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Physicochemical, Antioxidative and Sensory Properties of Muffin Using Different Drying Time of Cocoa Pulp

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A thesis submitted in fulfilment of the requirement for the degree of Bachelor of Applied Science (Product Development Technology) with Honours

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DECLARATION

I hereby declare that the work embodied in this report is the result of the original research and has not been submitted for a higher degree to any universities or institutions.

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Student

Name:

Date:

I certify that the report of this final year project entitled “Physicochemical, Antioxidative and Sensory Properties of Muffin Using Different Drying Time of Cocoa Pulp by Nur Suhaili binti Abdul Malek, matric number F15A0161 has been examined and all the correction recommended by examiners have been done for the degree of Bachelor of Applied Science (Product Development Technology) with Honours, Faculty of Agro-Based Industry, Universiti Malaysia Kelantan.

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Physicochemical, Antioxidative and Sensory Properties of Muffin Using Different Drying Time of Cocoa Pulp

ABSTRACT

There were abundance of cocoa pulp discarded to the environment. However, there were insufficient amount or little research being explored on utilizing edible by-product such as cocoa pulp to be incorporated into muffin. In this study, wheat flour was partially substituted with 10% of commercial cocoa powder, muffin without cocoa powder, 12 hours and 24 hours dried cocoa pulp.The textural properties, colour analysis and sensory acceptabilty were studied. The antioxidative properties were analyzed through DPPH and TPC method. There were no significant difference attributes(p≥0.05) in between muffin with 12 hours dried cocoa pulp compared to muffin with 24 hours dried cocoa pulp in all colour attributes The 12 hours dried cocoa pulp has the lowest value in hardness (5021 ± 15.62). Moreover, 12 hours dried cocoa pulp exhibited higher antioxidant properties than other muffin formulation (48.58 ± 0.00). Furthermore,the total phenolic content (TPC) of 12 hours dried cocoa pulp also was the highest (14.92 ± 0.94). For sensory acceptability, 12 hours dried cocoa pulp showed the highest overall acceptance (4.83 ± 10.8) than muffin with 24 hours dried cocoa pulp. In conclusion, this project had shown that it was potential to utilize the cocoa pulp into muffin in baking industry. The success of the project was depended on the acceptance of the consumers to have a new ingredients in the muffin. Cocoa pulp was an innovative idea and can be used to increase the economic use of cocoa. Besides, it providing a solution to the wastage problem occurred during harvesting cocoa fruit.

Keywords: Antioxidative properties, colour, muffins, texture, sensory acceptability

Fizikokimia, Antioksida dan Ciri Deria Muffin Menggunakan Masa Pengeringan yang Berbeza dari Pulpa Koko

ABSTRAK

Terdapat banyak pulpa koko dibuang ke persekitaran. Walau bagaimanapun, terdapat jumlah yang tidak mencukupi atau kajian yang banyak tentang penggunaan produk sampingan yang boleh dimakan untuk digantikan ke dalam muffin. Dalam kajian ini, tepung gandum telah digantikan sebanyak 10% dengan serbuk koko komersial, 12 jam dan 24 jam pulpa koko kering dan muffin tanpa serbuk koko. Sifat tekstur, analisis warna dan penerimaan sensori telah dikaji. Sifat antioksidan dianalisis menggunakan kaedah assay DPPH dan TPC. Tidak ada sifat perbezaan yang signifikan ($p \geq 0.05$) antara muffin dengan pulpa koko kering 12 jam dan muffin dengan pulpa koko kering 24 jam dalam semua sifat warna. Pulpa koko kering selama 12 jam mempunyai nilai terendah dalam kekerasan (5021 ± 15.62). Selain itu, muffin dengan pulpa koko kering selama 12 jam menghasilkan sifat antioksidan yang lebih tinggi daripada muffin yang lain (48.58 ± 0.00). Jumlah fenolik (TPC) pulpa koko kering selama 12 jam adalah (14.92 ± 0.94). Bagi penerimaan sensori, pulpa koko kering selama 12 jam menunjukkan penerimaan sensori yang tinggi (4.83 ± 1.08) daripada muffin yang dikeringkan selama 24 jam. Kesimpulannya, dari projek ini ia menunjukkan bahawa terdapat potensi untuk menggantikan pulpa koko ke dalam muffin dalam industri pembuatan pastri. Kejayaan projek ini bergantung kepada penerimaan pengguna untuk mendapatkan bahan-bahan baru dalam muffin. Pulpa koko adalah satu bentuk inovasi yang boleh digunakan untuk meningkatkan penggunaan buah koko secara meluas. Selain itu, ia dapat memberikan penyelesaian kepada masalah pembaziran yang berlaku semasa proses penuaian buah koko.

