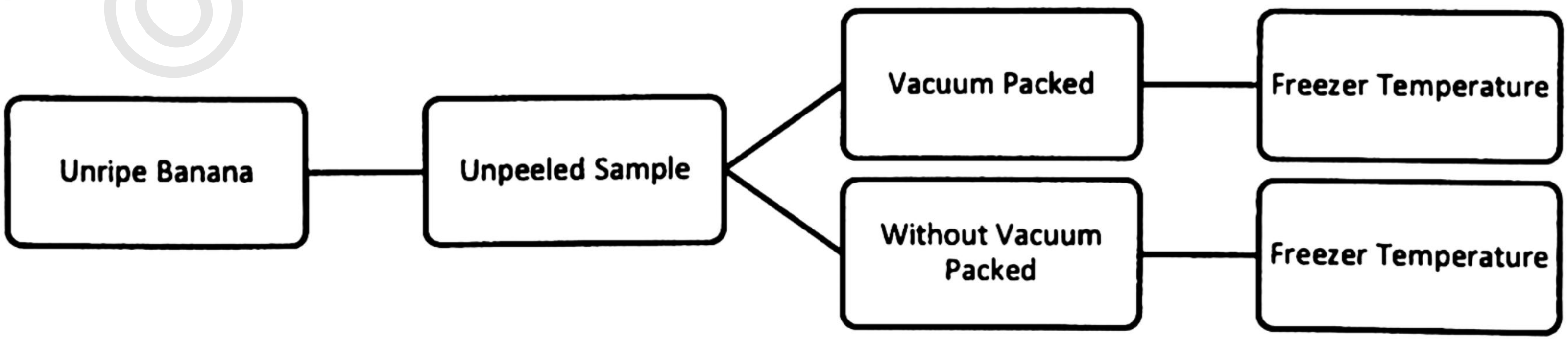



Figure 8: Summary of banana sample preparation for microbial analysis

## 3.2 Shelf Life Study

The shelf life of banana can be influenced by different kind of processing, storage conditions and packaging design. Therefore, the shelf life study was determined using different types of analysis such as physical analysis which include texture/firmness, changes of color, and percentage of moisture content. Besides that, antimicrobial analysis was also conducted using total plate count (TPC) analysis. The analysis was done every week during a month of storage.

**The procedure of testing :**

### 3.2.1 Texture Analysis

Firmness of banana was measured using a texture analyzer (TA XT Plus Texture Analyser, Stable Micro Systems, London) as shown in Figure 9, equipped with a 5kg load cell. The analyzer was linked to a computer that recorded data via a software programme called Texture Expert. Firmness evaluation was carried out by using whole peeled and unpeeled banana and penetrating it with a 2mm diameter cylindrical rod at a speed of 1.0mmsec$^{-1}$ and automatic return. The downward distance was set at 5mm and pre-test and post-test speeds were both set at 2mmsec$^{-1}$. Samples were positioned so that the rod penetrated their geometric center at the middle of the banana in longitudinal axis[3,38,25]. The test options and test settings to operate the machine are represented in Table 3.

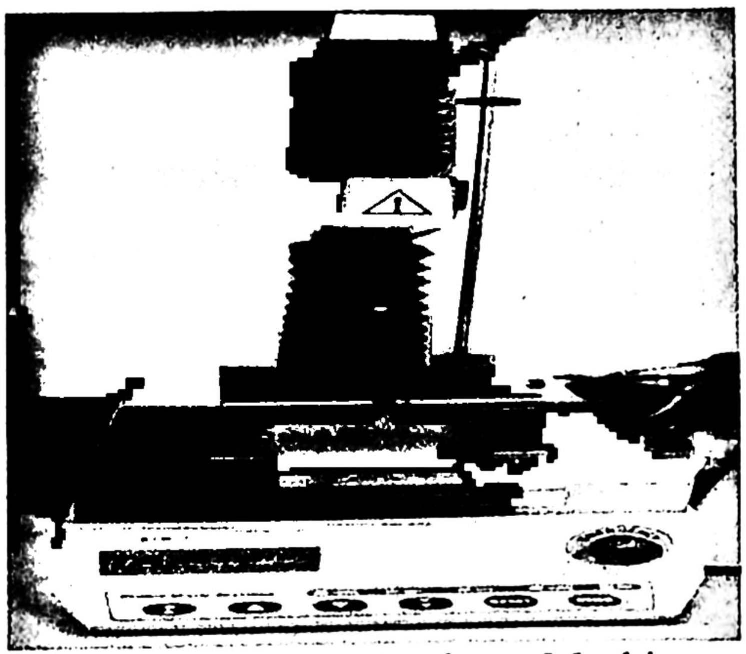


Figure 9: Texture Analyzer Machine

Table 3: Parameter to operate the texture analyzer

| Sl.No | Test options | Command |
|---|---|---|
| 1 | Type of probe used | Penetration rig |
| 2 | Test module | Measure force in compression |
| 3 | Test option | Return to start |

TEST SETTINGS FOR TEXTURE ANALYSER

| Test settings | Set values |
|---|---|
| Pre test speed | 2.0 mm/s |
| Test speed | 1.0 mm/s |
| Post test speed | 2.0 mm/s |
| Distance | 5 mm |
| Trigger force | 5 g |
| Load cell | 5 kg |

### 3.2.2 Color Analysis

In this project, the tri-stimulus color changes were recorded with color spectrophotometer/ spectrocolorimeter (UltraScan Pro, Hunter Associates Laboratory, Inc., Amerika) as shown in Figure 10. The instrument was calibrated using a standard ceramic white tile. The instrument was equipped with an inbuilt software (chromaflash) that denote L as well as a and b values. The color of the banana was measured as $L*$, $a*$ and $b*$ using *CIELAB* system (Chauhan et al., 2006);[4] and Delta E* (Total Color Difference) was calculated based on delta L*, a*,b* color differences and represents the distance of a line between the sample and standard using this formula:

$$\Delta E = \sqrt{(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2} \quad (1)$$

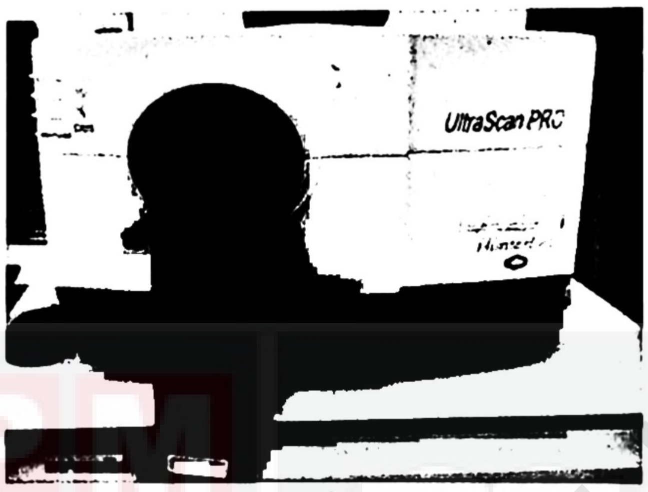


Figure 10: Spectrophotometer Machine

### 3.2.3 Moisture Content Analysis

Moisture analysis was conducted by first measuring the weight of an empty container (A) using sensitive weighing balance (Uni Bloc, Shimadzu Corporation, Philippine) as shown in Figure 11. Then, 30 g of chopped banana flesh was placed on the container and was measured (B). The chopped banana flesh was then placed in an oven (OF22, JEIO Tech, Korea) as shown in Figure 12, at 105 °C overnight (24 h). Lastly, the sample was weighed again after drying (C) and then the percentage of dry matter content of the flesh and percentage of moisture content was calculated as follows [14]:

$$\text{Weight of wet sample (D)} = B - A \quad (2)$$

$$\text{Weight of dry sample (E)} = C - A \quad (3)$$

$$\text{Moisture content (\%)} = \frac{D - E}{D} \times 100 \quad (4)$$

$$\text{Dry matter content (\%)} = 100 - (\text{\% moisture content}) \quad (5)$$

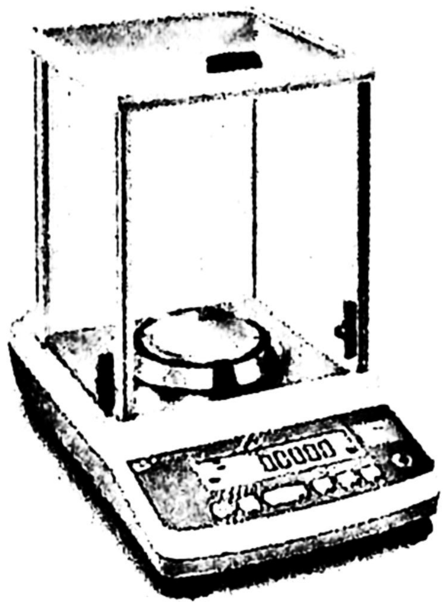


Figure 11: Sensitive Weighing Balance

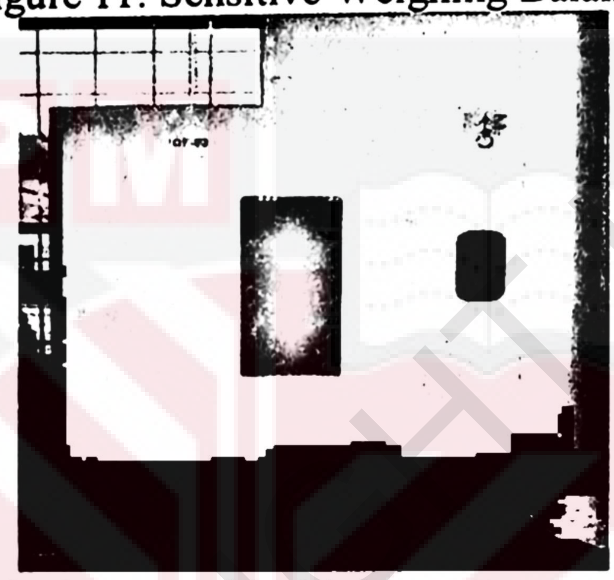


Figure 12: Oven used in Moisture Content Analysis

### 3.2.4 Total Plate Count Analysis (TPC)

#### 3.2.4.1 Media and Agar Preparation

Peptone water (0.1% w/v, OXOID Ltd., UK) was used as saline water and as a growth medium or as the basis of carbohydrate for bacterial growth. While, Plate Count Agar (PCA) (OXOID Ltd., England) also called Standard Methods Agar (SMA), is a microbiological growth medium commonly used to assess or to monitor total or viable bacterial growth of a sample. Both media was prepared before spread process took place. Then, all media, agar and equipment such as tips, hockey stick, universal bottle and beaker was sterilized for 15 minutes with temperature 121°C using autoclave (Tomy, Japan) as shown in Figure 13. The sterilization process was done to ensure that no contamination occurred.

Figure 13: Autoclave Machine

#### 3.2.4.2 Spread Plate and Bacteria Count Procedure

The samples of 10g of bananas were weighed aseptically and mixed with 90ml of peptone water (0.1%w/v) and homogenized in a blender (Panasonic, Japan) as shown in Figure 14. Then serial dilutions of bananas extract mixed with peptone water were prepared in universal bottle using Eppendorf Pipettes (Research plus, Germany) and tips (Research plus, Germany) as shown in Figure 15 and Figure 16. Then, the extract of banana was spread into the plate (Figure 17) containing plate count agar using hockey stick. All preparation was done under laminar flow (ESCO, Esco Micro (M), Singapore) cabinet to avoid contamination as shown in Figure 18 and ethanol (70%, Hmbg Chemicals, Germany) was used as sanitizer. After that, the plate was stored in incubator (Lab Companion, Chemopharm, Malaysia) at incubation temperature (37 °C±1 °C) for 18 hours as shown in Figure 19. The test was performed in triplicate and the number of colony formed was counted and calculated using the following equation :

$$Total\frac{CFU}{ml} = \frac{No.colony\ x\ Dilution\ factor}{volume\ plated} \quad (6)$$

Figure 14: Blender Used in Sample Preparation

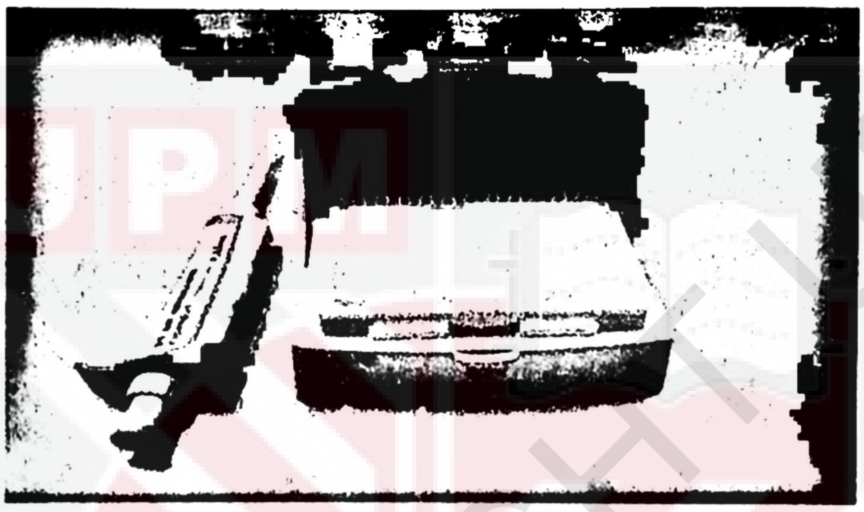


Figure 15: Eppendorf Pipette and Tips Used to Extract Bananas

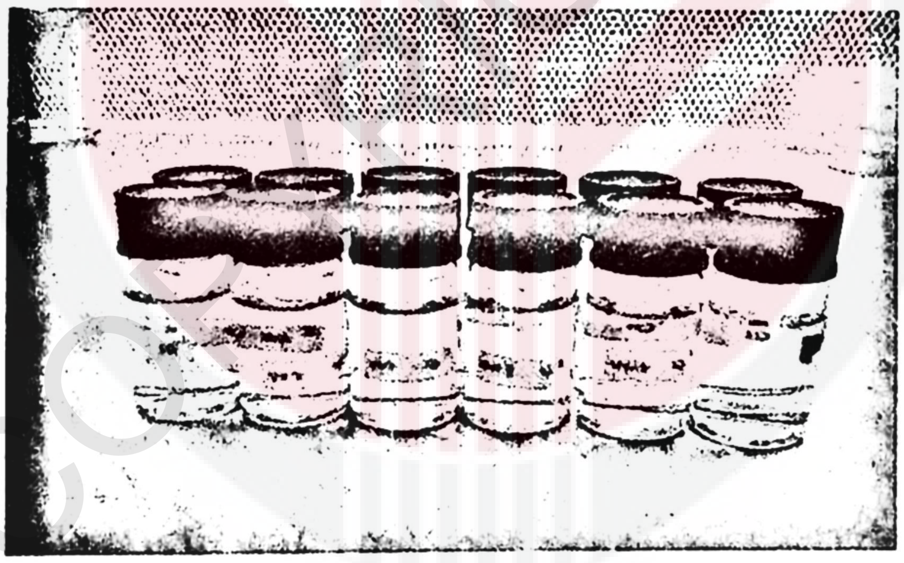


Figure 16: Universal Bottle Used in Serial Dilution

Figure 17: Plate Contain Plate Count Agar

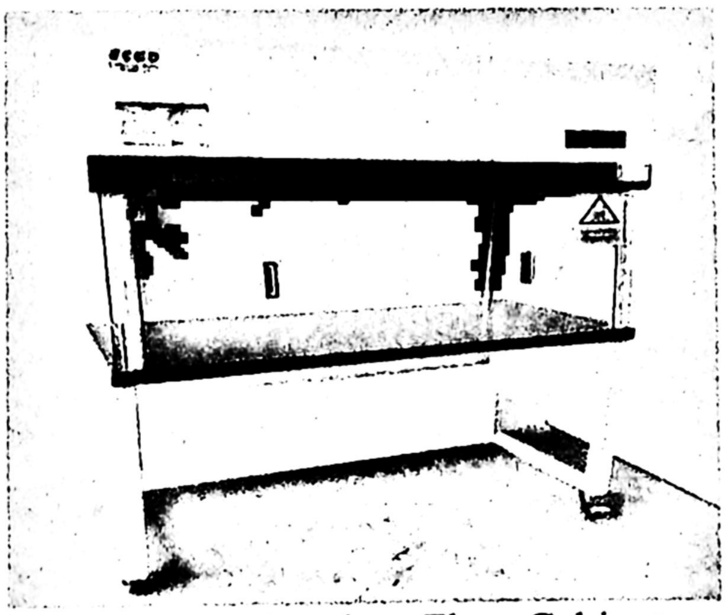


Figure 18: Laminar Flow Cabinet

Figure 19: Incubator

# CHAPTER 4

# RESULT AND DISCUSSION

This chapter presents the data analyse from the experiment conducted throughout the completion of this project. The analysis made started with physical analysis on the bananas such as texture analysis, color analysis and moisture content analysis. Images of the non-vacuum packed (NVP) and vacuum packed (VP) bananas were also taken over the storage period. After that, the antimicrobial analysis which involved Total Plate Count (TPC) was made to determine the total number of colonies formed throughout the storage of the bananas. The NVP and VP banana samples kept under various storage temperatures were analyzed for one month.

## 4.1 Physical Analysis for Unpeeled Banana

### 4.1.1 Texture Analysis

Texture analysis was done to evaluate the textural properties of banana via puncture test. This empirical method was used to determine the firmness of banana. Textural properties are one of the important indicators of food maturity. The penetration force of unpeeled Saba banana stored at different storage temperature throughout the study period was shown in Figures 20(a) – (c). Low penetration force indicates low firmness of banana because the banana only require low force to be penetrated while high penentration force indicate high firmness of banana because the banana require high force to be penetrated.

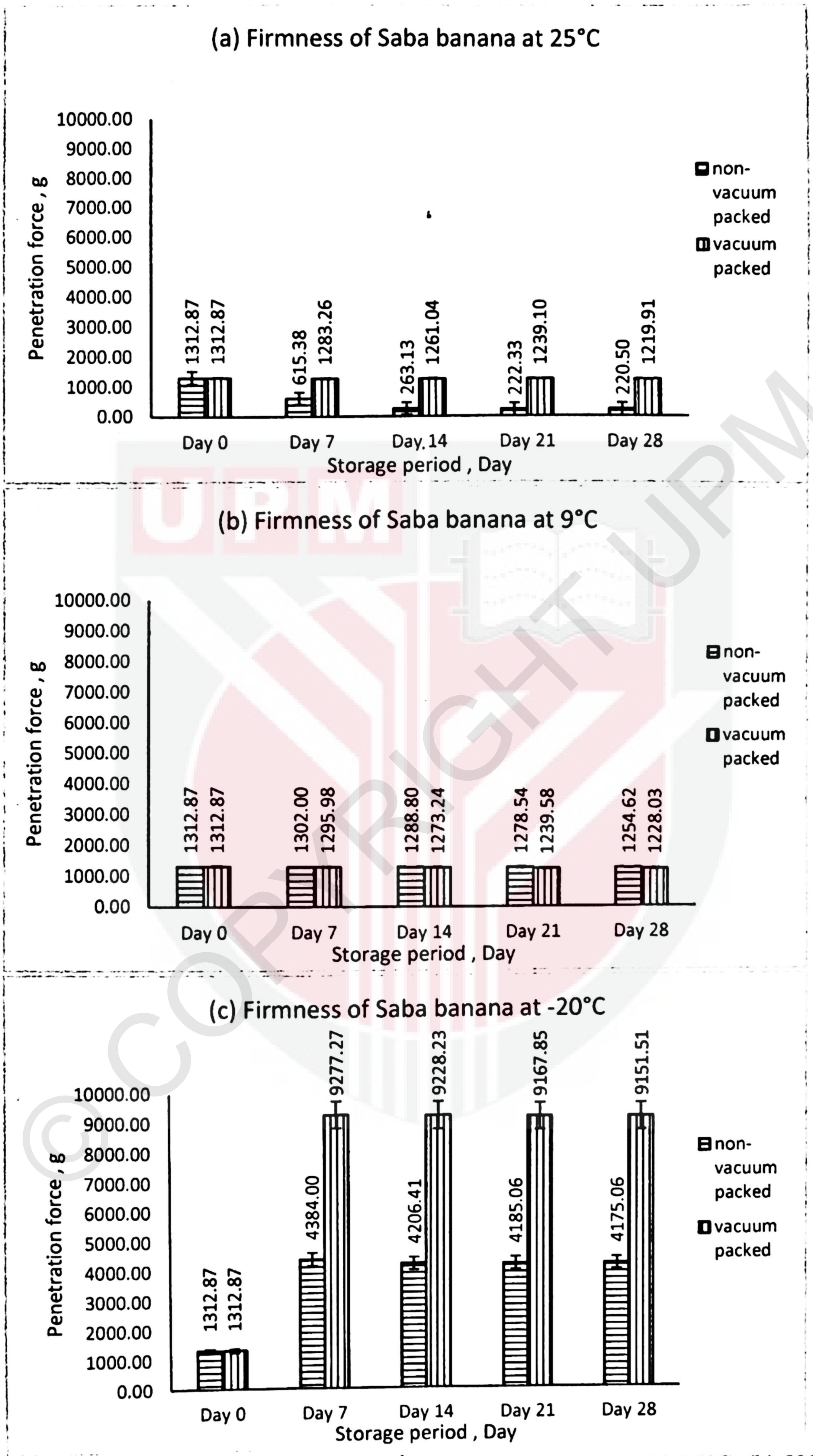



Figure 20: Penetration force of unpeeled Saba banana stored at (a) 25°C, (b) 9°C and (c) -20°C.

It was found that penetration force for both banana samples (NVP and VP) kept at 25°C and 9°C storage temperatures decreased with time due to decrease in firmness as the bananas became matured over time. For the NVP bananas stored at 25°C, the penetration force decreased significantly from $0^{th}$ day to $28^{th}$ day which was from 1312.87g to 220.50g while for the VP banana, the penetration force was slightly decreased from 1312.87g to 1219.91g from $0^{th}$ day to $28^{th}$ day. It was found that penentration force for VP bananas reduced slightly compared to NVP bananas that reduced significantly due to oxygen deficiency that delayed the maturity and ripening process. Based on Figure 20(b) which was for 9°C, the penetration force shown that both samples also decreased with time. The penetration values for NVP bananas decreased slightly from 1312.87g to 1254.62g while the penetration force for VP bananas also decreased slightly from 1312.87g to 1228.03g over one month period of storage. The decrement was not as significant as NVP and VP bananas kept at 25°C because lower temperature helped to delay the maturity and ripening process as the chemical reaction inside the banana was retarded.

However, contradict to the findings in Figures 20(a) and (b), from Figure 20(c) which was for -20°C, the penetration force for both samples was seen to increase from day 0 to day 7 and then from day 7 to the end of storage period the penetration force decreased slightly with time. The increase in penetration force of bananas kept at -20°C was due to the fact that all the moisture content inside the banana already frozen at that temperature, thus increased the firmness of the banana. While from day 7 to day 28 for NVP bananas, the penetration force decreased slightly from 4384.0g to 4175.06g while for VP bananas, the values also decreased slightly from 9277.27g to 9151.51g.

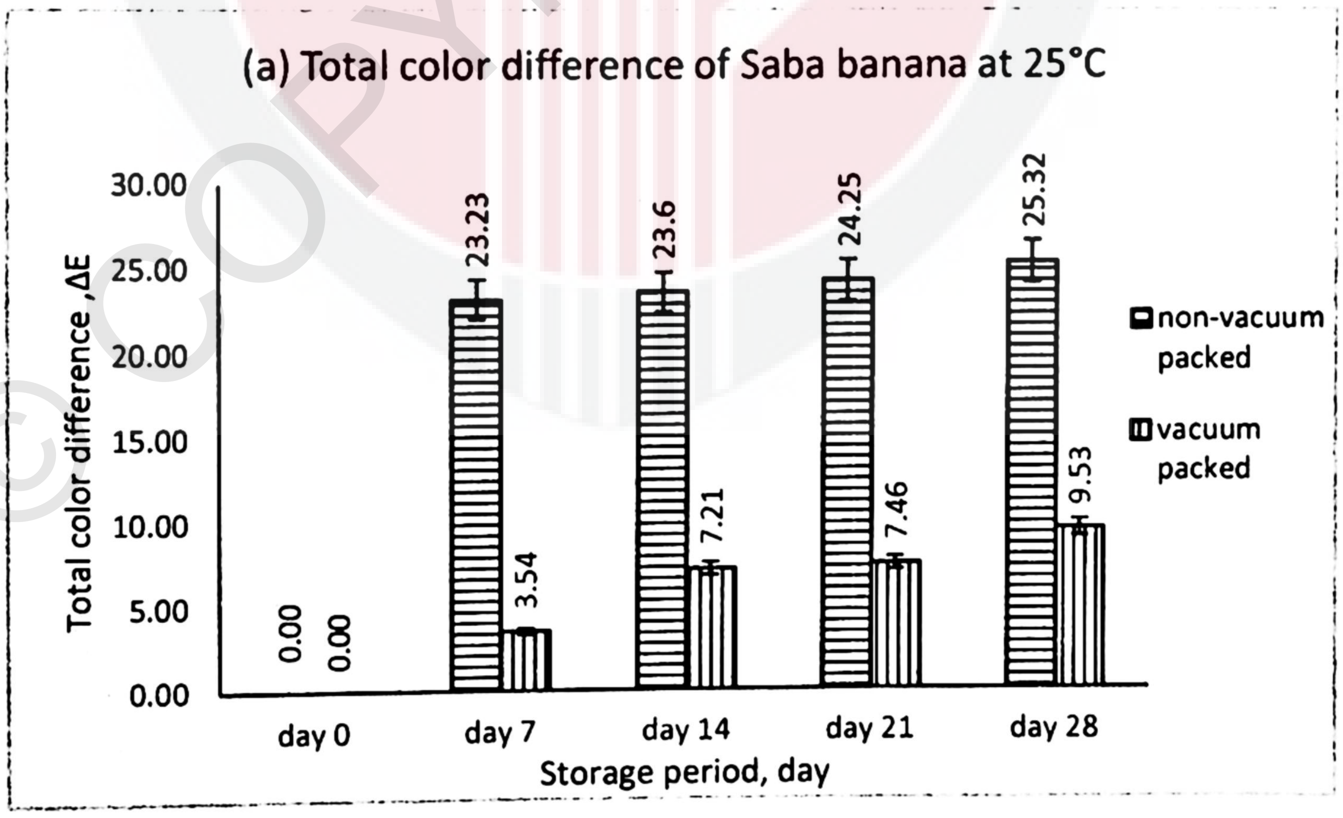
Bananas kept at all storage temperatures exhibited decrease in penetration force due to ripening process of the banana. The main reasons were due to the breakdown of starch into sugars, the breakdown of cell walls or reduction in cohesion of the middle lamella due to solubilization of pectic substances and also because of migration of water from skin to flesh due to osmosis process (Karthiayani et al., 2013). Similar result was also reported by Kharthiani et al., (2013) whom found that the firmness of Robusta and Poovan banana to decrease with increase in storage period. Reasearch done by Soltani et al., (2011) also showed the same result where firmness and hardness of *Cavendish* banana decreased from 7647.87g to 2753.23g as fruit ripened with the same reason as before.

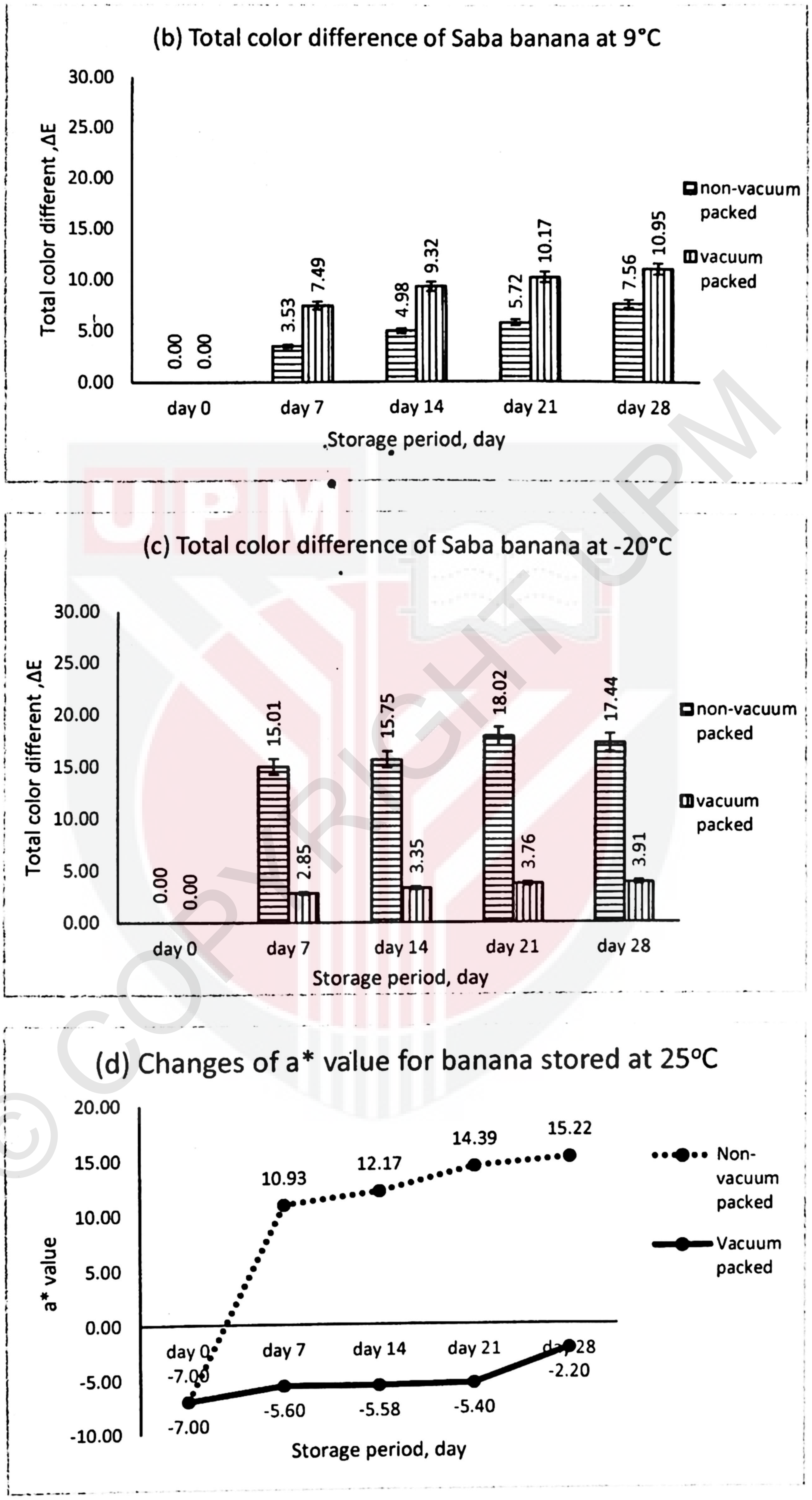
Even though all condition showed a decrease pattern for penetration force, the penetration force of VP bananas still exhibited high value compared to the NVP bananas except for the VP banana stored at 9°C, where the penetration force was lower than the NVP banana. The high value of penetration force for VP proved that the VP bananas were more firm compared to the NVP banana and this was due to the lack of oxygen in the VP bananas and good barrier properties of the vacuum packaging material that helped to retard the ripening process and thus maintaining the quality of the banana from the beginning to the end of storage period. Meanwhile, at 9°C the penetration force of VP bananas was lower than the NVP bananas because of the spoilage that occurred due to the temperature abuse during the storage period as the temperature inside the cold room where the samples were kept was not stable and need time to be stabilized the each time after the cold room was opened.

Comparing between the three storage temperatures, VP bananas kept at the -20°C storage temperature exhibited the highest value in penetration force compared to the others indicating high firmness of banana. This was because the bananas were packed in vacuum packaging thus the quality was maintained from the beginning and the frozen temperature had helped to increase the firmness due to frozen moisture content inside the bananas.

### 4.1.2 Color Analysis

Color is one of the factors that could affect consumer acceptance and preferable towards the food product. Therefore, the changes of color of the unpeeled bananas throughout the study period in this project was investigated. The color changes was analyzed in term of total color difference (ΔE) and the changes of a* value. The a* values (green to red) considers the positive values as red and negative values as green while 0 is neutral. The ΔE and changes of a* value of unpeeled Saba bananas stored at different storage temperature throughout the study period was shown in Figures 21(a) – (f). Based on the figures, ΔE and a* values of both banana samples NVP and VP kept at all storage temperatures increased with time.




(b) Total color difference of Saba banana at 9°C
Total color different ,ΔE
30.00
25.00
20.00
15.00
10.00
5.00
0.00
0.00
0.00
3.53
7.49
4.98
9.32
5.72
10.17
7.56
10.95
day 0
day 7
day 14
day 21
day 28
Storage period, day
non-vacuum packed
vacuum packed
(c) Total color difference of Saba banana at -20°C
Total color different ,ΔE
30.00
25.00
20.00
15.00
10.00
5.00
0.00
0.00
0.00
15.01
2.85
15.75
3.35
18.02
3.76
17.44
3.91
day 0
day 7
day 14
day 21
day 28
Storage period, day
non-vacuum packed
vacuum packed
(d) Changes of a* value for banana stored at 25°C
a* value
20.00
15.00
10.00
5.00
0.00
-5.00
-10.00
-7.00
10.93
12.17
14.39
15.22
-7.00
-5.60
-5.58
-5.40
-2.20
day 0
day 7
day 14
day 21
day 28
Storage period, day
Non-vacuum packed
Vacuum packed

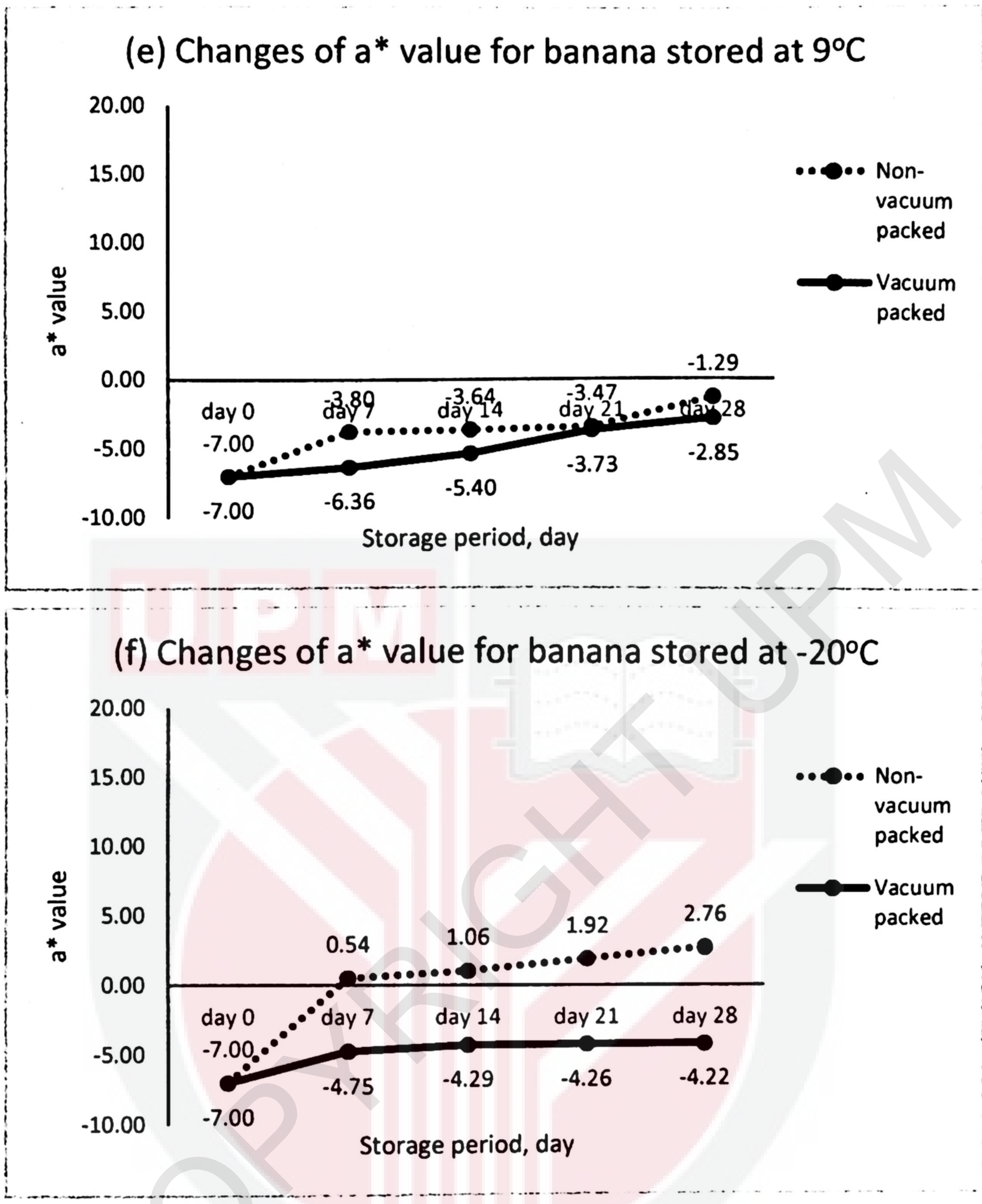



Figure 21: Total color difference of bananas kept at (a) 25°C, (b) 9°C, (c) -20°C and changes of a* value of bananas stored at (d) 25°C, (e) 9°C, (f) -20°C

For the NVP bananas stored at 25°C, ΔE (Figure 21(a)) increased significantly from 0th day to 28th day which was from 0 to 25.23 while the a* value (Figure 21(d)) also increased significantly from -7 to 15.22 indicating change in color from green to red. Whereas for the VP banana, ΔE (Figure 21(a)) increased slightly from 0 to 9.53 and a* value (Figure 21(d)) also increased slightly from -7 to -2.2 which indicates that the banana still preserve the green color at the end of the study period. The increment was not as significant as NVP bananas kept at 25°C because VP delay the ripening process, thus the change in color was not significant and the green color was still preserve.

Figure 21(b) and (e) which was for 9°C, the ΔE and a* value of both samples also increased from 0th day to 28th day. The ΔE of NVP values was increased slightly from 0 to 7.56 respectively while the a* value was increased from -7 to -2.85 which indicates that the banana still preserve the green color. The same goes for the ΔE (Figure 21(b)) for VP banana which was also increased from 0 to 10.95 and the a* value (Figure 21(e)) increased from -7 to -1.29 where both ΔE and a* value were higher than the NVP value. This higher value was related to the temperature abused occured during storage in cold room as stated before. It was found that the changes in ΔE and a* value for NVP banana samples kept at 9°C were lower than ΔE and a* value for NVP banana samples kept at 25°C due to the low storage temperature. Lower storage temperature retard in ripening, thus less changes in green color pigment occurred.

Meanwhile from Figures 21(c) and (f), which was for -20°C, the ΔE and a* value for both samples were also observed to increase from 0th day to 28th day where the ΔE of NVP banana increased significantly from 0 to 17.44 and the a* value was increased from -7 to 2.76 indicating that the green color had started to change into a tinge of red intensity. This changes occurred due to the very low storage temperature that caused chilling injury to the banana kept without any packaging[14]. While for VP banana, ΔE was increased slightly from 0 to 3.91 and the a* value increased from -7 to -4.22 which indicates that the bananas still preserve the green color.

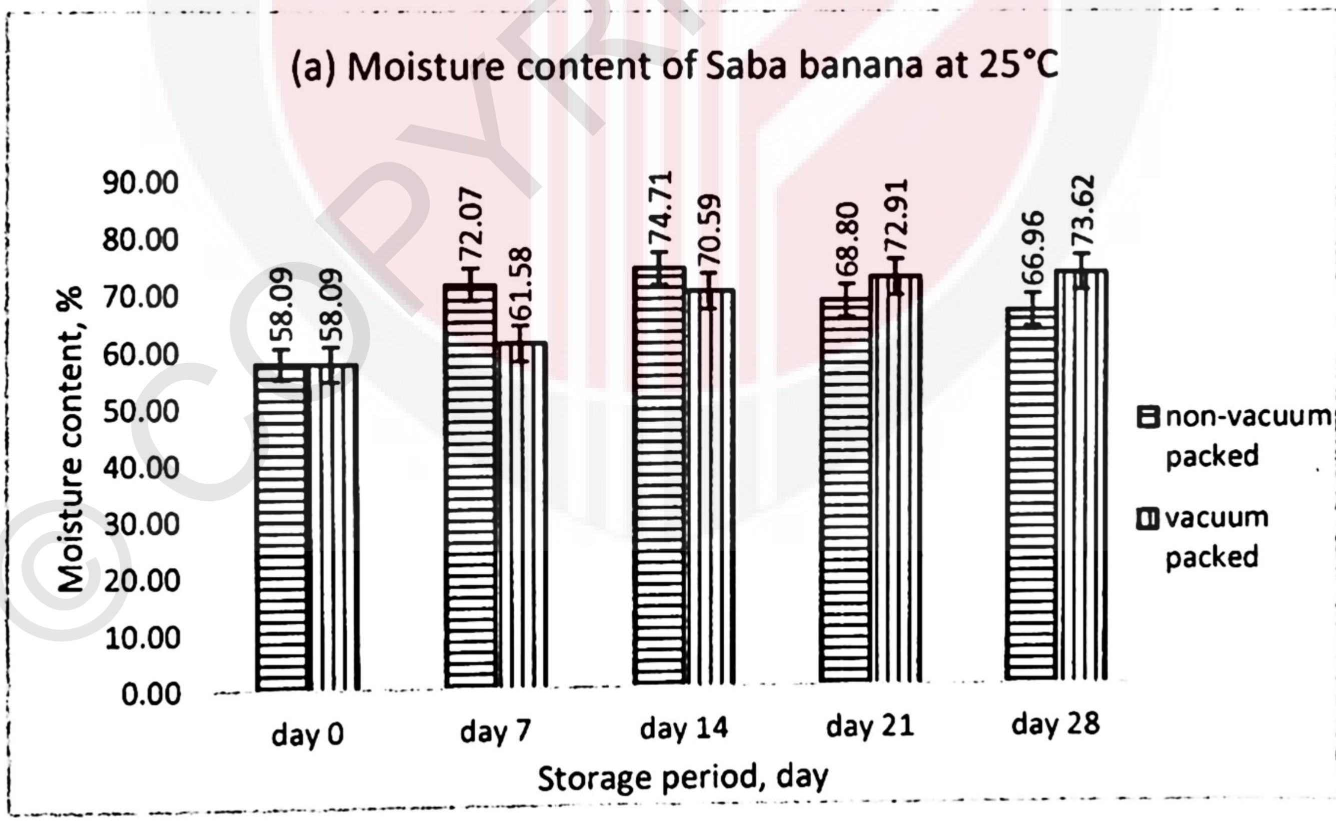
Overall, ΔE and a* value increased from 0th day to 28th day storage period. This increment was due to the metabolic processes linked to biosynthesis that had caused changes in chlorophyll pigments to carotenoid pigment in the fruit which demonstrated that ripening process occurred from time to time in which the rate of the ripening process was dependent on the storage temperature and packaging design[4]. Research done by Kudachikar et al., (2011) also reported that Robusta and Poovan banana exhibited increment in a* value of the peels of packed green banana across storage period. Besides that, similar trend of result was also reported by Soltani et al., (2013) that, there were color changes in the Cavendish banana due to the same reason[32].

The ΔE can also be an indicator to the changes in banana colors as the lower the total color difference or ΔE value, the closer the sample was to the standard[28,33]. Even though the color changes increased with the increase in time, VP banana exhibited lower ΔE and a* value compared to the NVP banana except for cold temperature (9°C) where the ΔE and a* value was higher than VP banana. The lower value for vacuum packed bananas proved that the absence of oxygen and the good barrier properties of packaging material limited the oxygen and moisture content to pass through the packaging thus, reduce the ripening process reaction and browning reaction. Meanwhile, the higher value for VP bananas stored at cold temperature (9°C) indicated the spoilage that occurred throughout the storage period as the VP banana turn blackish from time to time as shown in Figure 26. At 9°C although the NVP bananas exhibited lower ΔE and a* value than VP bananas, the number of black spot on the peels of NVP bananas increased from time to time as shown in Figure 25 due to lack of pre-treatment such as washing with fungicide solution.

Comparing between bananas kept at the three storage temperatures, VP bananas stored at -20°C exhibited the lowest value in ΔE and a* value compared to the other storage temperature due to the bananas were packed in vacuum packaging with good barrier properties and in addition, the frozen temperature had helped to inhibit or inactive the growth of bacteria thus the quality was maintained from the beginning to end of the study period.

### 4.1.3 Moisture Analysis

Moisture content is one of the most commonly measured properties of food materials. It is an important criteria to food for a number of different reasons and one of it is it determines food quality. The texture, taste, appearance and stability of foods are dependent on the amount of water they contain. The higher the moisture content, the higher the possibility for the banana to degrade with time. Therefore, the moisture analysis was done to relate with the firmness/texture of the banana. The percentage of moisture content for unpeeled NVP and VP bananas stored at 25°C, 9°C, and -20°C over 1 month storage period is shown in Figures 22(a) – (c).