Kata kunci: Mufin, sifat antioksidan, tekstur, penerimaan sensori, warna

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TPC	Total Phenolic Content
DPPH	2,2- diphenyl-1-picrylhydrazyl
g	Gram
mg	Miligram
mL	Milimeter
%	Percentage
X	Times
±	Plus/ minus
L*	Lightness
a*	Redness
b*	Yellowness
≥	More than
≤	Less than
ANOVA	Analysis of variance
SD	Standard deviation

CHAPTER 1

INTRODUCTION

1.0 Research Background

Biscuit, muffin, cake, bread, pastries and pies are example of bakery product. They use high amount of flours mixed with many other ingredients and will baked in an oven. These baked items are mostly consumed as it easily available and convenient to be eaten as snack. However, most of bakery products are high in carbohydrate, fat and calorie, but low in fibre content (Mishra & Chandra, 2012). This issue regards to unhealthy choice for frequent consumption. Therefore, it is important to have an alternative ingredient used in bakery products that have nutritive value in order to increase the nutritional quality of the bakery goods.

Cocoa (*Theobroma cacao* L.) is from genus Theombroma, origin in the rainforest of Amazon basin and other tropical sites of South and Central America. The cocoa is usually used in food industry such as chocolate, cakes, biscuits, ice-cream and developed countries make sweet (Guehi *et al*., 2007). According to Asiedu (1991) cocoa contain 50% of fat and called as cocoa butter used in cosmetics and pharmaceutical sector. However, due to the expensive price of cocoa butter, chocolate industries have to replace the cocoa with other vegetal fats (Hoda *et al*., 2006).

In order to overcome this problem, the cocoa trader should think an idea on how to exploit by-products namely shells and juice. During cocoa harvesting, cocoa beans is fermented in tanks. Nowadays, some strategies are being directed in cocoa by-product valorization. These researches are mainly associated to cocoa pods and mucilaginous juice widely used. In Nigeria and Ghana country, cocoa pods is for soap fabrication (Antonio *et al*., 1993). Meanwhile, in Brasilia, the juice is found in alcoholic beverage, vinegar (Samsiah *et al*., 1991) and jelly fabrication (Wood & Lass, 1985).

Cocoa pulp from cocoa fruit could be substitute the common ingredients in muffin or other bakery product. Cocoa pulp is surrounded in the cocoa beans which is sweet and free of alkaloids and other toxic substances as well as does not easily get these substances on heating. "The mucilaginous pulp is composed of spongy parenchymtous cells containing cell sap rich in sugars, citric acid and salts." (Lopez *et al*., 1984). The cocoa pulp are usually been discarded and there is no further research on its potential use to increase the economic value.

Recent years, new and lot of methods to process fruits have been studied in order to decrease production losses, increasing farmer's income and to market the new product (Duarte et al.,2010). Therefore, the aim of this project to investigate the effect of antioxidative properties and acceptance of muffin as well as its physicochemical properties when added with dried cocoa pulp. Sensory evaluation of muffin with drying cocoa pulp was performed.

1.1 Problem Statements

The usage of cocoa fruit to produce cocoa powder and cocoa butter is only 10% of the overall weight of cocoa pod. The problem is the unutilized by-products and one of it is cocoa pulp in order to minimize loss and generate profit by exploiting them (Adomako, 1974 ; International Cocoa Organization, 2006). Then, the limited research and effort been done to utilize the cocoa pulp into processed food sector.

1.2 Hypothesis

H_o: There are no significant differences between colour, texture, antioxidative properties and sensory acceptability of mufins incorporated with dried cocoa pulp for 12 hours and 24 hours.