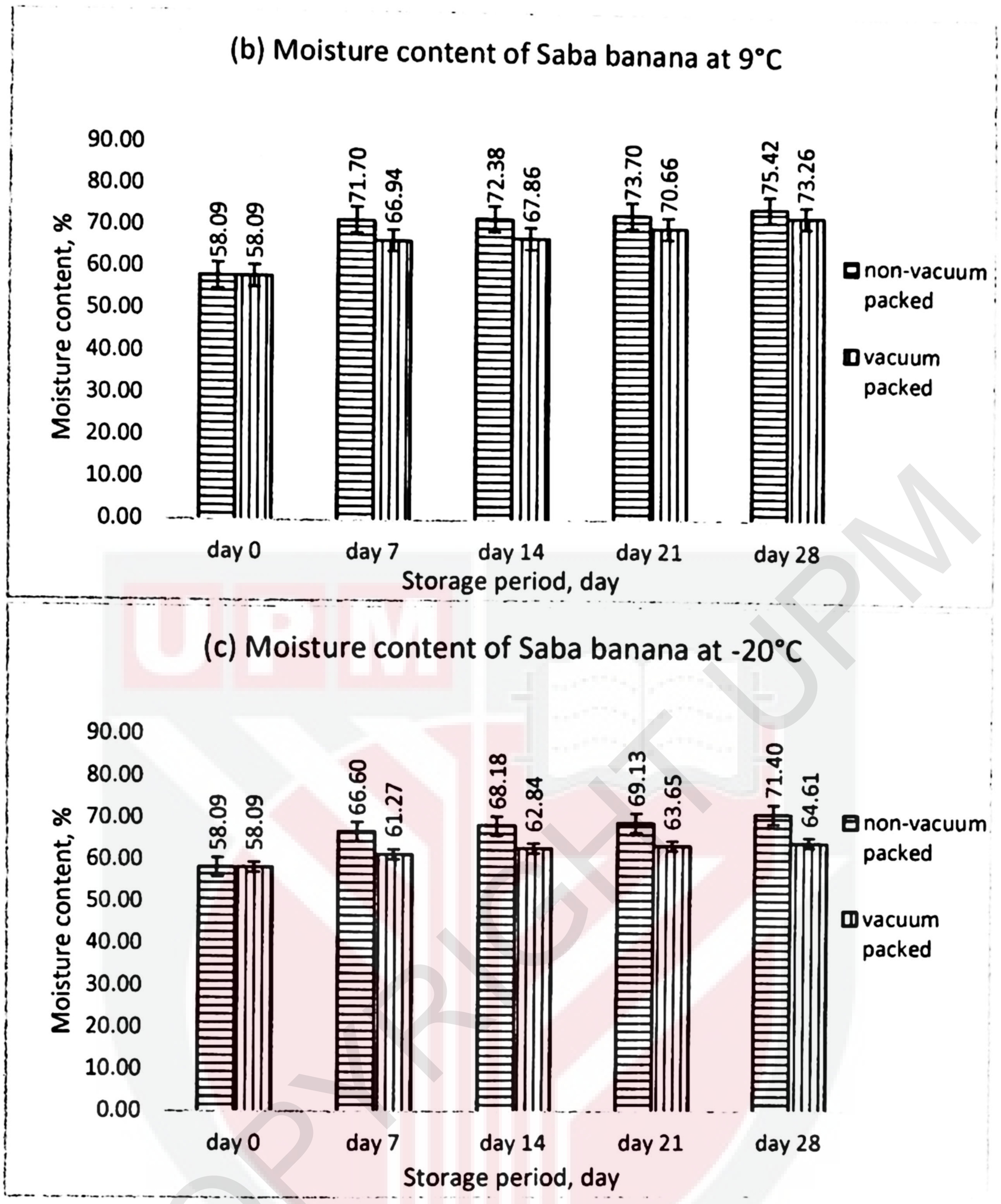


Figure 22: Moisture content of bananas stored at (a) 25°C, (b) 9°C, and (c) -20°C

It can be seen that the amount of moisture content increased with the increase in storage period due to ripening process occured. From Figure 22(a) which was for the bananas stored at 25°C, the percentage of moisture content for NVP banana increased from 58.09% on the $0^{th}$ day to 74.71% on the $14^{th}$ day due to the moisture at surrounding being absorbed[28] and also the ripening process occurred which resulted to change from starch into glucose[25]. After $14^{th}$ day the moisture decreased to 66.96% at the end of the study period. As shown in Figure 23 the decrement was due to the fact that the bananas after 1 month period of storage had became overripe and spoiled thus dried from time to time. Whilst for the VP banana, the moisture content was slightly increased from 58.09% to 73.62% from 0th day to 28th day respectively.

Based on Figure 22(b) which was for 9°C, the moisture content shown by both samples also increased from time to time. The NVP values increased from 58.08% to 75.42% while the moisture content for VP banana also increased from 58.08% to 73.26%. The increment in moisture content for NVP bananas was due to high humidity at 9°C and thus the bananas absorbed the moisture at surrounding[28]. While for the VP bananas the increment in moisture content was because of the bananas that have became spoiled and bruises occurred. As can be seen from Figure 22(c) which was for -20°C, the moisture content for both samples was also increased from day 0 to day 28th. For the NVP bananas, moisture content was increased from 58.09% to 71.40% whilst for VP banana, the moisture was increased from 58.09% to 64.61%. The increment in moisture content in bananas was also due to high humidity at -20°C and the bananas absorbed the moisture at surrounding[28].

The increase in moisture content at the end of the storage period for all banana samples was also due to the ripening process that occurred throughout the storage period and this occurrence was because of osmotic transfer of moisture from peel to pulp during the storage (Sau et al. 2017; Kudachikar, 2011). Besides that, the storage conditions that have high relative humidity might also favor the moisture migration from the environment to the banana. This led to increase in moisture content and also known to lead to increase of reactant mobility (Bassey et al. 2013). In addition, the increase in moisture content was also due to the breakdown of starch into sugars hence high moisture content (Karthiayani et al., 2013). Similar result was by Kudachikar et al. (2011), where they found that there was an increase value in moisture content for Robusta and Poovan bananas as the storage period was increased. Research conducted by Bassey at al., (2013) also gave similar result pattern when they analyzed moisture content of other type of cooking banana.

Even though all conditions showed an increased pattern for moisture content, the VP bananas exhibited lower moisture compared to the NVP bananas except for the VP bananas stored at 25°C, where the moisture content at the end of the storage period was higher than the NVP bananas. The lower moisture content of VP bananas than NVP bananas proved that the VP bananas were more firm and contain less water content compared to the NVP banana due to the absence of oxygen and good barrier properties of the vacuum packaging material that helped to retard the ripening process and thus maintained the quality of the banana from the beginning to the end. Meanwhile, at 25°C the moisture content of NVP bananas was lower than the VP bananas due to the spoilage that occurred to the NVP samples thus dried the banana as storage period increased .

Comparing between the three storage temperatures, the -20°C storage temperature for VP bananas exhibited the lowest moisture content compared to the others indicating that the VP bananas still maintaining their quality and firmness and that exhibited longer shelf life due to the slower ripening process. This was because the bananas were vacuum packed. The absence of oxygen delayed the ripening process and reduce the moisture content. Besides, the good moisture barrier properties of packaging material inhibited the movement of moisture from surrounding to pass through the package, thus, the quality was maintained.

### 4.1.4 Observation on Banana Images Over the Storage Period

Physical analysis done also include capturing and observing images of the NVP and VP bananas on day 0, 7, 14, 21 and 28th over the storage period as shown in Figures 23 to Figure 28. It was found that these figures supported all the other physical analysis results explained in Section 4.1.1 – 4.1.3 which include texture analysis, color analysis and moisture content analysis.

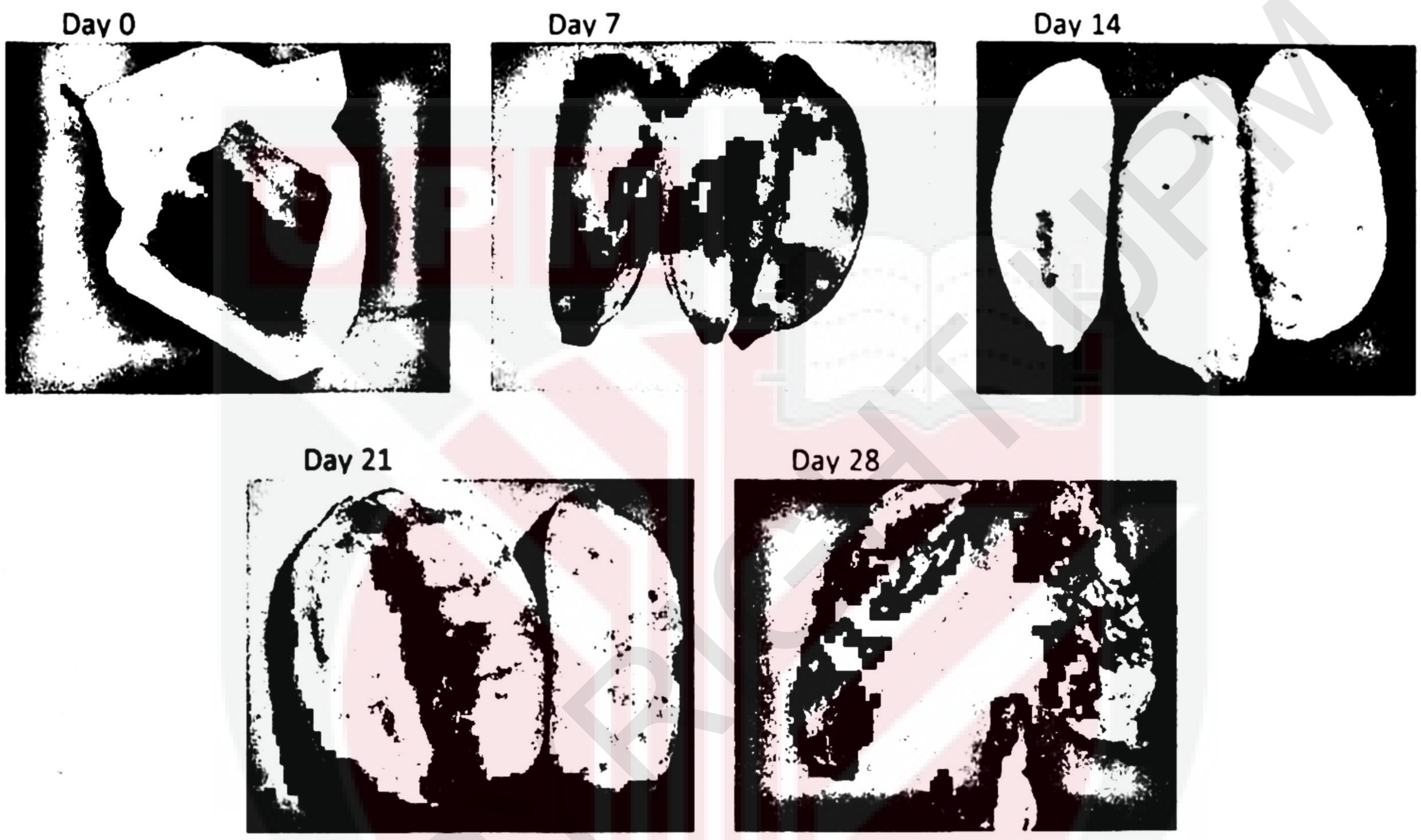



Figure 23: Non-vacuum packed banana stored at 25°C

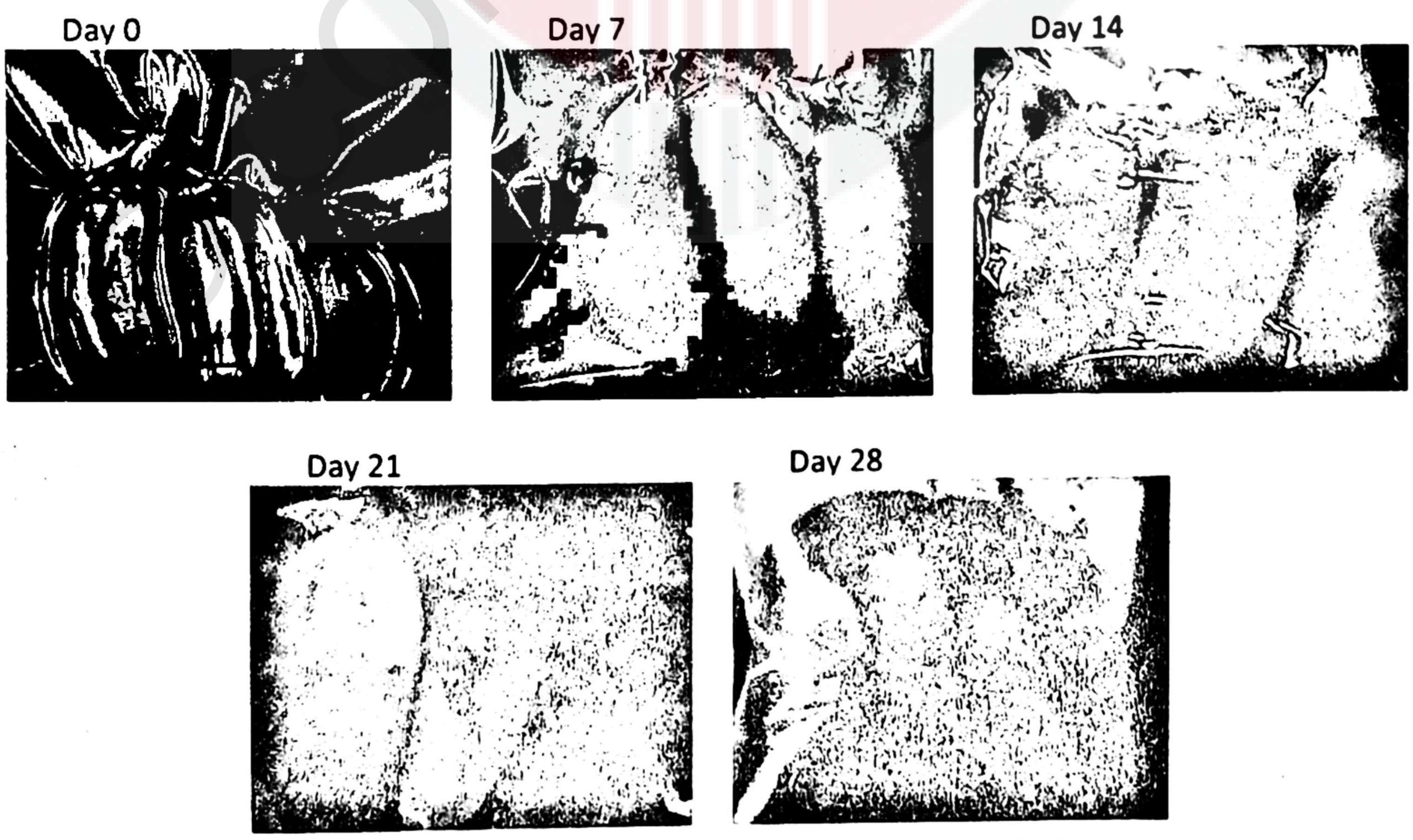



Figure 24: Vacuum packed banana stored at 25°C

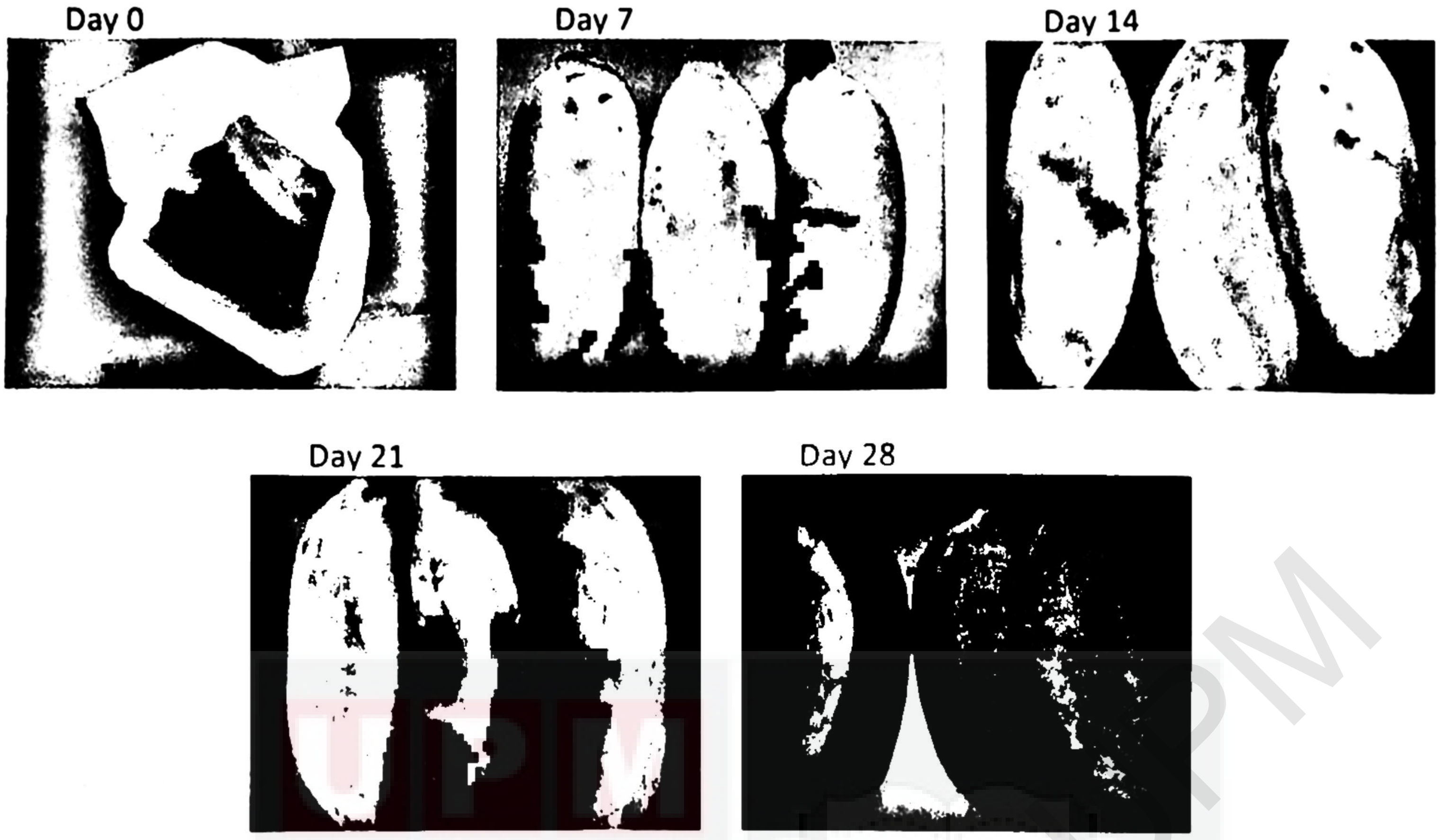



Figure 25: Non-vacuum packed banana stored at 9°C

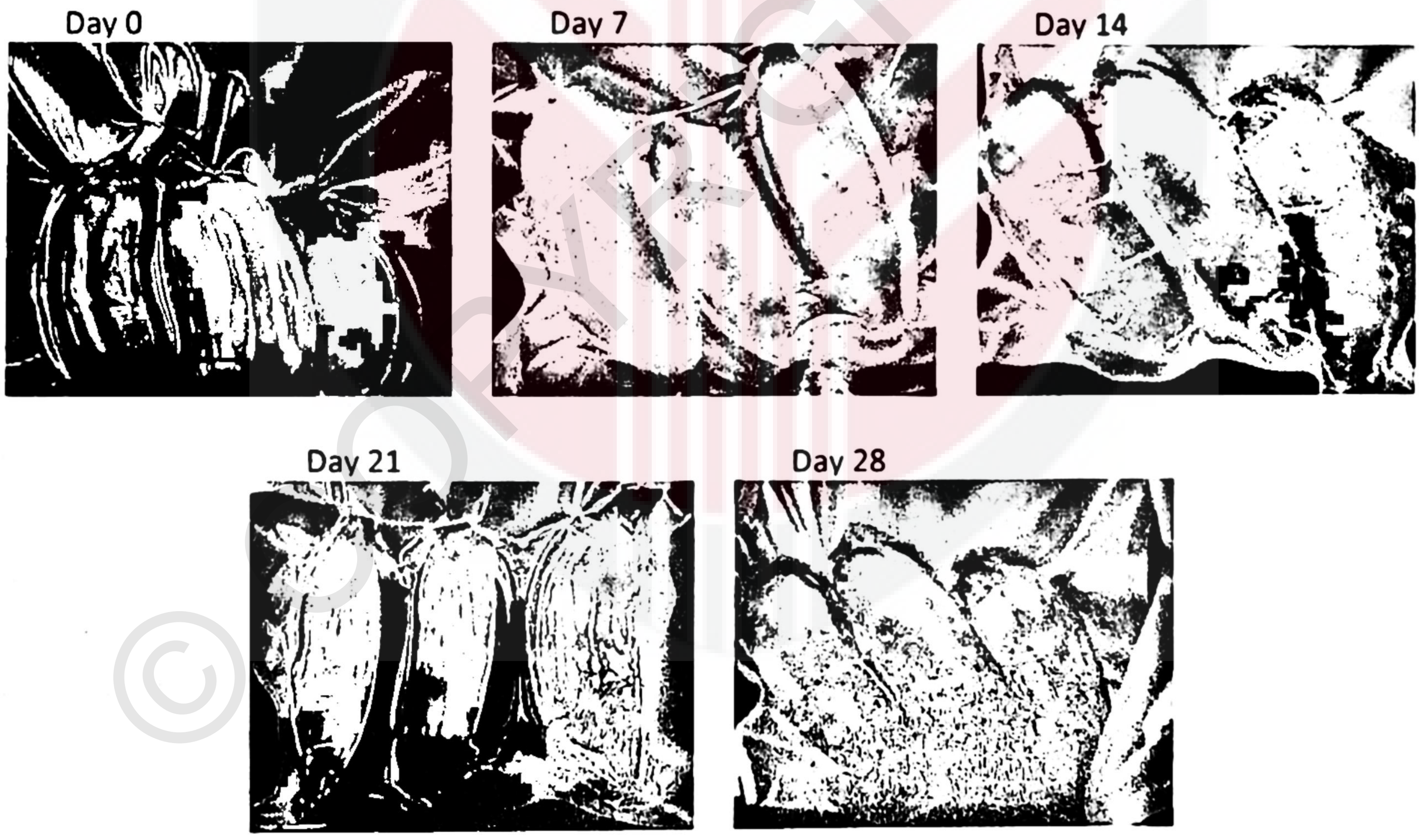



Figure 26: Vacuum packed banana stored at 9°C

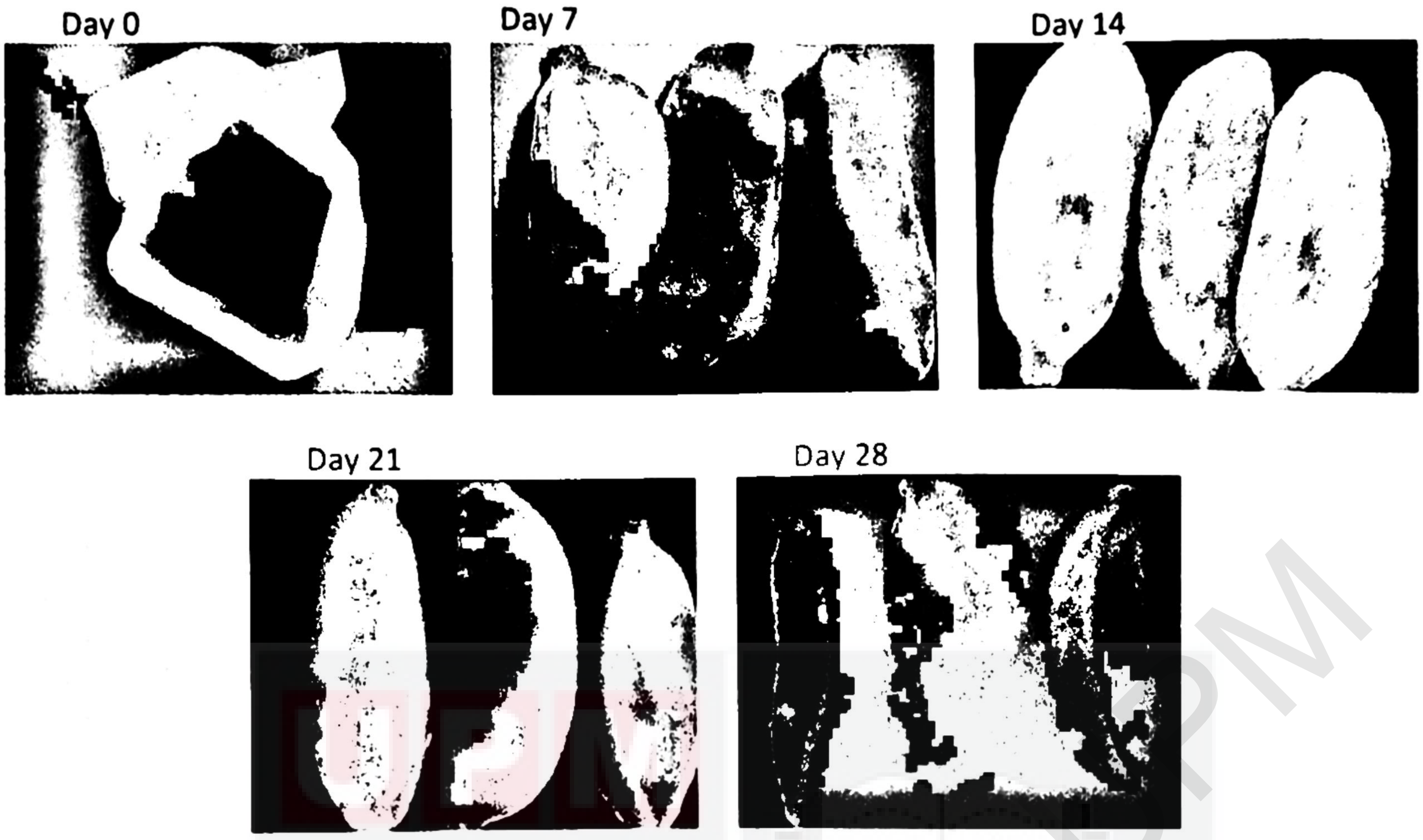



Figure 27: Non-vacuum packed banana stored at -20°C

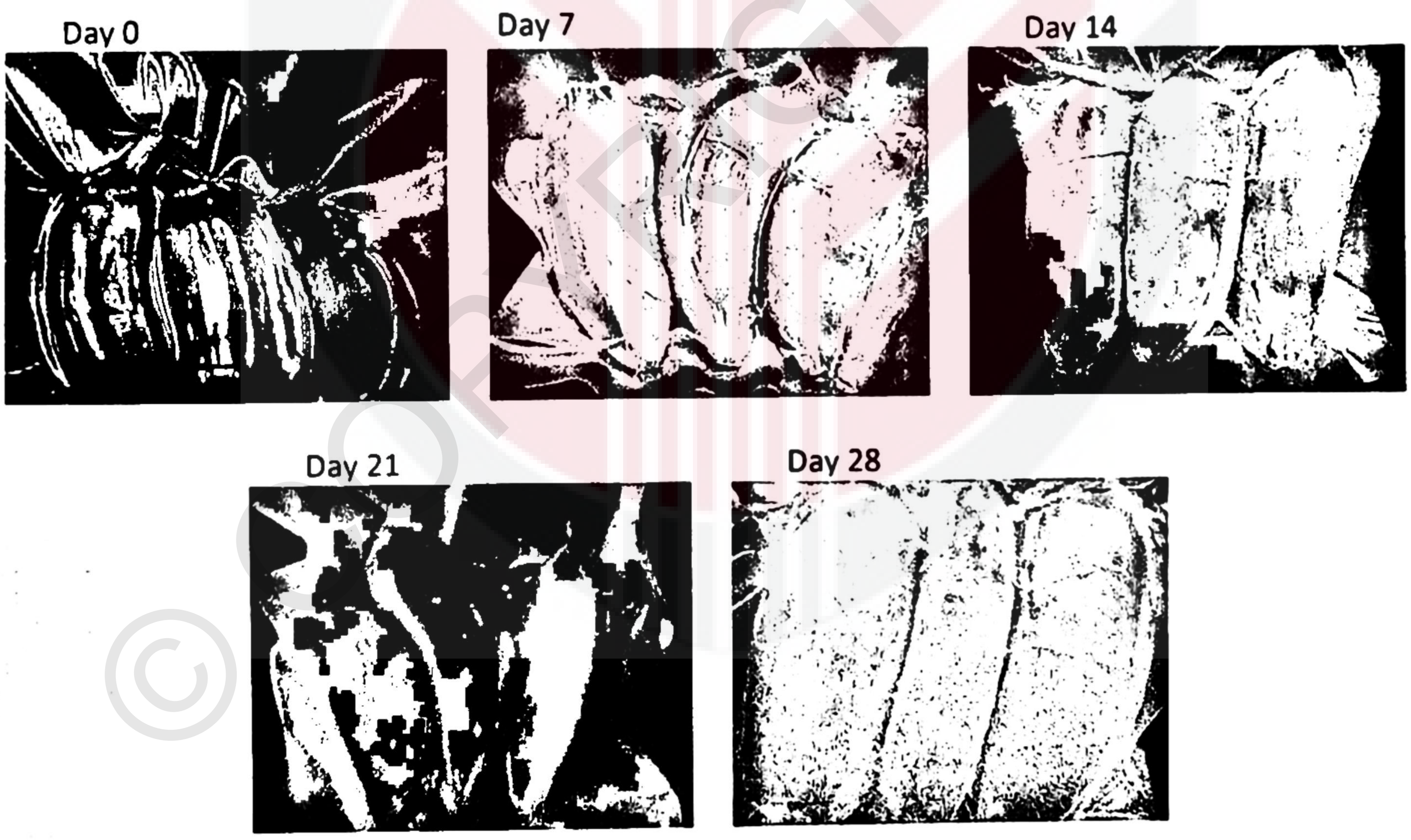
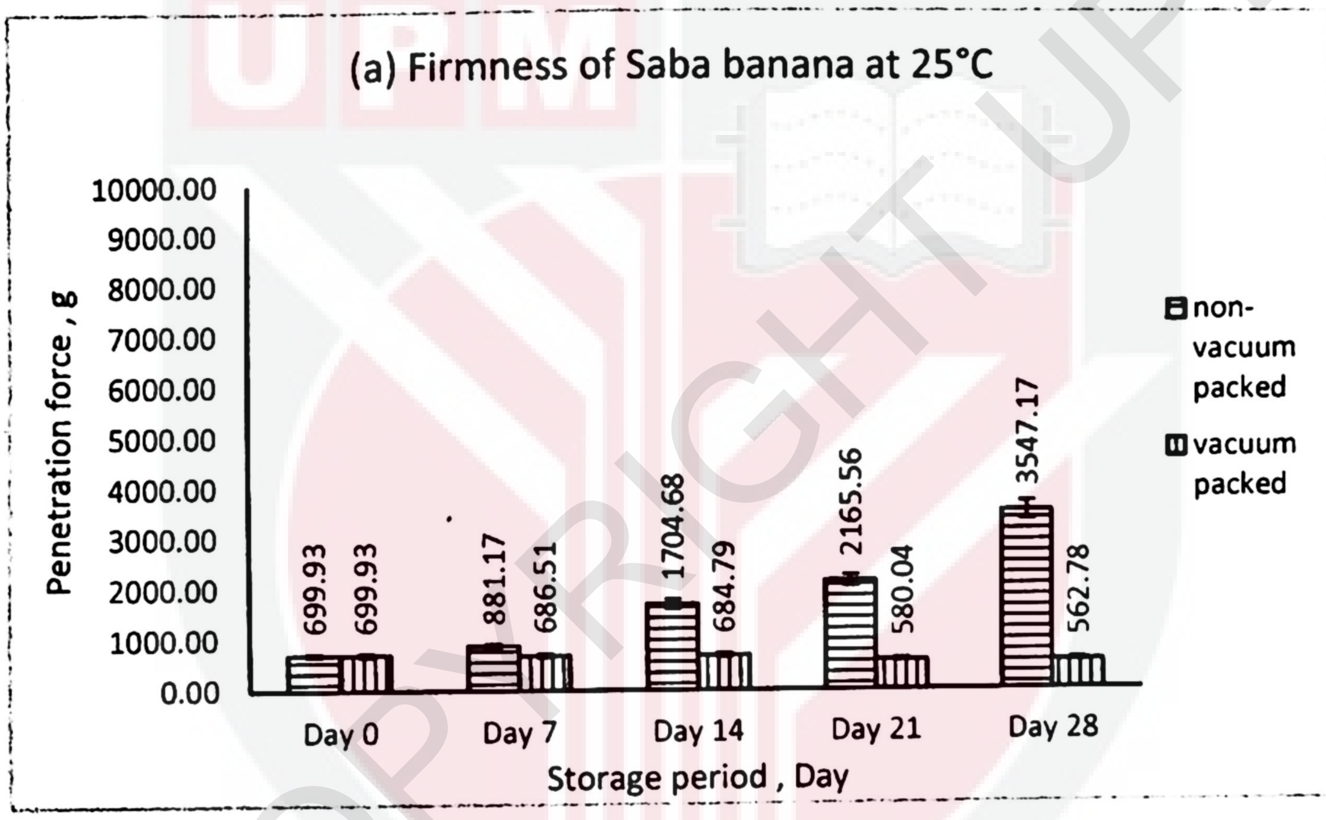


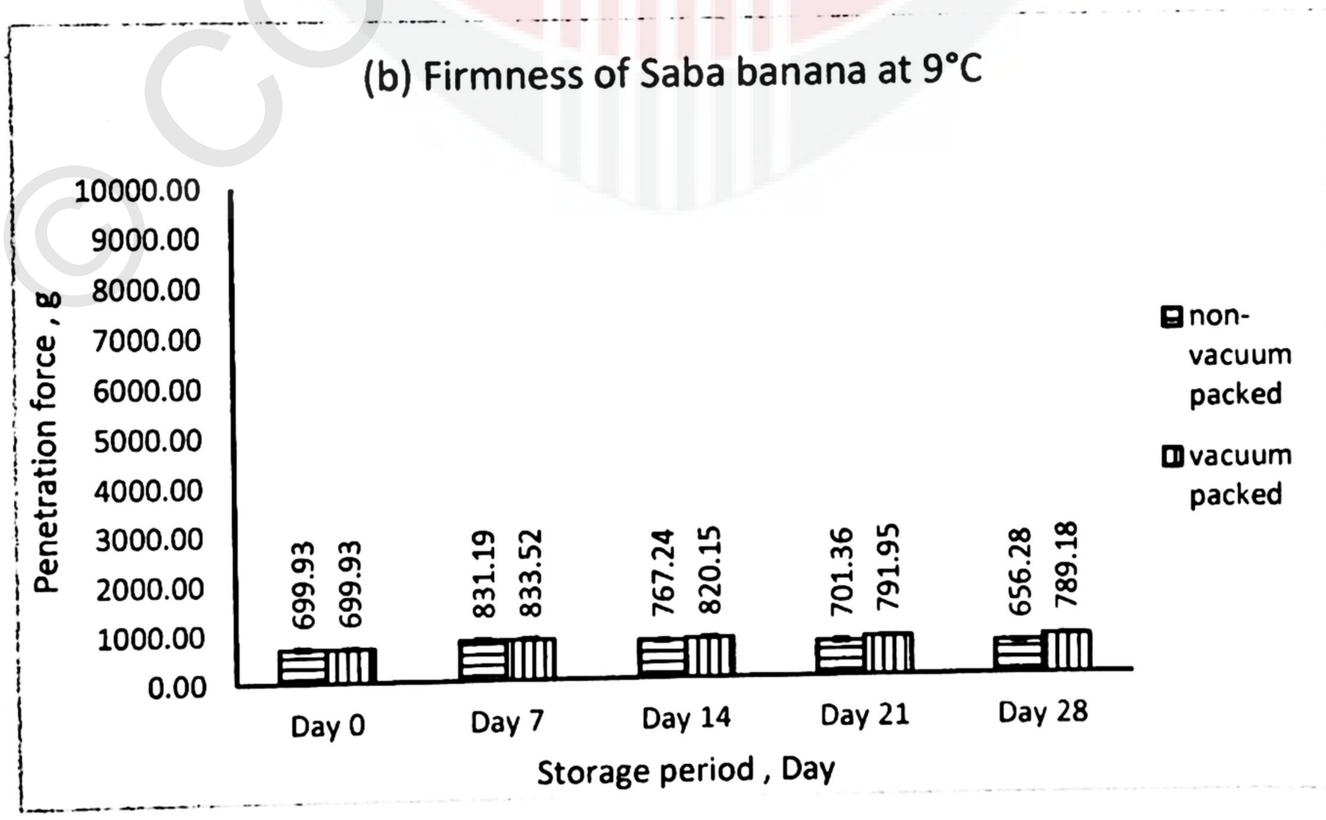

Figure 28: Vacuum packed banana stored at -20°C

## 4.2 Physical Analysis for Peeled Banana

### 4.2.1 Texture Analysis

The penetration force of peeled Saba banana stored at different storage temperature throughout the study period was shown in Figures 29(a) – (c). Low penetration force indicates low firmness of banana because the banana only requires low force to be penetrated while high penetration force indicate high firmness of banana because the banana require high force to be penetrated.



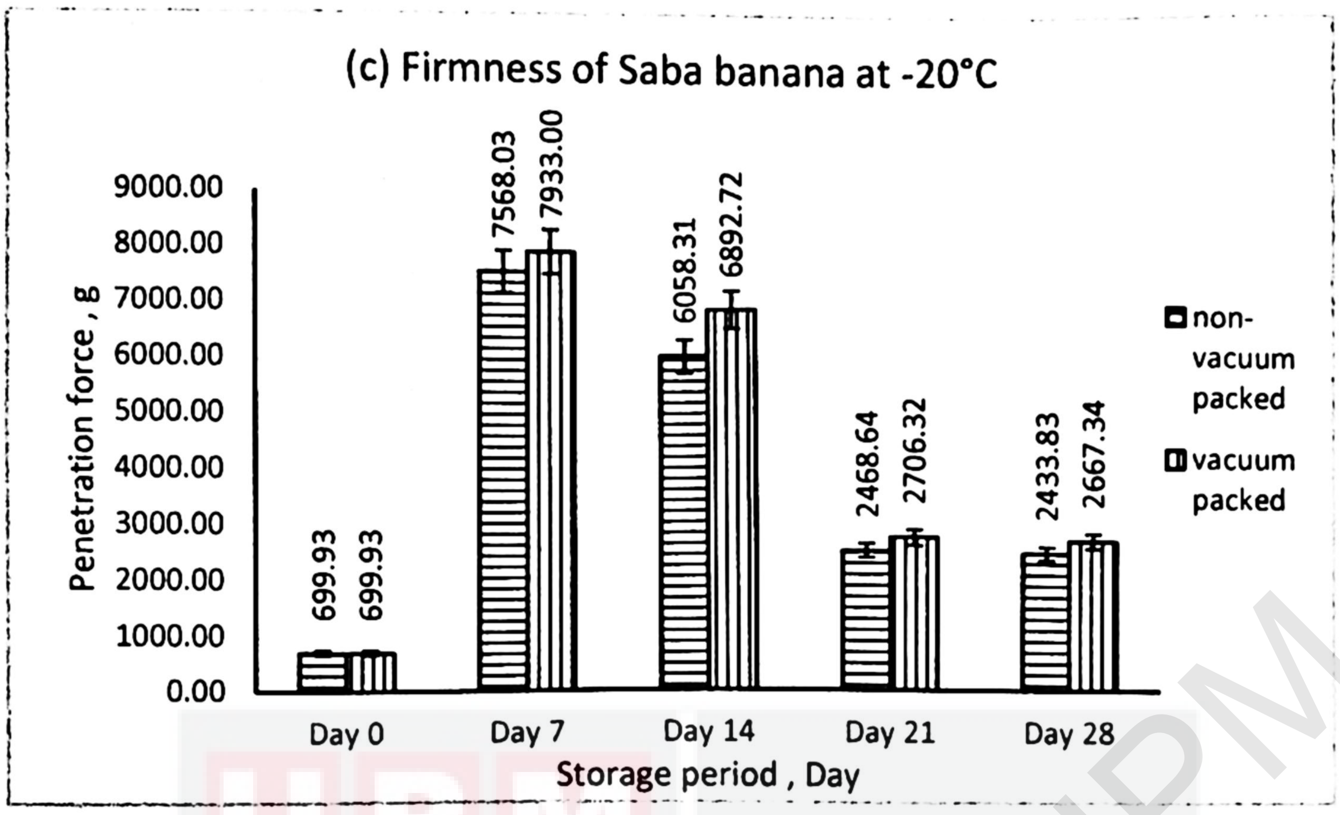



Figure 29: Penetration force of peeled Saba banana stored at (a) 25°C, (b) 9°C and (c) -20°C.

It was found that penetration force for both banana samples (NVP and VP) kept at 25°C, 9°C and -20°C storage temperatures decreased with time due to decrease in firmness as the bananas became matured over time except for the NVP bananas kept at 25°C where the penetration force increased significantly with time from 699.93g to 3547.17g due to the increase in firmness as the bananas became spoiled and dried over time. For the VP bananas stored at 25°C, the penetration force decreased slightly from 0$^{th}$ day to 28$^{th}$ day which was from 699.93g to 562.78g. It was found that penetration force for VP bananas reduced slightly due to oxygen deficiency that delayed the maturity and at the same time ripening process retarded as the main source of the ethylene from peels had been removed. Based on Figure 29(b) which was for 9°C, the penetration force shown that both samples also decreased with time except from day 0 to day 7 where the penetration force increased. The increase in penetration force of bananas kept at 9°C was due to the fact that all the moisture content inside the banana slowly froze at that temperature, thus increased the firmness of the banana. The penetration force from day 7 to day 28 for NVP bananas decreased slightly from 831.19g to 656.28g while the penetration force for VP bananas also decreased slightly from 833.52g to 789.18g over one month period of storage. The decrement was not as significant as NVP and VP bananas kept at 25°C because lower temperature helped to delay the maturity and ripening process as the chemical reaction inside the banana was lowered which assists in extending product shelf-life [24].

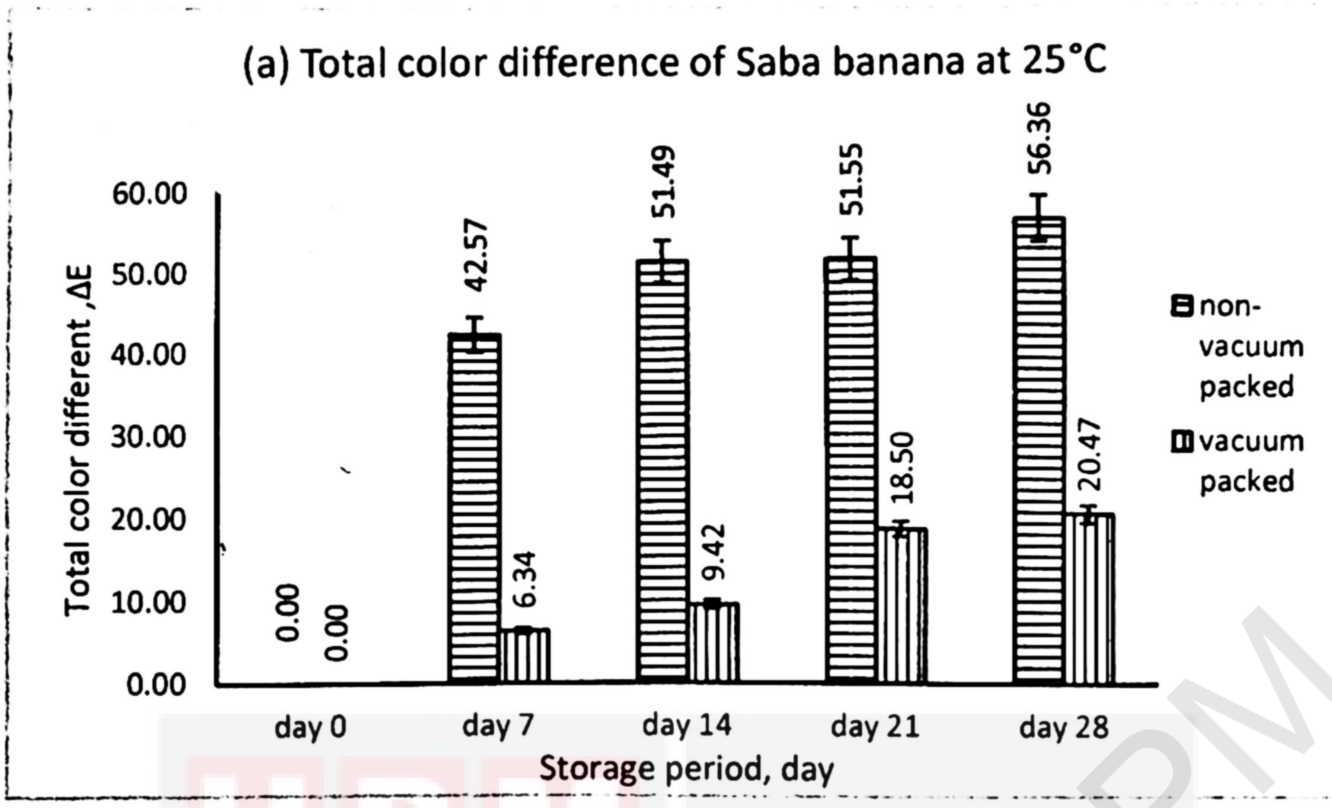
Similar to the findings in Figures 29(b), from Figure 29(c) which was for -20°C, the penetration force for both samples was seen to increase from day 0 to day 7 and then from day 7 to the end of storage period the penetration force decreased significantly with time. The increase in penetration force of bananas kept at -20°C was due to the fact that all the moisture content inside the banana already frozen at that temperature, thus increased the firmness of the banana. While from day 7 to day 28 for NVP bananas, the penetration force decreased significantly from 7568.03g to 2433.83g while for VP bananas, the value also decreased slightly from 7933.00g to 2667.34g.