H_a: There are significant difference between colour, texture, antioxidative properties and sensory acceptability of muffins incorporated with dried cocoa pulp for 12 hours and 24 hours.

1.3 Objectives

1. To examine the effect of colour and texture of muffins that incorporated with commercial cocoa powder, without cocoa powder, cocoa pulp dried for 12 hours and 24 hours

2. To evaluate the sensory acceptability of muffins incorporated with commercial cocoa powder, without cocoa powder, 12 hours and 24 hours dried cocoa pulp among consumers by conducting sensory evaluation.
3. To determine the antioxidative properties (DPPH,TPC) of muffins that incorporated with commercial cocoa powder, without cocoa powder, cocoa pulp dried for 12 hours and 24 hours

1.4 Scope of Study

The main activity of this research was to determine the physicochemical, antioxidative and sensory properties of muffin incorporated with dried cocoa pulp. The cocoa pulp were dried for 12 hours and 24 hours. Cocoa fruit was obtained from Sungai Satan Farm, Ayer Lanas, Kelantan and the experiment was conducted at Universiti Malaysia Kelantan Jeli Campus. The colour analysis was performed using a chroma meter. The L*, a* and b* reading were taken. Then, the texture testing was use a texture analyzer and attributes such as hardness, springiness, cohesiveness and chewiness was selected.

To determine the antioxidant activity, 2,2-diphenyl-1-picrylhydrazyl (DPPH) was used to estimate the antioxidant activity on each muffin. Ascorbic acid was used as a standard and the percentage of DPPH inhibition was calculated. On the other hand, Folin-ciocalteu method was performed to identify the total phenolic content for each muffin. In this method, gallic acid standard was used to construct standard calibration curve and the result for phenolic content in each muffin was obtained through the absorbance value by spectrophotometer machine.

1.5 Significance of Study

This project is to determine the physicochemical, antioxidative and sensory properties in muffin incorporated with dried cocoa pulp for 12 hours and 24 hours. By performing this research, we should be able to determine whether dried cocoa pulp will result in highest total phenolic and antioxidant activity as well as it physicochemical and sensory acceptability among consumers. Furthermore, we also can identify the correlation between the total phenolic content and antioxidant activity of each muffin. Phenolic compound in fruits are commonly known in contributing a good effect towards human health. Thus, it believed that there is potential to develop a muffin with the cocoa pulp in baking industry. The success of the project is depending on the acceptability of the consumers to have new ingredient in the muffin. Cocoa pulp is an innovative idea and can be utilized to increase the economic use of cocoa.

1.6 Limitation of Study

Due to the insufficient time, total plate count analysis cannot be performed. As the cocoa pulp contain a citric acid (Guehi *et al*., 2010) thus it may act as a preservative and can prolong the shelf life.

CHAPTER 2

LITERATURE REVIEW

2.0 Literature review

2.1 Muffin

2.1. 1 Baking

Baking is a method of cooking which is transfer of heat via oven on the exterior of the food by radiation and convection together with conduction of heat from container to the sides and bottom of the product. The difference temperature cause changes of heat into interior of the cold batter (Majzoobi *et al*., 2015).

2.1.2 Ingredients

The ingredients used in baked goods specifically muffin had come to health concern to consumer as it usually have a moderate to high sugar content. The common ingredients used in muffin are flour, eggs ,fat, baking powder and milk. All these ingredients will be mixed that results in bubbles formation to the mixture. A successful muffin depends on its crumb texture and final volume.

2.1.3 Structure

The fine final structure is when the centre crumb contains small and evenly distributed gas cells. In this part the temperature increases at first and then will keep constant around 100 °C due to the availability of water is high. The structure of the top part is coarse with a larger distribution of gas cell sizes. The gelatinizing still occur so the gas cell walls are not rigid and extend to expand. The bottom part of the crumb play a roles in the migration to colder zones. The starch gelatinizes due to the availability of water falls greater in short time thus gas cell size is larger compared to the rest of the product (Ning-Bin, 1980).