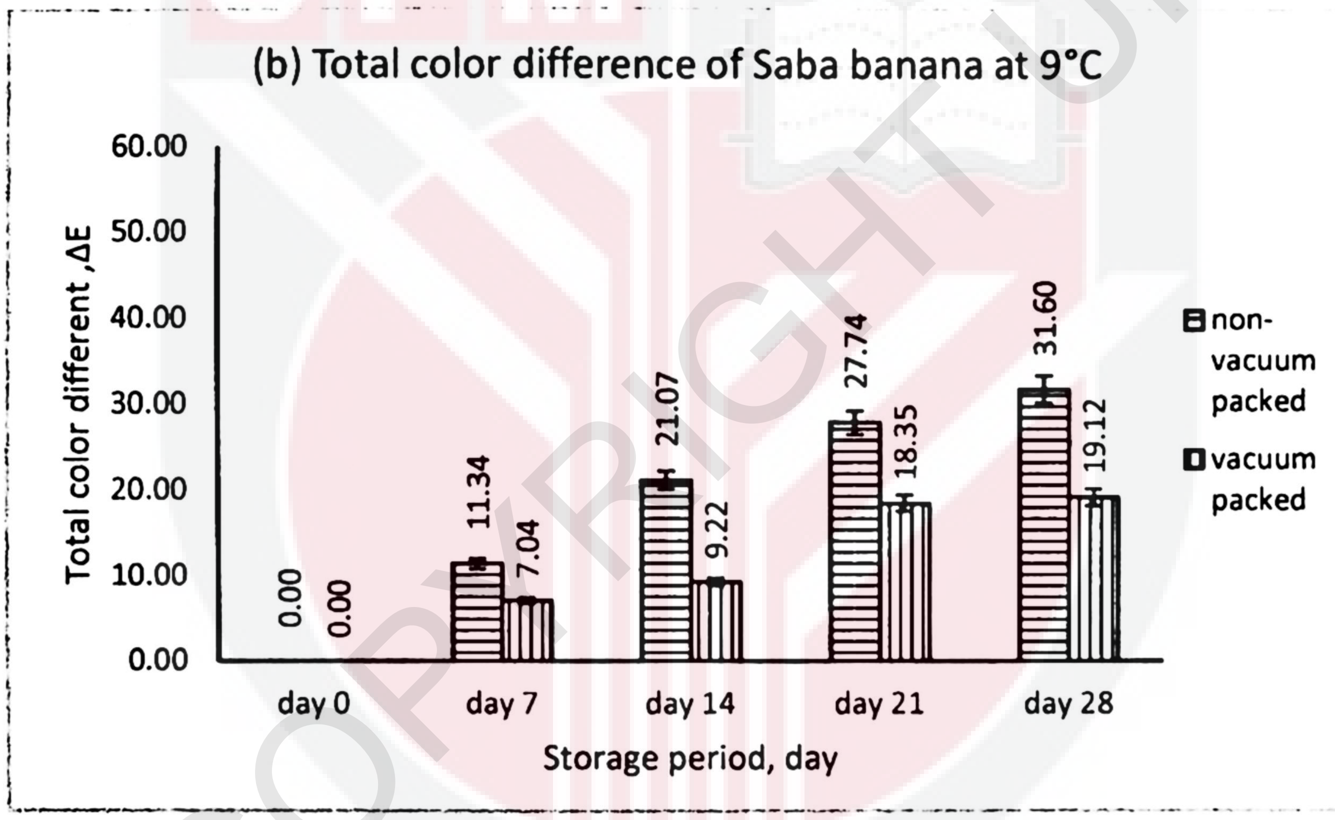
Bananas kept at all storage temperatures exhibited decrease in penetration force due to ripening process of the banana even though the main source of ripening agent from peels had been removed. The main reasons were due to the breakdown of starch into sugars, the breakdown of cell walls or reduction in cohesion of the middle lamella due to solubilization of pectic substances (Karthiayani et al., 2013). Even though all condition showed a decrease pattern for penetration force, the penetration force of VP bananas still exhibited high value compared to the NVP bananas except for the VP banana stored at 25°C, where the penetration force was lower than the NVP banana. The high value of penetration force for VP proved that the VP bananas were more firm compared to the NVP banana and this was due to the lack of oxygen in the VP bananas and good barrier properties of the vacuum packaging material that helped to retard the ripening process and thus maintaining the quality of the banana from the beginning to the end of storage period. Meanwhile, at 25°C the penetration force of VP bananas was lower than the NVP bananas due to the unsuitable storage temperature that had caused the bananas became spoiled and dried over time.

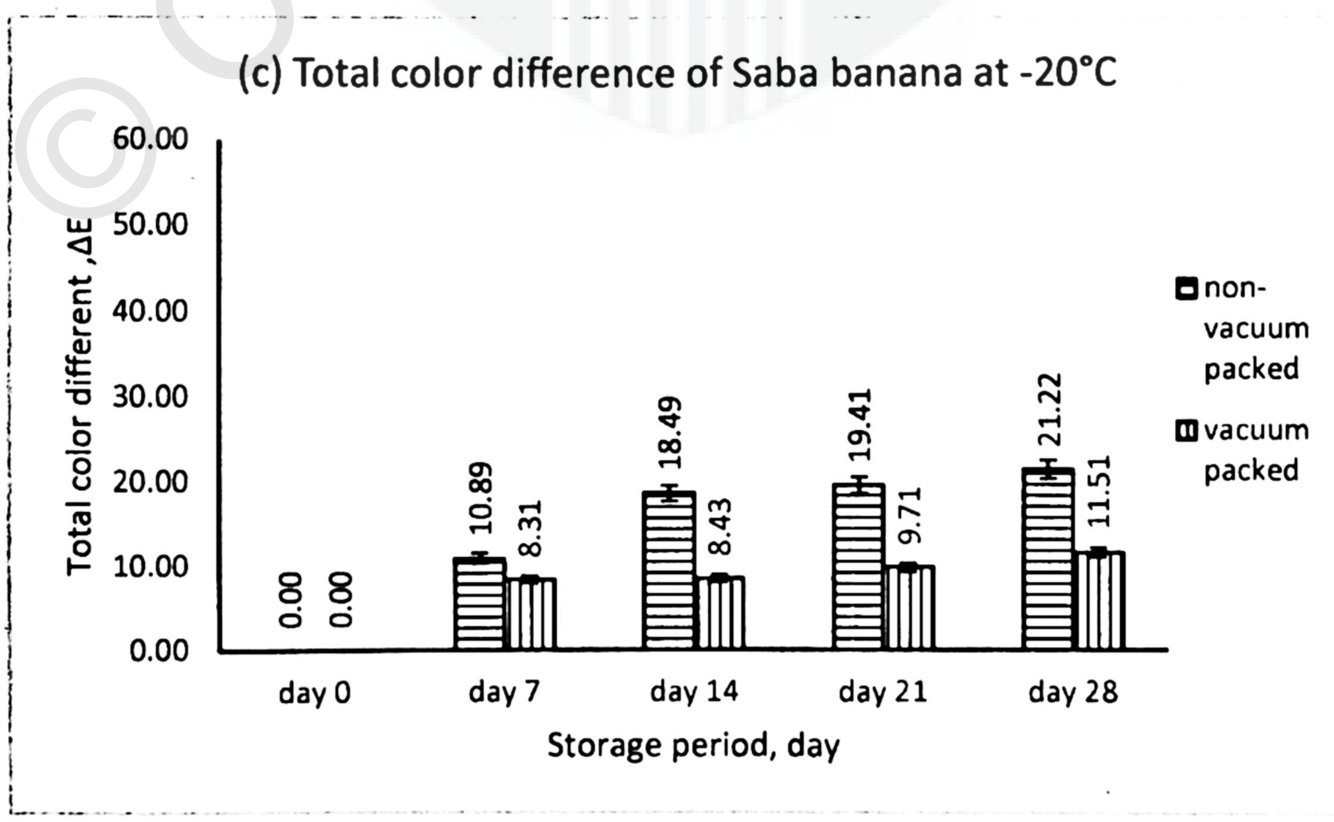
Comparing between the three storage temperatures, VP bananas kept at the -20°C storage temperature exhibited the highest value in penetration force compared to the others indicating high firmness of banana. This was because the bananas were packed in vacuum packaging thus the quality was maintained from the beginning and the frozen temperature had helped to increase the firmness due to frozen moisture content inside the bananas.

### 4.2.2 Color Analysis

The changes of color of the bananas throughout the study period in this project was investigated. The color changes was analyzed in term of total color difference (ΔE) and the changes of L* value. The L* values (lightness) which axis goes vertically from 0 (perfect black) to 100 (perfect white). The ΔE and changes of L* value of peeled Saba bananas stored at different storage temperature throughout the study period was shown in Figures 30(a) – (f). Based on the figures, ΔE value of both banana samples NVP and VP kept at all storage temperatures increased with time while L* value of both banana samples NVP and VP kept at all storage temperatures decreased with time.


(a) Total color difference of Saba banana at 25°C
Total color different ,ΔE
60.00
50.00
40.00
30.00
20.00
10.00
0.00
0.00
0.00
42.57
6.34
51.49
9.42
51.55
18.50
56.36
20.47
day 0
day 7
day 14
day 21
day 28
Storage period, day
non-vacuum packed
vacuum packed


(b) Total color difference of Saba banana at 9°C
Total color different ,ΔE
60.00
50.00
40.00
30.00
20.00
10.00
0.00
0.00
0.00
11.34
7.04
21.07
9.22
27.74
18.35
31.60
19.12
day 0
day 7
day 14
day 21
day 28
Storage period, day
non-vacuum packed
vacuum packed


(c) Total color difference of Saba banana at -20°C
Total color different ,ΔE
60.00
50.00
40.00
30.00
20.00
10.00
0.00
0.00
0.00
10.89
8.31
18.49
8.43
19.41
9.71
21.22
11.51
day 0
day 7
day 14
day 21
day 28
Storage period, day
non-vacuum packed
vacuum packed

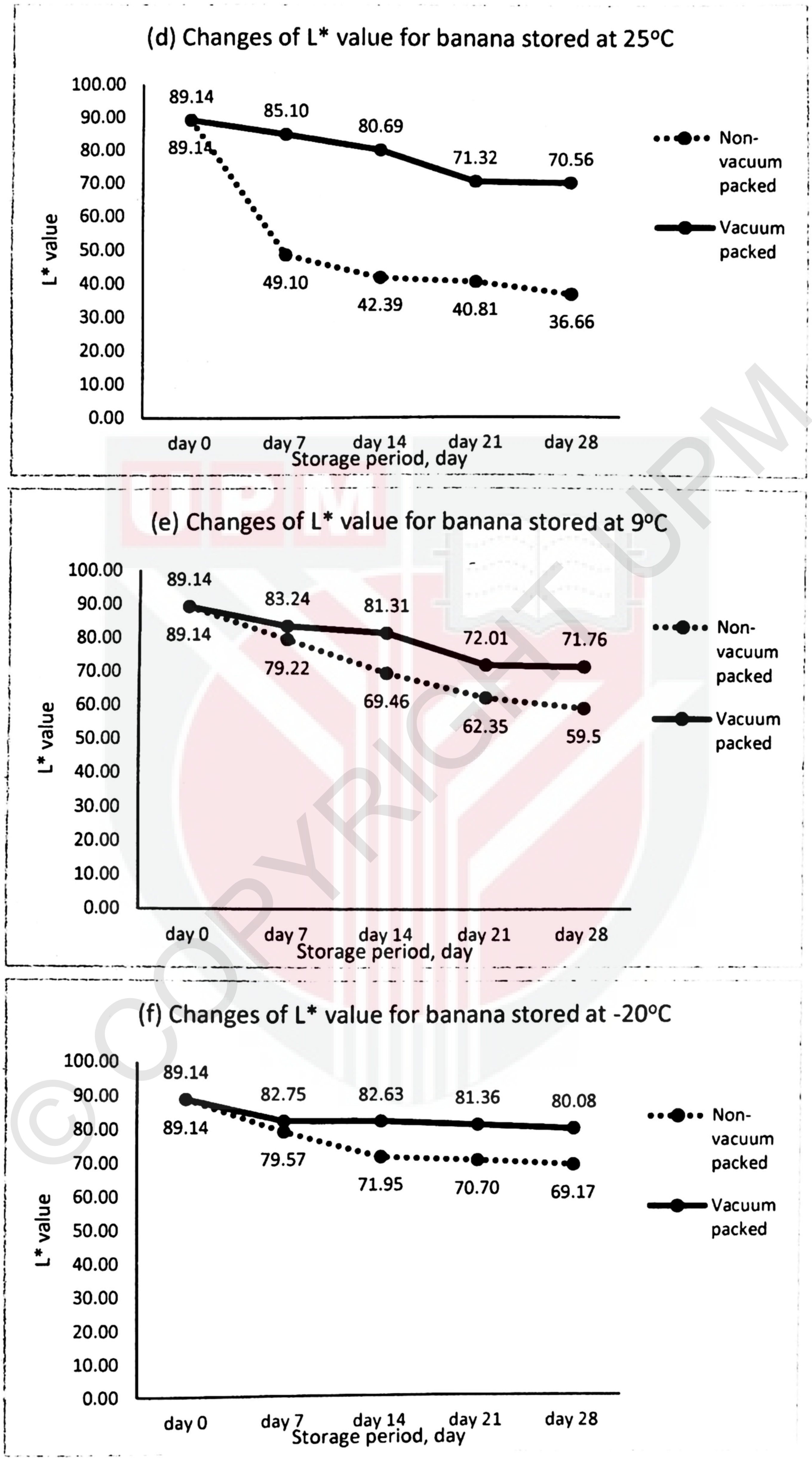



Figure 30: Total color difference of bananas kept at (a) 25°C, (b) 9°C, (c) -20°C and changes of L* value of bananas stored at (d) 25°C, (e) 9°C, (f) -20°C

For the NVP bananas stored at 25°C, ΔE (Figure 30(a)) increased significantly from 0th day to 28th day which was from 0 to 56.36 while the L* value (Figure 30(d)) was decreased significantly from 89.14 to 36.66 indicating change in color from whiter to blacker due to browning reaction. Whereas for the VP banana, ΔE (Figure 30(a)) increased slightly from 0 to 20.47 and L* value (Figure 30(d)) also decreased slightly from 89.14 to 70.56 which indicates that the banana still preserves the white color at the end of the study period but not as good as VP bananas kept at -20°C. The increment of ΔE and decrement of L* value was not as significant as NVP bananas kept at 25°C because VP delay the ripening process and limited the oxygen contact with bananas, thus the change in color was not significant.

Figure 30(b) and (e) which was for 9°C, the ΔE value of both banana samples NVP and VP were increased with time while L* value of both banana samples NVP and VP were decreased with time. The ΔE of NVP values was increased significantly from 0 to 31.60 respectively while the L* value decreased from 89.14 to 58.5 which indicates that the bananas had increased the blackish color due to browning reaction. The same goes for the ΔE (Figure 30(b)) for VP banana which was also increased from 0 to 19.12 and the L* value (Figure 30(e)) decreased from 89.14 to 71.76. It was found that the changes in ΔE and L* value for NVP and VP banana samples kept at 9°C were lower than ΔE and L* value for NVP and VP banana samples kept at 25°C due to the low storage temperature. Lower storage temperature retarded the ripening and lowered the chemical reaction including browning reaction, thus less changes in white color of bananas.

Meanwhile from Figures 30(c) and (f), which was for -20°C, the ΔE value of both banana samples NVP and VP were also observed to increase from 0th day to 28th while L* value of both banana samples NVP and VP were decreased from 0th day to

28th. From Figure 30(c) The ΔE of NVP banana was increased slightly from 0 to 21.22 and the L* value was decreased from 89.14 to 69.17 as shown in Figure 30(f) indicating that the white color had started to change into a blackish intensity and this change occurred due to the low storage temperature that caused chilling injury to the banana kept without any packaging[14] and also browning reaction. While for VP banana, ΔE was increased slightly from 0 to 11.51 as shown in Figure 30(c) and the L* value was increased from 89.14 to 80.08 as shown in Figure 30(f) which indicates that the bananas still preserve the white color due to oxygen barrier of vacuum packaging and lower temperature that had retarded the chemical reaction.

Overall, ΔE value increased from 0th day to 28th day storage period had shown that there are color changes occurred while period of storage while and L* value was decreased had shown that the white color of bananas at the beginning turned to blackish slowly. These changes might be because of enzymatic browning reaction during storage period. Enzymatic browning is one of the most limiting factors on the shelf-life of fresh-cut products [24]. Cells are broken causing enzymes to be liberated from tissues and put in contact with their substrates. Enzymatic browning is the discoloration which results from the action of a group of enzymes called polyphenol oxidases (PPO), which have been reported to occur in all plants including banana. That why PPO has been considered one of the most damaging enzymes to quality maintenance[24]. Usually, brown pigments are formed, but in addition, reddish-brown, blue-gray and even black discolorations can be produced on some bruised plant tissues [24]. Color variation in products of enzymatic oxidation is related to the phenolic compounds involved in the reaction and bananas PPO activity is higher in the pulp than in the peel [24].

The ΔE can also be an indicator to the changes in banana colors as the lower the total color difference or ΔE value, the closer the sample was to the standard (Bassey et al. 2013; Hunterlab, 2016). Even though the color changes increased with the increase in time, VP banana exhibited lower ΔE and L* value compared to the NVP banana. The lower value for vacuum packed bananas proved that the absence of oxygen and the good barrier properties of packaging material limited the oxygen and moisture content to pass through the packaging thus, reduce the ripening process reaction and browning reaction. Comparing between bananas kept at the three storage temperatures, VP bananas stored at -20°C exhibited the lowest value in ΔE and L* value compared to the other storage temperature due to the bananas were packed in vacuum packaging with good barrier properties and in addition, the frozen temperature had helped to inhibit or inactive the growth of bacteria thus the quality was maintained from the beginning to end of the study period.

### 4.2.3 Moisture Analysis

Moisture content is one of the most commonly measured properties of food materials. It is an important criteria to food for a number of different reasons and one of it is it determines food quality. The texture, taste, appearance and stability of foods are dependent on the amount of water they contain [39]. Therefore, the moisture analysis was done to relate with the firmness/texture of the banana. The percentage of moisture content for NVP and VP bananas stored at 25°C, 9°C, and -20°C over 1 month storage period is shown in Figures 31(a) – (c).

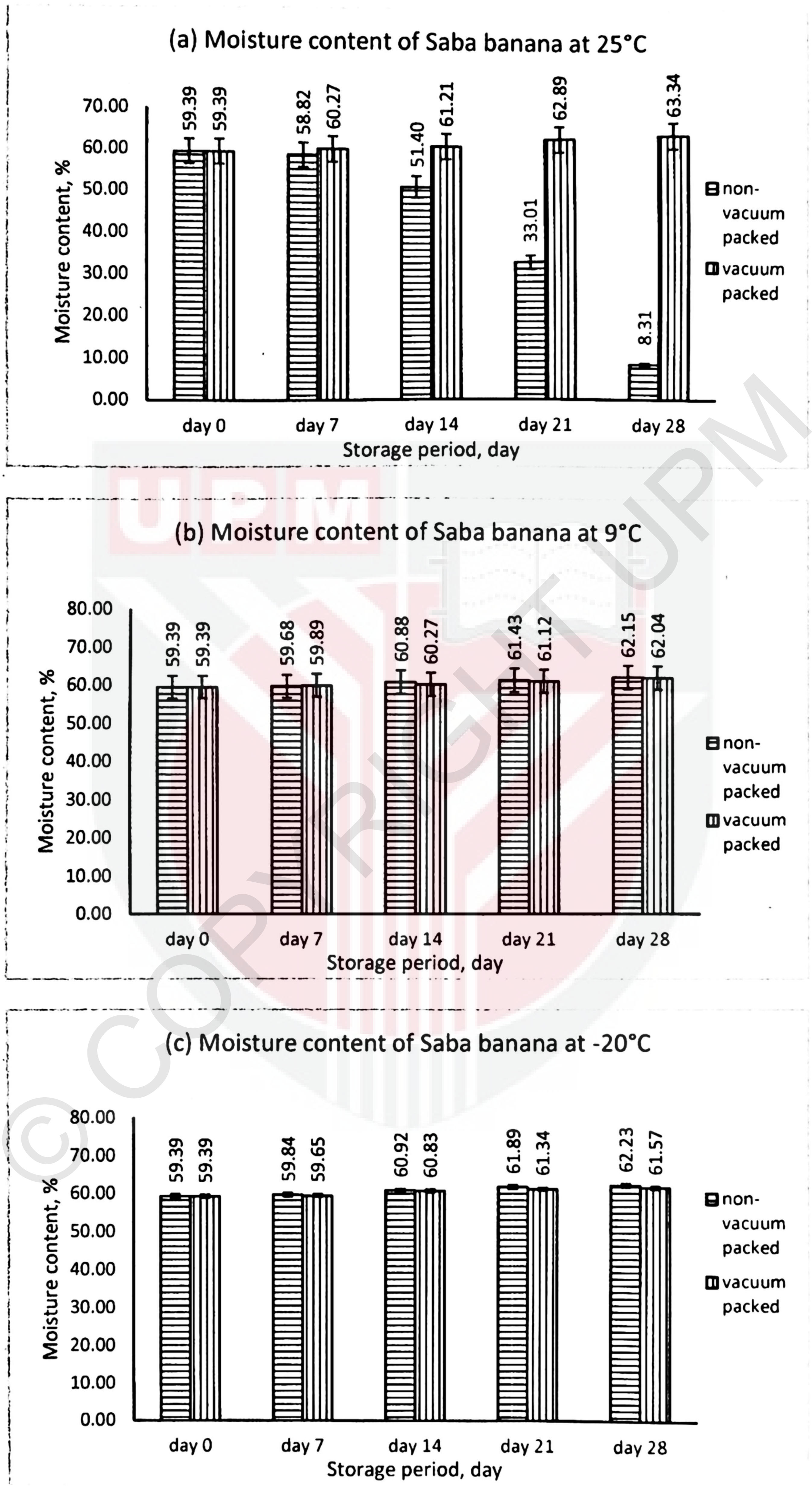



Figure 31: Moisture content of bananas stored at (a) 25°C, (b) 9°C, and (c) -20°C

It can be seen that the amount of moisture content increased slightly with the increase in storage period except for the NVP bananas stored at 25°C where the percentage of moisture content was decreased significantly across storage period. The increment was basically due to the ripening process occurred thus increase the moisture content. From Figure 31(a) which was for the bananas stored at 25°C, the percentage of moisture content for NVP banana decreased significantly from 59.39% on the $0^{th}$ day to 8.31% on the $28^{th}$ day. As shown in Figure 32 the decrement was due to the fact that the bananas after 1 month period of storage had became overripe and spoiled thus dried from time to time. Whilst for the VP banana, the moisture content was slightly increased from 59.39% to 63.34% from $0^{th}$ day to $28^{th}$ day respectively due to the moisture at surrounding being absorbed and also the ripening process occurred which resulted to change from starch into glucose [25].

Based on Figure 31(b) which was for 9°C, the moisture content shown by both samples was increased from time to time. The NVP values increased slightly from 59.39% to 62.15% while the moisture content for VP banana also increased slightly from 59.39% to 62.04%. The increment in moisture content for NVP and VP bananas was due to ripening process that occurred slowly and storage condition that have high relative humidity had caused moisture migration from surrounding to the bananas [28]. As can be seen from Figure 31(c) which was for -20°C, similar pattern showed as the bananas stored at 9°C which the moisture content for both samples was also increased from day $0^{th}$ day to $28^{th}$ day. For the NVP bananas, moisture content was increased from 59.39% to 62.23% whilst for VP banana, the moisture was increased from 59.39% to 61.57%. The increment in moisture content in bananas was also due to the similar reasons as stated before.

The increase in moisture content at the end of the storage period for all banana samples was basically due to the ripening process that occurred throughout the storage period and this occurrence was because of the breakdown of starch into sugars hence high moisture content (Karthiayani et al., 2013). Besides that, the storage conditions that have high relative humidity might also favor the moisture migration from the environment to the banana [28]. This led to increase in moisture content and also known to lead to increase of reactant mobility (Bassey et al. 2013).

Even though all conditions showed an increased pattern for moisture content except for the NVP bananas stored at 25°C, where the moisture content decreased across storage period, the VP bananas exhibited lower moisture compared to the NVP bananas. The lower moisture content of VP bananas than NVP bananas proved that the VP bananas were more firm and contain less water content compared to the NVP banana due to the absence of oxygen and good barrier properties of the vacuum packaging material that helped to retard the ripening process and thus maintained the quality of the banana from the beginning to the end. Meanwhile, at 25°C the moisture content of NVP bananas was decreased significantly and this was due to the spoilage that occurred to the NVP samples thus dried the banana as storage period increased.

Comparing between the three storage temperatures, the -20°C storage temperature for VP bananas exhibited the lowest moisture content compared to the others indicating that the VP bananas still maintaining their quality and firmness and that exhibited longer shelf life due to the slower ripening process. This was because the bananas were vacuum packed. The absence of oxygen delayed the ripening process and reduce the moisture content. Besides, the good moisture barrier properties of packaging material inhibited the movement of moisture from surrounding to pass through the package, thus, the quality was maintained.

### 4.2.4 Observation on Banana Images Over the Storage Period

Physical analysis done also include capturing and observing images of the NVP and VP bananas on day 0, 7, 14, 21 and 28th over the storage period as shown in Figures 32 to Figure 37. It was found that these figures supported all the other physical analysis results explained in Section 4.2.1 – 4.2.3 which include texture analysis, color analysis and moisture content analysis.

Figure 32: Non-vacuum packed banana stored at 25°C

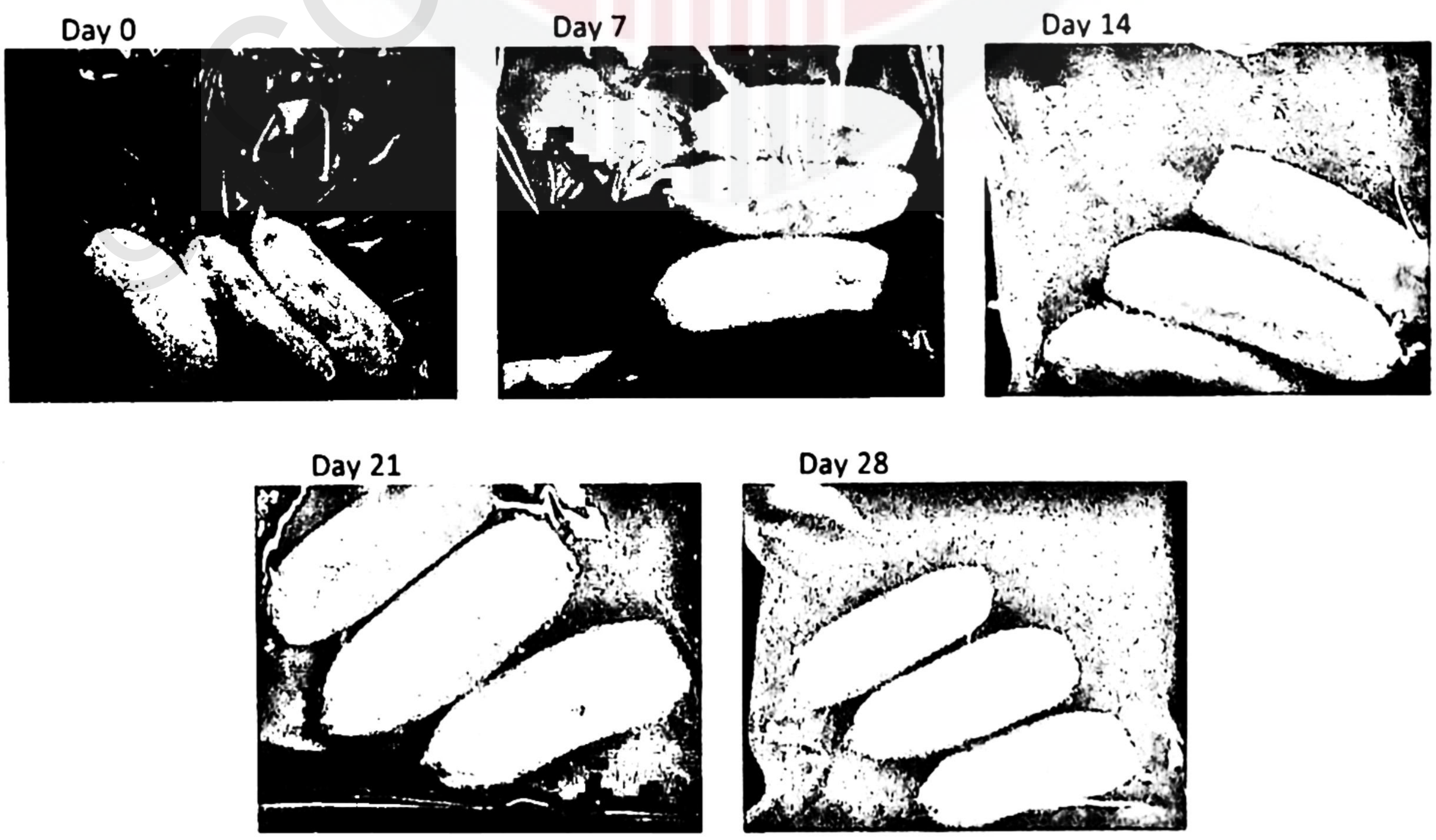


Figure 33: Vacuum packed banana stored at 25°C

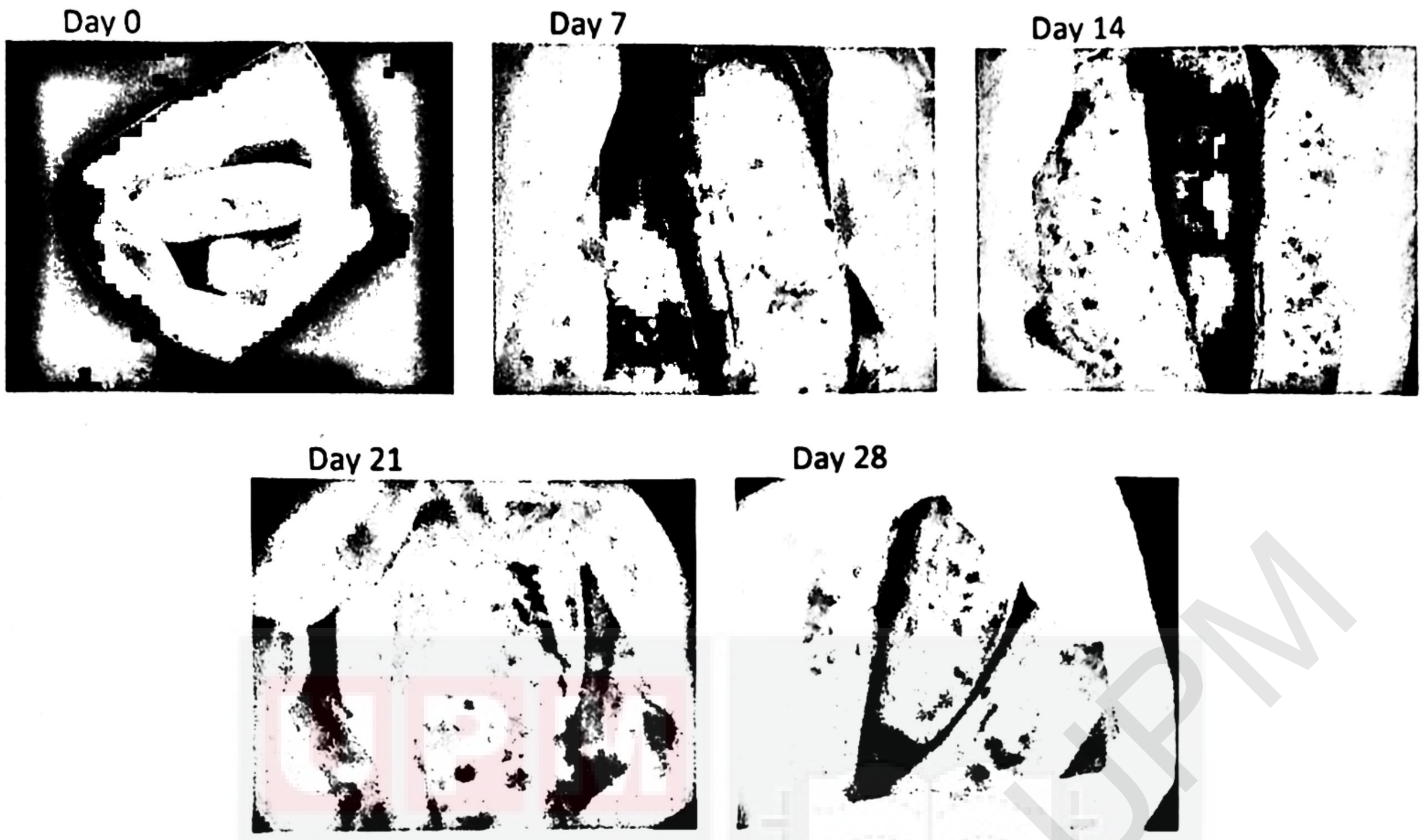



Figure 34: Non-vacuum packed banana stored at 9°C


Figure 35: Vacuum packed banana stored at 9°C

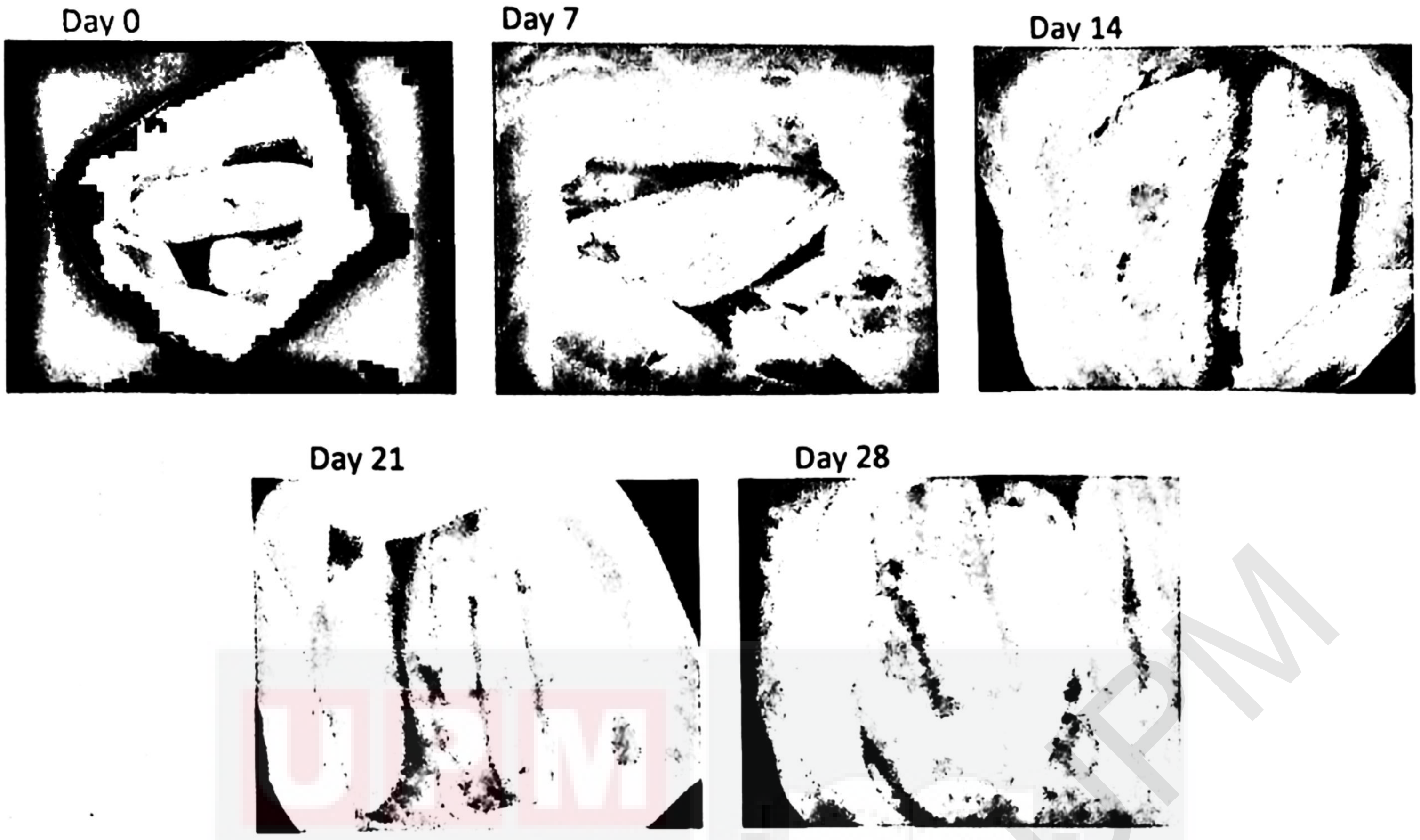



Figure 36: Non-vacuum packed banana stored at -20°C

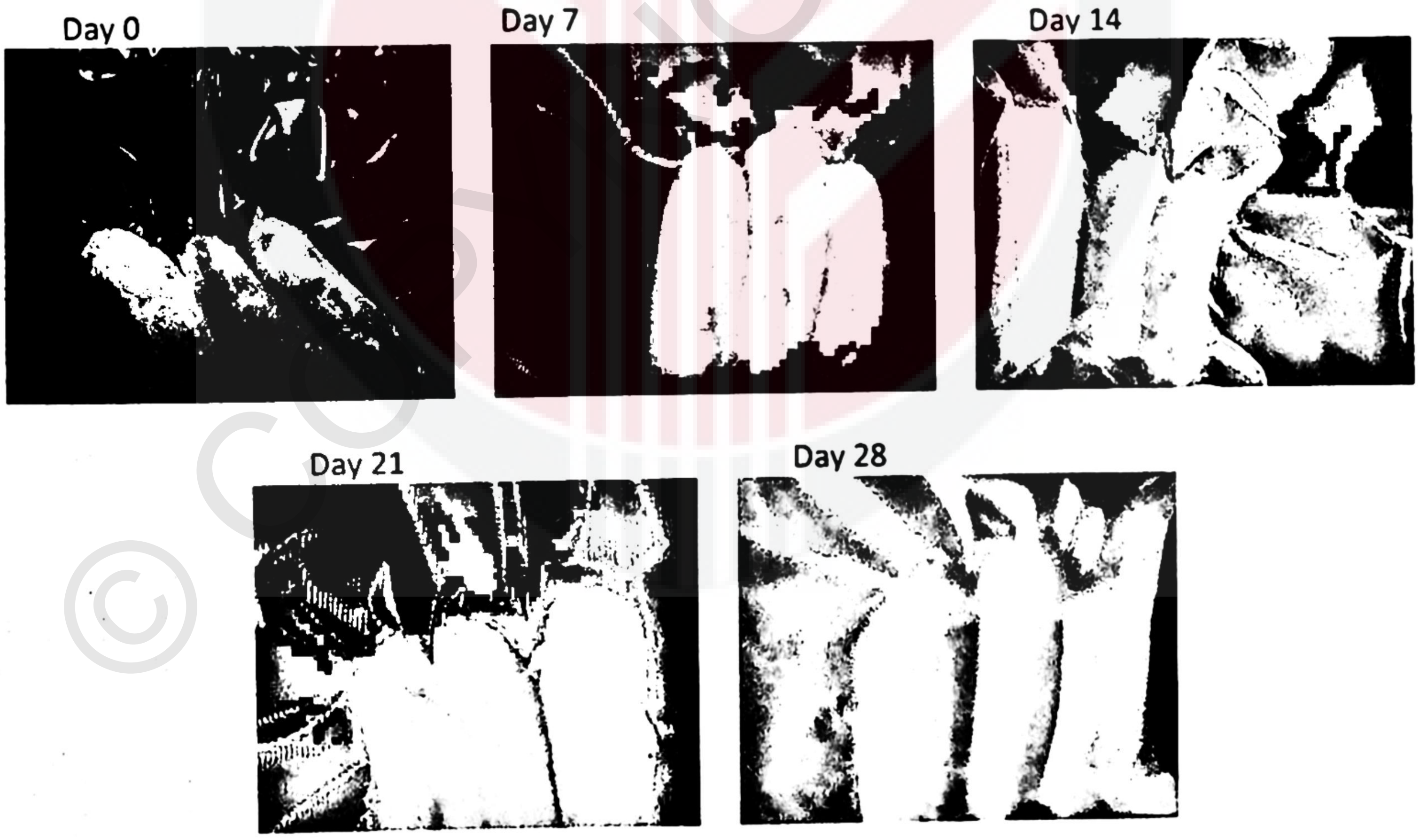



Figure 37: Vacuum packed banana stored at -20°C

## 4.3 Summary of Physical Analysis

Based on physical analysis made for both unpeeled banana and peeled banana and based on Figure 23 to Figure 28 and Figure 32 to Figure 37 the most suitable method and temperature to prolong the shelf life of the bananas was that the bananas must be in vacuum packed and stored in frozen temperature (-20°C). With this treatment, the ripening process was retarded thus prolong the lifespan of the bananas. Based on this result, the antimicrobial analysis was proceeded using this condition in order to investigate the microbial activity of the bananas over the 1 month storage period.

## 4.4 Antimicrobial Analysis

### 4.4.1 Total Plate Count Analysis (TPC)

Figure 38 presents the number of colony formed throughout the 1 month study period using the total plate count (TPC) method for NVP and VP bananas stored at -20°C. On 0$^{th}$ day, the number of colony formed for both banana samples was $2.0 \times 10^3$ CFU/ml and this value served as the reference point for all the other samples. It was found that for both samples, there was an increase pattern in TPC over the period of storage due to bacteria growth over time. Microbial count for NVP banana increased rapidly from $2.0 \times 10^3$ CFU/ml to $2.02 \times 10^4$ CFU/ml after 28 days period of storage. This increment was due to the existent of bacteria such as psychrophilic bacteria in the freezer that contaminated with the bananas, thus increase the microbial count [27]. Furthermore, cold and moist surrounding in the freezer with the availability of nutrients from banana provide good conditions for bacteria growth [27].

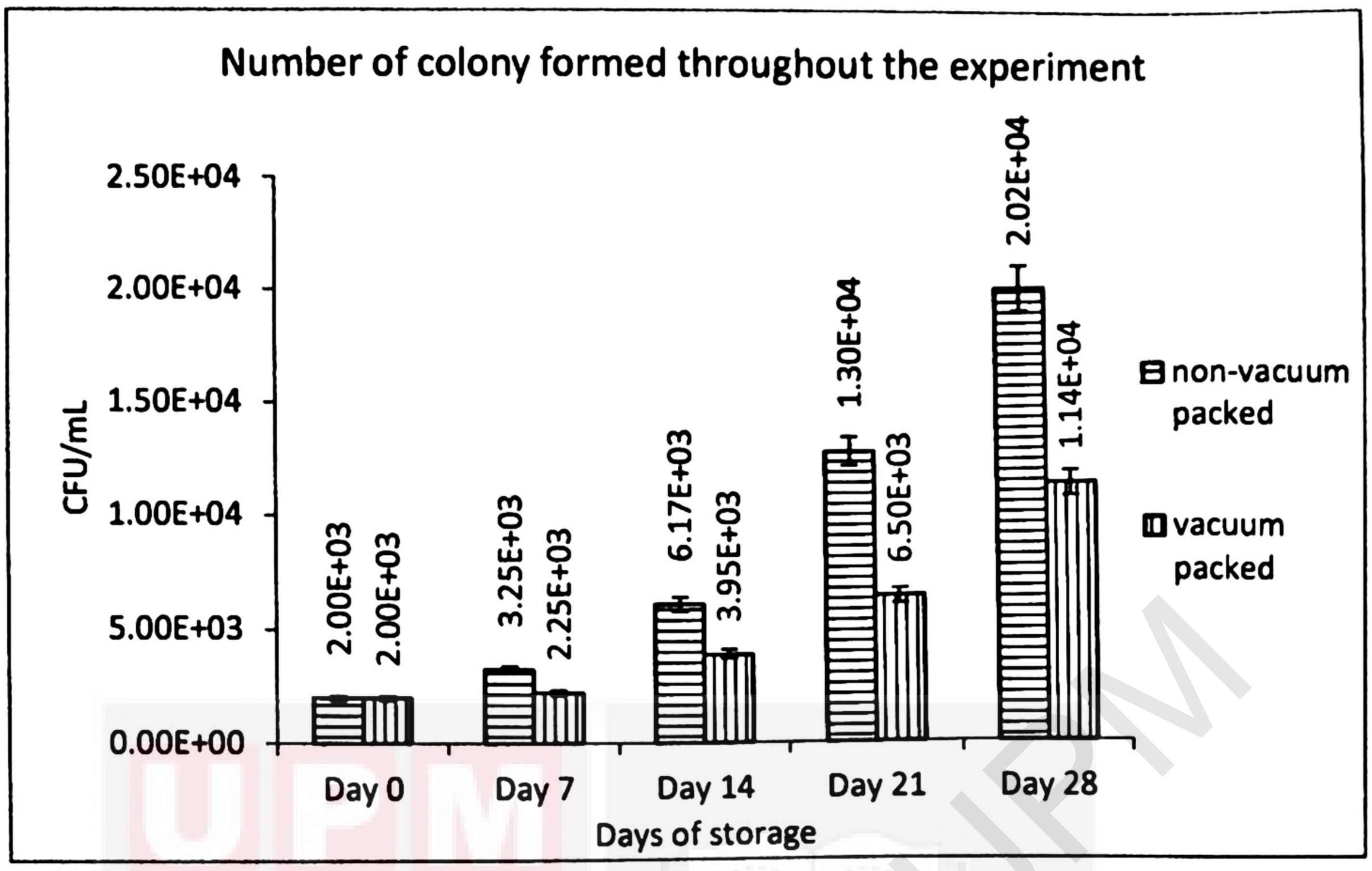



Figure 38: Number of colony formed throughout the experiment

Figure 38 also shows that the microbial growth for VP banana was lower than NVP banana whereby the value increased from 2.0 x $10^3$ CFU/ml to 1.14 x $10^4$ CFU/ml after 28th days of storage. The lower TPC was due to the absence of oxygen in the VP bananas that could promote the growth of aerobic bacteria. Besides, the role of LDPE vacuum packaging material that acted as a barrier against oxygen and moisture, thus, this barrier could prevent the growth of pathogenic bacteria also contributed to the low TPC of bacteria compared to NVP bananas. According to French regulations, an aerobic plate count (APC) which is also known as total plate count (TPC) of 5x$10^7$ colony forming units (CFU)/ml is the maximum acceptable value at the end of the microbiological shelf-life of numerous for fruits and vegetables [35]. Therefore, in this work, since the TPC was still below the maximum acceptable value of 5x$10^7$ CFU/ml, the bananas were still consumable and acceptable after 1 month period of storage. However, bananas stored in vacuum packed exhibited better TPC than non-vacuum packed. This proved that vacuum packaging method is very promising in maintaining the quality and extending the shelf life of bananas.

Figures 39 and 40 shows the images of colonies formed after incubated at 37°C for 18 hours on 0, 7, 14, 21 and 28th day of storage respectively. Both figures supported the previous findings whereby the number of colonies increased with increasing storage period and that number of colonies on VP bananas were lower than NVP bananas.

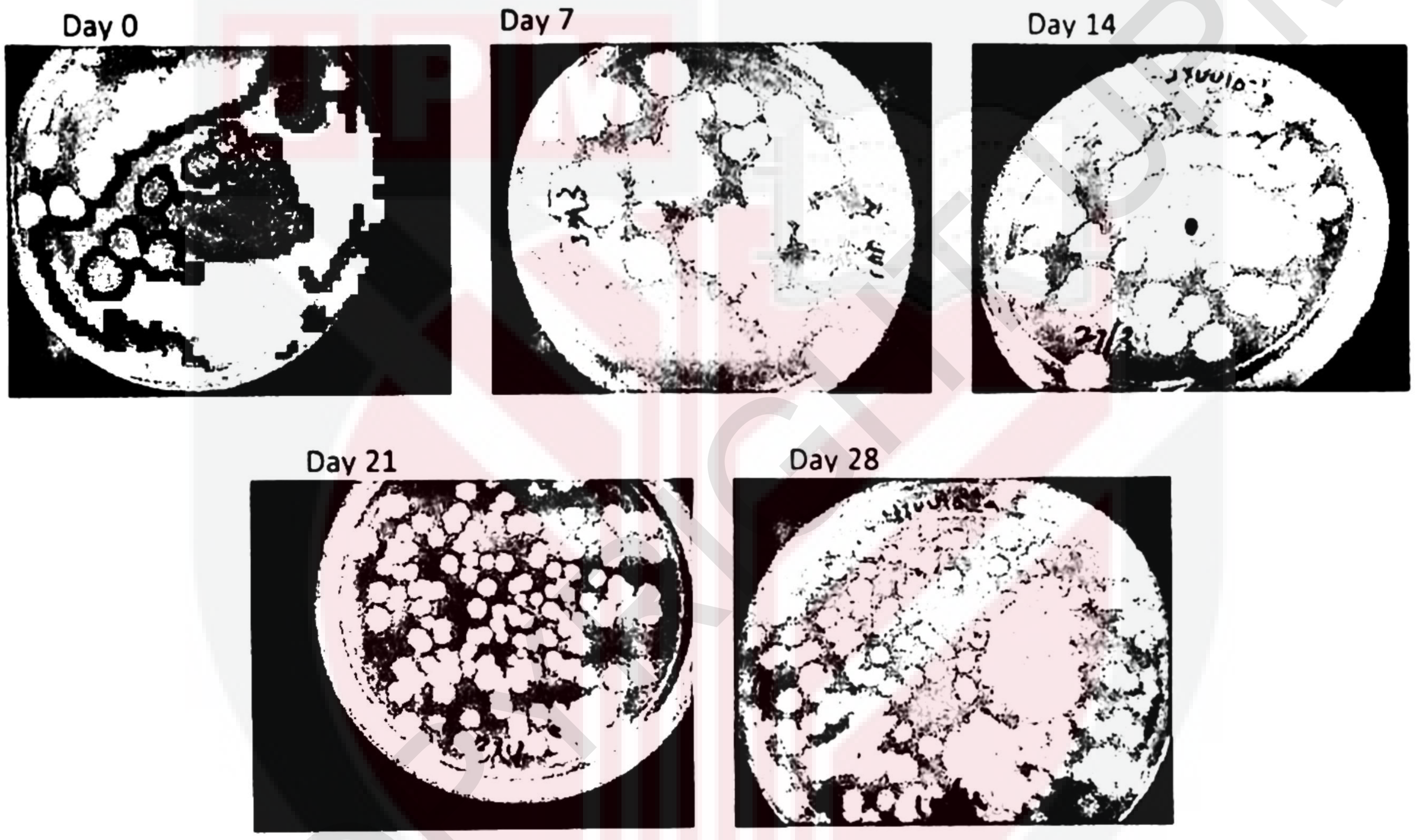



Figure 39: Colonies formed at vacuum packed banana stored at -20°C

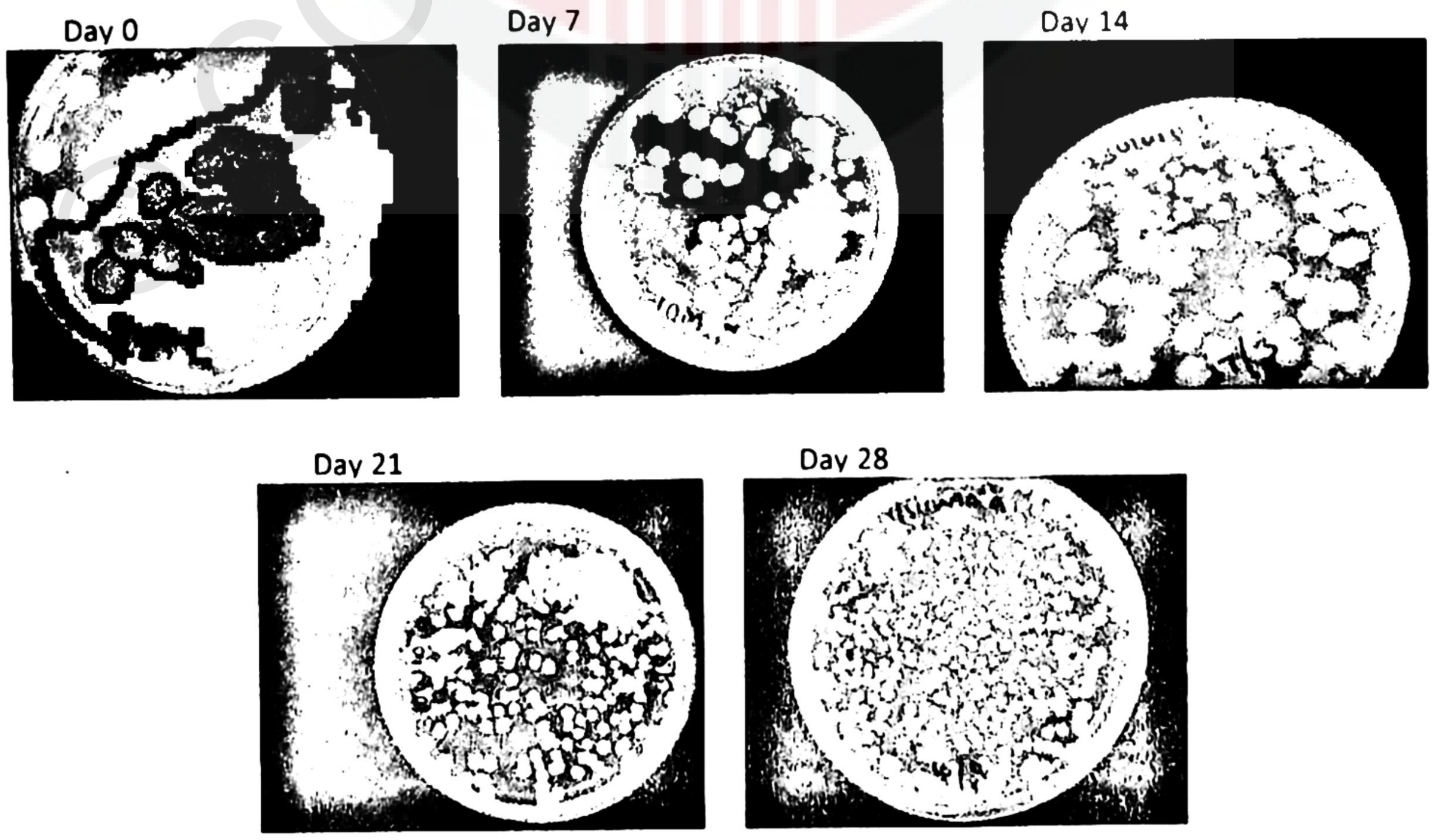



Figure 40: Colonies formed at non-vacuum packed banana stored at -20°C

# CHAPTER 5

# CONCLUSION AND RECOMENDATION

## 5.1 Conclusion

Based on the analysis conducted, vacuum packaging and storage temperature was found to really impacted shelf life of Saba banana. Vacuum packaging removed out all the air inside the package especially oxygen while Low Density Polyethylene (LDPE) packaging material provided excellent barrier against moisture and oxygen thus retarded the ripening process of banana [40]. Low storage temperature especially -20 $^{0}$C contributed towards prolonging the shelf life of banana as at low temperature, the growth of bacteria was inhibited and became inactive. In this work vacuum packaging and low storage temperature were found to be promising to increase the shelf life of Saba banana. This could prevent the spoilage and wastage of abundance Saba banana in Sabah thus increase economic growth.

## 5.2 Recommendations

Besides vacuum packaging, there are many methods that can be utilized to retard the ripening process of banana in order ot increase the shelf life of banana. It is recommended for future study to explore different methods such as by applying the pre-treatment of bananas with ascorbic acid, citric acid, and/or sodium metabisulphite and also a pre treatment prior to vacuum packed such as washing the bananas with fungicide solution. Besides that, the effect of addition of oxygen absorber and ethylene absorber sached before packed in vacuum packaging can also be investigated. Other method can be the combination effect of chemical dip and/or edible coating and/or controlled atmosphere (CA) on quality of banana. The banana can be dip into a solution containing calcium chloride, ascorbic acid cysteine, and/or combined with a carrageenan coating and/or combined with controlled atmosphere (3% O2 + 10% CO2). Besides variying the method to retard the ripening process, shelf life of bananas can also be investigated by determining different properties such as chemical properties of banana (pH, total soluble solid (TSS)), sensory analysis on the banana and barrier properties of packaging material (oxygen transmission rate (OTR), water vapor transmission rate of packaging material (WVTR)). The improvement on the banana analysis can also be done to get more firm result such as repeating the experiment, done for triplicate sample in one batch, and also thawing the frozen banana before characterized/testing.

## REFERENCES


1. Sen C. Modified atmosphere packaging and active packaging of banana (Musa spp.): A review on control of ripening and extension of shelf life. *J Stored Prod Postharvest Res*. 2012;3(9):122-132. doi:10.5897/JSPPR11.057.
2. Food and Agriculture Organization of the United Nations. EST: Banana facts and figures. 2016:2-4. http://www.fao.org/economic/est/est-commodities/bananas/bananafacts/en/#.WjG_YEpl_IV.
3. Madan A, Jain RK, Nandane AS. Development of Active Modified Atmosphere Lab Scale Setup to Study the Effect on Shelf-life of Banana. 2014;3(1):1-10.
4. Kudachikar VB, Kulkarni SG, Prakash MNK. Effect of modified atmosphere packaging on quality and shelf life of "Robusta" banana (Musa sp.) stored at low temperature. *J Food Sci Technol*. 2011;48(3):319-324. doi:10.1007/s13197-011-0238-y.
5. Department of Agriculture (DOA). Fruit Crops Statistic. *Dep Agric Malaysia*. 2016:1-180. http://www.doa.gov.my/index/resources/aktiviti_sumber/sumber_awam/maklumat_pertanian/perangkaan_tanaman/perangkaan_buah_2016.pdf.
6. Ramos B, Miller FA, Brandão TRS, Teixeira P, Silva CLM. Fresh fruits and vegetables - An overview on applied methodologies to improve its quality and safety. *Innov Food Sci Emerg Technol*. 2013;20:1-15. doi:10.1016/j.ifset.2013.07.002.
7. Castillo-Israel KAT, Baguio SF, Diasanta MDB, Lizardo RCM, Dizon EI, Mejico MIF. Extraction and characterization of pectin from Saba banana [Musa "saba"(Musa acuminata x Musa balbisiana)] peel wastes: A preliminary study. *Int Food Res J*. 2015;22(1):202-207.
8. Barrett DM, Beaulieu JC, Shewfelt R. Color, flavor, texture, and nutritional quality of fresh-cut fruits and vegetables: Desirable levels, instrumental and sensory measurement, and the effects of processing. *Crit Rev Food Sci Nutr*. 2010;50(5):369-389. doi:10.1080/10408391003626322.
9. Encyclopaedia Britannica. Banana , fruit of the genus. 2017:1-3. https://www.britannica.com/plant/banana-plant.
10. Stoffelen R, Verlinden R, Pinochet J, Swennen RL, De Waele D. Host plant response of Fusarium wilt resistant Musa genotypes to Radopholus similis and Pratylenchus coffeae. *Int J Pest Manag*. 2000;46(4):289-293. doi:10.1080/09670870050206064.
11. Rabbani M.G. MAAAHHMK. Effects of Variety and Postharvest Treatments on Shelf Life and Quality of Tomato Effects of Variety and Postharvest Treatments on Shelf Life and Quality of Tomato. 2011;6(May):163-175.
12. Siriwardana H, Abeywickrama K, Kannangara S, Jayawardena B. Efficacy of alum treatment and vacuum packaging in controlling crown rot disease of Cavendish banana. 2016;5(August):7-23.
13. Siriwardana H, Abeywickrama K, Kannangara S, Jayawardena B. Control of Postharvest Crown Rot Disease in Cavendish Banana with Aluminium Sulfate and Vacuum Packaging. 2017;12(3):162-171.
14. Hailu M, Seyoum Workneh T, Belew D. Effect of packaging materials on shelf life and quality of banana cultivars (Musa spp.). *J Food Sci Technol*. 2012;51(11):2947-2963. doi:10.1007/s13197-012-0826-5.
15. Mohapatra D, Mishra S, Singh CB, Jayas DS. Post-harvest Processing of Banana: Opportunities and Challenges. *Food Bioprocess Technol*.

2011;4(3):327-339. doi:10.1007/s11947-010-0377-6.
16. Food Safety Authority of Ireland (FSAI). *Validation of Product Shelf-Life (Revision 3).*; 2017.
17. Nelson S. Banana ripening : principles and practice.
18. Ma L, Zhang M, Bhandari B, Gao Z. Recent developments in novel shelf life extension technologies of fresh-cut fruits and vegetables. *Trends Food Sci Technol.* 2017;64:23-38. doi:10.1016/j.tifs.2017.03.005.
19. Meireles A, Giaouris E, Simões M. Alternative disinfection methods to chlorine for use in the fresh-cut industry. *Food Res Int.* 2016;82:71-85. doi:10.1016/j.foodres.2016.01.021.
20. Bico SLS, Raposo MFJ, Morais RMSC, Morais AMMB. Combined effects of chemical dip and/or carrageenan coating and/or controlled atmosphere on quality of fresh-cut banana. *Food Control.* 2009;20(5):508-514. doi:10.1016/j.foodcont.2008.07.017.
21. Tapre A, Jain R. Study of advanced maturity stages of banana. *Int J Adv Eng Res Stud.* 2012;I(III):272-274. http://www.technicaljournalsonline.com/ijaers/VOL I/IJAERS VOL I ISSUE III APRIL JUNE 2012/178.pdf.
22. Cultivars P. Banana Ripening. 2015;(c):6-9.
23. Mohapatra D, Mishra S, Sutar N. Banana post harvest practices: current status and future prospects - a review. *Agric Rev.* 2010;31(1):56-62. http://www.indianjournals.com/ijor.aspx?target=ijor:ar&volume=31&issue=1&article=007.
24. Garcia E, Barrett DM. Preservative treatments for fresh-cut fruits and vegetables. *CRC Press.* 2002. doi:10.1201/9781420031874.ch9.
25. Karthiayani A, Varadharaju N, Siddharth M. Textural Properties Of Banana (Musa Acuminata) Stored Under Modified Atmosphere Conditions Using Diffusion Channel. *J Agric Eng Biotechnol.* 2013;1(May):17-22. doi:10.18005/JAEB0101003.
26. *Modern Food Microbiology.*; 2005. doi:10.1007/b100840.
27. Troller J. Chapter 3: Factors that Influence Microbial Growth. 2001. http://msue.anr.msu.edu/uploads/234/48511/Safe_Practices_for_Food_Processes_Chpt._3_Factors_that_Influence_Microbial_Growth.pdf.
28. Bassey FI, Chinnan MS, Ebenso EE, Edem CA, Iwegbue CMA. Colour change: An indicator of the extent of maillard browning reaction in food system. *Asian J Chem.* 2013;25(16):9325-9328. doi:10.14233/ajchem.2013.15504.
29. Mk M, MB C, CM N, Naik MC. Vacuum Packaging Technology : a Novel Approach for Extending the Storability and Quality of Agricultural Produce. In: Vol 7. ; 2017:1-6. doi:10.15406/apar.2017.07.00242.
30. Siracusa V. Food packaging permeability behaviour: A report. *Int J Polym Sci.* 2012;2012(May 2012). doi:10.1155/2012/302029.
31. Shin J, Selke SEM. Food Packaging. *Food Process Princ Appl.* 2014:249-273.
32. Soltani M, Alimardani R, Omid M. Changes in physico-mechanical properties of banana fruit during ripening treatment. *J Am Sci.* 2011;7(5):5-10. http://www.americanscience.org.
33. Hunterlab. Brief Explanation of delta E or delta E*. https://support.hunterlab.com/hc/en-us/articles/203023559-Brief-Explanation-of-delta-E-or-delta-E-. Published 2016.
34. Sau S, Chandra B, Viswavidyalaya K, et al. Effect of Modified and Active Packaging on Shelf Life and Quality of Banana Cv . Grand Naine Aging on

Shelf Life Active PA Quality. 2017;(May).
35. Fan L, Song J. Microbial quality assessment methods for fresh-cut fruits and vegetables. *Stewart Postharvest Rev.* 2008;4(3). doi:10.2212/spr.2008.3.10.
36. United Fresh Produce Association U. Microbiological Testing of Fresh Produce A White Paper on Considerations in Developing and Using Microbiological Sampling and Testing Procedures if Used as Part of a Food Safety Program for Fresh Fruit and Vegetable Products. 2010:1-18.
37. virtual Amrita Laboratories Universalizing Education V amrita. ed. *Bacterial Growth Curve Objectives : Principle* :; 2014.
38. Chauhan OP, Raju PS, Dasgupta DK, Bawa AS. Modified Atmosphere Packaging of Banana (cv. Pachbale) with Ethylene, Carbon di-oxide and Moisture Scrubbers and Effect on its Ripening Behaviour. *Am J Food Technol.* 2006;1(2):179-189.
39. McClemens J. Determination of Moisture and Total Solids. *Course Anal Food Prod UMASS.* 2003:3-7. http://people.umass.edu/~mcclemen/581Moisture.html.
40. Julianti E, Yusraini E. Effect of Modified Atmosphere Packaging on Postharvest Quality of Barangan Banana. 2011;(October):544-547.
41. Cindy. "How Long Do Bananas Last? Shelf Life, Storage, Expiration." Eat By Date, 2012, www.eatbydate.com/fruits/fresh/bananas-shelf-life-expiration-date/.
42. "StillTasty." How Long Do Bananas Last?, www.stilltasty.com/Fooditems/index/16451.

## APPENDICES

### 6.1 Result of Analysis

Table 4: Texture analysis of the banana in different storage temperature

| Sample | Peeled (vacuum packed) | | |
|---|---|---|---|
| Days | Penetration force, g | | |
| | Room | Cold | Freeze |
| Day 0 | 699.93 | 699.93 | 699.93 |
| Day 7 | 686.51 | 833.52 | 7933.00 |
| Day 14 | 684.79 | 820.15 | 6892.72 |
| Day 21 | 580.04 | 791.95 | 2706.32 |
| Day 28 | 562.78 | 789.18 | 2667.34 |
| | | | |
| Sample | Peeled (non-vacuum packed) | | |
| Days | Penetration force, g | | |
| | Room | Cold | Freeze |
| Day 0 | 699.93 | 699.93 | 699.93 |
| Day 7 | 881.17 | 831.19 | 7568.03 |
| Day 14 | 1704.68 | 767.24 | 6058.31 |
| Day 21 | 2165.56 | 701.36 | 2468.64 |
| Day 28 | 3547.17 | 656.28 | 2433.83 |
| | | | |
| Sample | Unpeeled (Vacuum packed) | | |
| Days | Penetration force, g | | |
| | Room | Cold | Freeze |
| Day 0 | 1312.87 | 1312.87 | 1312.87 |
| Day 7 | 1295.98 | 1283.26 | 9277.27 |
| Day 14 | 1273.24 | 1261.04 | 9228.23 |
| Day 21 | 1239.58 | 1239.10 | 9167.85 |
| Day 28 | 1228.03 | 1219.91 | 9151.51 |
| | | | |
| Sample | Unpeeled (Non-vacuum packed) | | |
| Days | Penetration force, g | | |
| | Room | Cold | Freeze |
| Day 0 | 1312.87 | 1312.87 | 1312.87 |
| Day 7 | 615.38 | 1302.00 | 4384.00 |
| Day 14 | 263.13 | 1288.80 | 4206.41 |
| Day 21 | 222.33 | 1278.54 | 4185.06 |
| Day 28 | 220.50 | 1254.62 | 4175.06 |

Table 5: Color analysis of banana stored in different storage temperature

| Sample | Peeled (vacuum packed) | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Condition | Room | | | | Cold | | | | Freeze | | | |
| Color / Days | L | A | b | ΔE | L | a | b | ΔE | L | a | b | ΔE |
| Day 0 | 89.14 | 0.87 | 22.97 | 0.00 | 89.14 | 0.87 | 22.97 | 0.00 | 89.14 | 0.87 | 22.97 | 0.00 |
| Day 7 | 85.10 | 2.87 | 18.51 | 6.34 | 83.24 | 4.17 | 21.02 | 7.04 | 82.75 | 5.39 | 25.77 | 8.31 |
| Day 14 | 80.69 | 4.01 | 20.24 | 9.42 | 81.31 | 5.43 | 21.24 | 9.22 | 82.63 | 5.44 | 25.76 | 8.43 |
| Day 21 | 71.32 | 3.93 | 19.07 | 18.50 | 72.01 | 6.24 | 19.16 | 18.35 | 81.36 | 6.04 | 25.62 | 9.71 |
| Day 28 | 70.56 | 4.16 | 15.03 | 20.47 | 71.76 | 6.84 | 17.69 | 19.12 | 80.08 | 6.51 | 27.28 | 11.51 |
| | | | | | | | | | | | | |
| Sample | Peeled (non-vacuum packed) | | | | | | | | | | | |
| Condition | Room | | | | Cold | | | | Freeze | | | |
| Color / Days | L | A | b | ΔE | L | a | b | ΔE | L | a | b | ΔE |
| Day 0 | 89.14 | 0.87 | 22.97 | 0.00 | 89.14 | 0.87 | 22.97 | 0.00 | 89.14 | 0.87 | 22.97 | 0.00 |
| Day 7 | 49.10 | 5.05 | 9.12 | 42.57 | 79.22 | 5.25 | 19.64 | 11.34 | 79.57 | 4.37 | 19.13 | 10.89 |
| Day 14 | 42.39 | 0.39 | 1.39 | 51.49 | 69.46 | 5.81 | 17.29 | 21.07 | 71.95 | 5.36 | 17.85 | 18.49 |
| Day 21 | 40.81 | 1.90 | 5.07 | 51.55 | 62.35 | 5.98 | 17.91 | 27.74 | 70.70 | 5.57 | 19.17 | 19.41 |
| Day 28 | 36.66 | 0.25 | 2.42 | 56.36 | 59.5 | 6.18 | 13.38 | 31.60 | 69.17 | 6.60 | 18.64 | 21.22 |

| Sample | Unpeeled (vacuum packed) | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Condition | Room | | | | Cold | | | | Freeze | | | |
| Color / Days | L | A | b | ΔE | L | a | b | ΔE | L | a | b | ΔE |
| Day 0 | 43.58 | -7.00 | 19.75 | 0.00 | 43.58 | -7.00 | 19.75 | 0.00 | 43.58 | -7.00 | 19.75 | 0.00 |
| Day 7 | 45.63 | -5.60 | 17.22 | 3.54 | 49.17 | -6.36 | 14.81 | 7.49 | 43.67 | -4.75 | 18 | 2.85 |
| Day 14 | 49.55 | -5.58 | 15.97 | 7.21 | 47.15 | -5.40 | 11.29 | 9.32 | 43.88 | -4.29 | 17.80 | 3.35 |
| Day 21 | 49.26 | -5.40 | 15.19 | 7.46 | 46.94 | -3.73 | 10.73 | 10.17 | 43.21 | -4.26 | 17.20 | 3.76 |
| Day 28 | 51.81 | -2.20 | 19.52 | 9.53 | 47.64 | -2.85 | 10.47 | 10.95 | 43.71 | -4.22 | 17 | 3.91 |

| Sample | Unpeeled unwrapped | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Condition | Room | | | | Cold | | | | Freeze | | | |
| Color / Days | L | A | b | ΔE | L | a | b | ΔE | L | a | b | ΔE |
| Day 0 | 43.58 | -7.00 | 19.75 | 0.00 | 43.58 | -7.00 | 19.75 | 0.00 | 43.58 | -7.00 | 19.75 | 0.00 |
| Day 7 | 61.43 | 10.93 | 14.65 | 25.81 | 43.20 | -3.80 | 18.31 | 3.53 | 47.9 | 0.54 | 7.51 | 15.01 |
| Day 14 | 57.08 | 12.17 | 13.57 | 24.25 | 43.11 | -3.64 | 16.10 | 4.98 | 46.61 | 1.06 | 6.56 | 15.75 |
| Day 21 | 51.20 | 14.39 | 13.33 | 23.60 | 42.61 | -3.47 | 15.35 | 5.72 | 50.60 | 1.92 | 5.75 | 18.02 |
| Day 28 | 42.01 | 15.22 | 13.15 | 23.23 | 42.5 | -1.29 | 14.91 | 7.56 | 45.75 | 2.76 | 5.46 | 17.44 |

Table 6: Moisture analysis of banana stored at 25°C

| Sample | Peeled (vacuum packed) | | | | | | |
|---|---|---|---|---|---|---|---|
| **Items** | **Weight of container (g)** | **Weight of container + sample (g)** | **Weight of wet sample (g)** | **Weight after drying (g)** | **Weight of dry sample(g)** | **Moisture (%)** | **Dry matter (%)** |
| **Day 0** | 0.30 | 31.25 | 30.95 | 12.87 | 12.57 | 59.39 | 40.61 |
| **Day 7** | 0.50 | 30.88 | 30.38 | 12.57 | 12.07 | 60.27 | 39.73 |
| **Day 14** | 0.27 | 30.77 | 30.50 | 12.10 | 11.83 | 61.21 | 38.79 |
| **Day 21** | 0.30 | 30.67 | 30.37 | 11.57 | 11.27 | 62.89 | 37.11 |
| **Day 28** | 0.50 | 31.13 | 30.63 | 11.73 | 11.23 | 63.34 | 36.66 |
| | | | | | | | |
| **Sample** | **Peeled (non-vacuum packed)** | | | | | | |
| **Items** | **Weight of container (g)** | **Weight of container + sample (g)** | **Weight of wet sample (g)** | **Weight after drying (g)** | **Weight of dry sample(g)** | **Moisture (%)** | **Dry matter (%)** |
| **Day 0** | 0.30 | 31.25 | 30.95 | 12.87 | 12.57 | 59.39 | 40.61 |
| **Day 7** | 0.30 | 30.63 | 30.33 | 12.79 | 12.49 | 58.82 | 41.18 |
| **Day 14** | 0.50 | 30.13 | 29.63 | 14.90 | 14.40 | 51.40 | 48.60 |
| **Day 21** | 0.40 | 30.97 | 30.57 | 20.88 | 20.48 | 33.01 | 66.99 |
| **Day 28** | 0.50 | 31.08 | 30.58 | 28.54 | 28.04 | 8.31 | 91.69 |

| Sample | Unpeeled (vacuum packed) | | | | | | |
|---|---|---|---|---|---|---|---|
| Items | Weight of container (g) | Weight of container + sample (g) | Weight of wet sample (g) | Weight after drying (g) | Weight of dry sample(g) | Moisture (%) | Dry matter (%) |
| Day 0 | 0.30 | 30.65 | 30.35 | 13.02 | 12.72 | 58.09 | 41.91 |
| Day 7 | 0.30 | 30.57 | 30.27 | 11.93 | 11.63 | 61.58 | 38.42 |
| Day 14 | 0.20 | 30.70 | 30.50 | 9.17 | 8.97 | 70.59 | 29.41 |
| Day 21 | 0.40 | 31.30 | 30.90 | 8.77 | 8.37 | 72.91 | 27.09 |
| Day 28 | 0.50 | 31.20 | 30.70 | 8.60 | 8.10 | 73.62 | 26.38 |
| | | | | | | | |
| Sample | Unpeeled (non-vacuum packed) | | | | | | |
| Items | Weight of container (g) | Weight of container + sample (g) | Weight of wet sample (g) | Weight after drying (g) | Weight of dry sample(g) | Moisture (%) | Dry matter (%) |
| Day 0 | 0.30 | 30.65 | 30.35 | 13.02 | 12.72 | 58.09 | 41.91 |
| Day 7 | 0.30 | 30.63 | 30.33 | 8.77 | 8.47 | 72.07 | 27.93 |
| Day 14 | 0.27 | 30.83 | 30.56 | 8.00 | 7.73 | 74.71 | 25.29 |
| Day 21 | 0.40 | 31.17 | 30.77 | 10.00 | 9.60 | 68.80 | 31.20 |
| Day 28 | 0.36 | 31.20 | 30.84 | 10.55 | 10.19 | 66.96 | 33.04 |

Table 7: Moisture analysis of banana stored at 9°C

| Sample | Peeled (vacuum packed) | | | | | | |
|---|---|---|---|---|---|---|---|
| **Items** | **Weight of container (g)** | **Weight of container + sample (g)** | **Weight of wet sample (g)** | **Weight after drying (g)** | **Weight of dry sample(g)** | **Moisture(%)** | **Dry matter(%)** |
| **Day 0** | 0.30 | 31.25 | 30.95 | 12.87 | 12.57 | 59.39 | 40.61 |
| **Day 7** | 0.30 | 30.47 | 30.17 | 12.40 | 12.10 | 59.89 | 40.11 |
| **Day 14** | 0.37 | 31.33 | 30.96 | 12.67 | 12.30 | 60.27 | 39.73 |
| **Day 21** | 0.40 | 31.03 | 30.63 | 12.31 | 11.91 | 61.12 | 38.88 |
| **Day 28** | 0.50 | 31.27 | 30.77 | 12.18 | 11.68 | 62.04 | 37.96 |
| | | | | | | | |
| **sample** | **Peeled (non-vacuum packed)** | | | | | | |
| **Items** | **Weight of container (g)** | **Weight of container + sample (g)** | **Weight of wet sample (g)** | **Weight after drying (g)** | **Weight of dry sample(g)** | **Moisture(%)** | **Dry matter(%)** |
| **Day 0** | 0.30 | 31.25 | 30.95 | 12.87 | 12.57 | 59.39 | 40.61 |
| **Day 7** | 0.30 | 30.73 | 30.43 | 12.57 | 12.27 | 59.68 | 40.32 |
| **Day 14** | 0.50 | 31.05 | 30.55 | 12.45 | 11.95 | 60.88 | 39.12 |
| **Day 21** | 0.40 | 30.50 | 30.10 | 12.01 | 11.61 | 61.43 | 38.57 |
| **Day 28** | 0.47 | 30.80 | 30.33 | 11.95 | 11.48 | 62.15 | 37.85 |

| sample | Unpeeled (vacuum packed) | | | | | | |
|---|---|---|---|---|---|---|---|
| **Items** | **Weight of container (g)** | **Weight of container + sample (g)** | **Weight of wet sample (g)** | **Weight after drying (g)** | **Weight of dry sample(g)** | **Moisture(%)** | **Dry matter(%)** |
| **Day 0** | 0.30 | 30.65 | 30.35 | 13.02 | 12.72 | 58.09 | 41.91 |
| **Day 7** | 0.30 | 30.73 | 30.43 | 10.36 | 10.06 | 66.94 | 33.06 |
| **Day 14** | 0.20 | 31.00 | 30.80 | 9.50 | 9.90 | 67.86 | 32.14 |
| **Day 21** | 0.40 | 31.07 | 30.67 | 9.10 | 9.00 | 70.66 | 29.34 |
| **Day 28** | 0.50 | 31.13 | 30.63 | 6.00 | 8.19 | 73.26 | 26.74 |

| sample | Unpeeled (non-vacuum packed) | | | | | | |
|---|---|---|---|---|---|---|---|
| **Items** | **Weight of container (g)** | **Weight of container + sample (g)** | **Weight of wet sample (g)** | **Weight after drying (g)** | **Weight of dry sample(g)** | **Moisture(%)** | **Dry matter(%)** |
| **Day 0** | 0.30 | 30.65 | 30.35 | 13.02 | 12.72 | 58.09 | 41.91 |
| **Day 7** | 0.30 | 30.80 | 30.50 | 8.93 | 8.63 | 71.70 | 28.30 |
| **Day 14** | 0.30 | 31.07 | 30.77 | 8.80 | 8.50 | 72.38 | 27.62 |
| **Day 21** | 0.40 | 31.20 | 30.80 | 8.50 | 8.10 | 73.70 | 26.30 |
| **Day 28** | 0.43 | 31.23 | 30.80 | 8.00 | 7.57 | 75.42 | 24.58 |

Table 8: Moisture analysis of banana stored -20°C

| sample | Peeled (vacuum packed) | | | | | | |
|---|---|---|---|---|---|---|---|
| **Items** | **Weight of container (g)** | **Weight of container + sample (g)** | **Weight of wet sample (g)** | **Weight after drying (g)** | **Weight of dry sample(g)** | **Moisture (%)** | **Dry matter (%)** |
| **Day 0** | 0.30 | 31.25 | 30.95 | 12.87 | 12.57 | 59.39 | 40.61 |
| **Day 7** | 0.50 | 31.23 | 30.73 | 12.90 | 12.40 | 59.65 | 40.35 |
| **Day 14** | 0.30 | 30.83 | 30.53 | 12.26 | 11.96 | 60.83 | 39.17 |
| **Day 21** | 0.40 | 31.00 | 30.60 | 12.23 | 11.83 | 61.34 | 38.66 |
| **Day 28** | 0.50 | 31.13 | 30.63 | 12.27 | 11.77 | 61.57 | 38.43 |
| | | | | | | | |
| **sample** | **Peeled (non-vacuum packed)** | | | | | | |
| **Items** | **Weight of container (g)** | **Weight of container + sample (g)** | **Weight of wet sample (g)** | **Weight after drying (g)** | **Weight of dry sample(g)** | **Moisture (%)** | **Dry matter (%)** |
| **Day 0** | 0.30 | 31.25 | 30.95 | 12.87 | 12.57 | 59.39 | 40.61 |
| **Day 7** | 0.30 | 30.80 | 30.50 | 12.55 | 12.25 | 59.84 | 40.16 |
| **Day 14** | 0.50 | 31.13 | 30.63 | 12.47 | 11.97 | 60.92 | 39.08 |
| **Day 21** | 0.40 | 31.23 | 30.83 | 12.15 | 11.75 | 61.89 | 38.11 |
| **Day 28** | 0.50 | 31.13 | 30.63 | 12.07 | 11.57 | 62.23 | 37.77 |

| sample | Unpeeled (vacuum packed) | | | | | | |
|---|---|---|---|---|---|---|---|
| **Items** | **Weight of container (g)** | **Weight of container + sample (g)** | **Weight of wet sample (g)** | **Weight after drying (g)** | **Weight of dry sample(g)** | **Moisture (%)** | **Dry matter (%)** |
| **Day 0** | 0.30 | 30.65 | 30.35 | 13.02 | 12.72 | 58.09 | 41.91 |
| **Day 7** | 0.30 | 30.97 | 30.67 | 12.18 | 11.88 | 61.27 | 38.73 |
| **Day 14** | 0.50 | 31.23 | 30.73 | 11.92 | 11.42 | 62.84 | 37.16 |
| **Day 21** | 0.40 | 31.13 | 30.73 | 11.57 | 11.17 | 63.65 | 36.35 |
| **Day 28** | 0.50 | 31.13 | 30.63 | 11.34 | 10.84 | 64.61 | 35.39 |

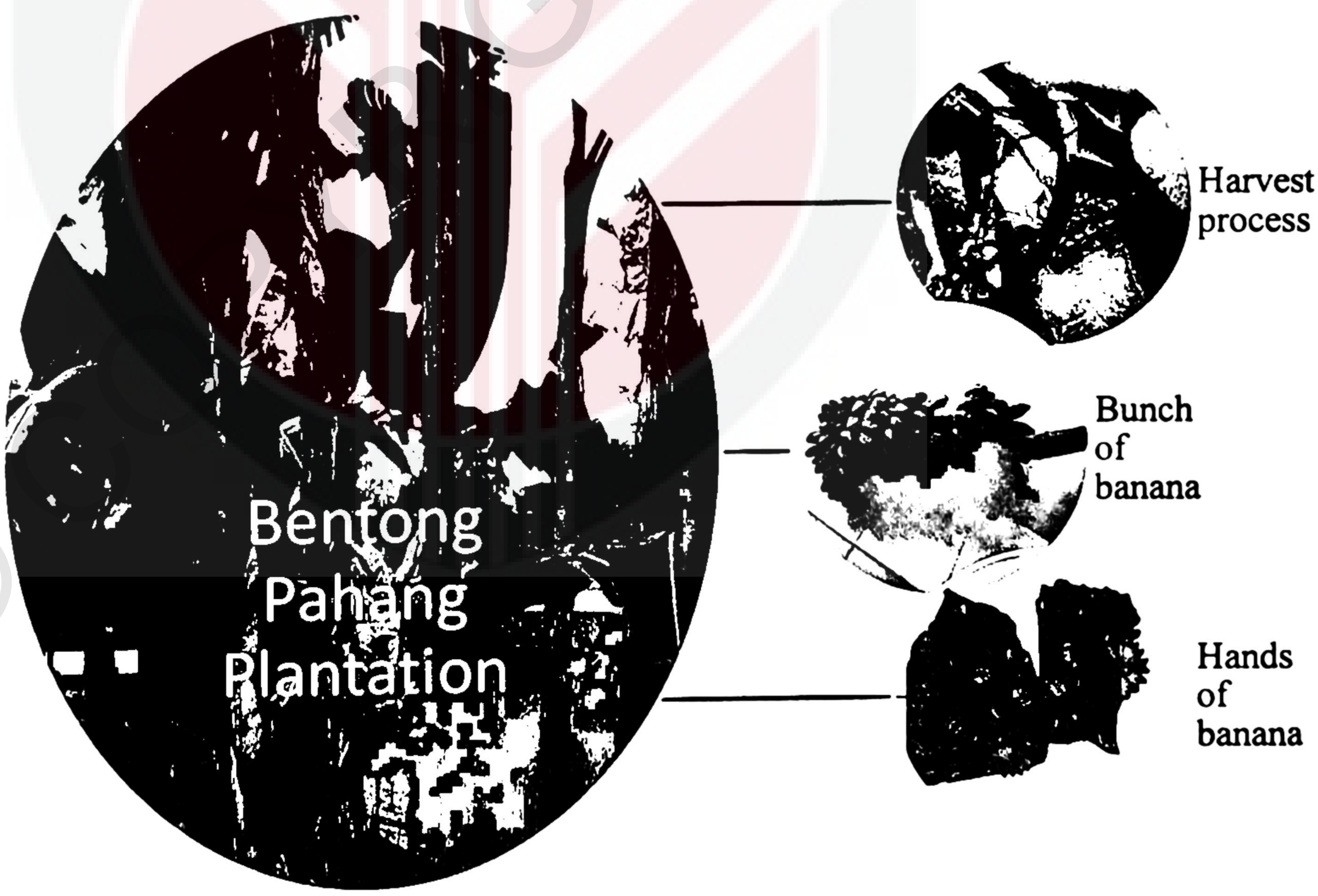
| sample | Unpeeled (non-vacuum packed) | | | | | | |
|---|---|---|---|---|---|---|---|
| **Items** | **Weight of container (g)** | **Weight of container + sample (g)** | **Weight of wet sample (g)** | **Weight after drying (g)** | **Weight of dry sample(g)** | **Moisture (%)** | **Dry matter (%)** |
| **Day 0** | 0.30 | 30.65 | 30.35 | 13.02 | 12.72 | 58.09 | 41.91 |
| **Day 7** | 0.30 | 30.63 | 30.33 | 10.43 | 10.13 | 66.60 | 33.40 |
| **Day 14** | 0.50 | 31.30 | 30.80 | 10.30 | 9.80 | 68.18 | 31.82 |
| **Day 21** | 0.40 | 31.17 | 30.77 | 9.90 | 9.50 | 69.13 | 30.87 |
| **Day 28** | 0.50 | 31.27 | 30.77 | 9.30 | 8.80 | 71.40 | 28.60 |

Table 9: Antimicrobial analysis of banana stored in -20ºC

| Weeks | No. of Colony | |
|---|---|---|
| | Vacuum packed | Non-vacuum packed |
| Day 0 | 20.00 | 20.00 |
| Day 7 | 22.50 | 32.50 |
| Day 14 | 39.50 | 61.67 |
| Day 21 | 65.00 | 129.50 |
| Day 28 | 114.00 | 202.00 |

| Weeks | Total CFU/mL | |
|---|---|---|
| | Vacuum packed | Non-vacuum packed |
| Day 0 | 2.00E+03 | 2.00E+03 |
| Day 7 | 2.25E+03 | 3.25E+03 |
| Day 14 | 3.95E+03 | 6.17E+03 |
| Day 21 | 6.50E+03 | 1.30E+04 |
| Day 28 | 1.14E+04 | 2.02E+04 |

## 6.2 Preparation of Sample




appendix 1: Harvesting Saba Banana from Bentong Plantation


appendix 2: First day preparation

## 6.3 Analysis Progress


appendix 3: Progress during sample analysis