2.1.4 Wheat flour

Wheat flour as one of the main ingredients in bakery goods. It is a challenging part due to wheat is imported to Malaysia as it never make its own. Increasing need and want for wheat-based products will cause an increasing importation of wheat flour thus looking other local source of replacement ingredient to substitute wheat flour in bakery products. It would also give many benefits to reduce relying on wheat as well as could significantly save foreign exchange (Ning-Bin, 1980).

2.1.5 Flour substitution

The substitution of whole wheat flour, wheat germ, rolled oats or bran as part of the flour in the muffin can increase the fiber content . Other flour usually used in muffins such as cornmeal, soy, oat, potato, and peanut (Ning-Bin, 1980). An acceptable product is possible when cowpea or peanut flours are substituted for 25% or when whole-wheat flour or corn meal is substituted for 50% of all-purpose flour (Holt *et al*., 1992). Acceptable muffins have been prepared when soy protein flour was substituted for 10–20% (Sim & Tam, 2001) or 100% of all-purpose flour (Bordi *et al*., 2001). All these flours do not contain glutenin or gliadin except whole wheat and bran in whole wheat flour cut and decrease gluten formation. Hence, there is little gluten development when these flours are used but the texture of muffins tend to be crumbly and compact unless other improvement are made in the formulation.

2.2 Cocoa fruit

Cocoa (*Theobroma cacao* L.) was from genus Theombroma, a group of small trees which was originated in the rainforest of Amazon basin and other tropical sites of South and Central America. The height of cocoa trees were around 4-8 meter and it starts bearing fruits after 5 years and takes 10 years to reach maximum yield. The mature cacao tree produces fruit in the shape of elongated podsit may produce 70 such fruits yearly. The pod has many ridges attached along its length and contains 20 to 60 seeds or cocoa beans in the long axis of the pod with colour ranges from brownish yellow to purple.The cocoa beans were about 2.5 cm long and were surrounded with a sweet sticky white pulp.

About 71% of world cocoa beans production was from Invory Coast, Africa with 33% of overall cocoa world supply (WCF, 2012). Meanwhile, in Asian-Ocenia, Indonesia was the highest producer of cocoa followed by Papua New Guinea and Malaysia based on 2014/2015 statistics by World Cocoa Foundation (Anon, 2015).

2.2.1 Location, production and statistics of cocoa tree in Malaysia

The cocoa plantation was started in year 1950 and the first commercial cultivation area was in Jerangau, Terenganu. Malaysia was one of the biggest producer of cocoa manufacturing and processing country in the world (ICCO, 2007). The Malaysian cocoa planting sector increase at peak production of 247,000 tonnes cocoa beans in 1990 and then decrease to 31,937 tonnes in 2006, while the local cocoa grindings industry progressed with increasing to 270,261 tonnes in 2006 The unstable growth of cocoa activities caused the requirement for importation of sufficient amount of cocoa beans to preserve the local grindings need (MCB, 2007).

According to the previous study, 95% of cocoa producers were smallholders which means they were majority producers of cocoa in Malaysia (Anon, 2015). Altough the production of cocoa in Malaysia was inconsistent, Malaysia was a major exporter of cocoa products such as cocoa paste, cocoa butter and cocoa powder. Cocoa powder and cocoa butter were important in processing food product and non food product such as chocolate, beverage, toiletries, pharmaceutical and cosmetic (Keen, 2001).

2.2.2 Sensory evaluation of cocoa

Cocoa product was important to deliver a flavour and the flavour can be obtained through fermentation and drying of cocoa beans (Wood GAR & Lass RA, 2001). The cocoa flavour obtained via fermentation and drying interact in the roasting process in order to produce a good chocolate flavour. The brown colour, mild aroma and texture of roasted beans were developed during roasting process. The flavour produced was the integration of 400-500 compounds such as pyrazines, aldehydes, ethers, thiazoles, phenols, ketones, alcohols, furans and esters (The SS & Birch EJ, 2014). Many compounds developed during roasting was aldehydes and pyrazines.

Ramli *et al*. (2006) investigate the effect of roasting process on sensory evaluation of dark chocolates. The author in his study revealed that higher temperature and longer duration of roasting cause to lower astringent taste. This was due to that polyphenol compund that act for the astringent cocoa degrade at high temperature and long roasting time (Ramli *et al*., 2006). The increasing of temperature from 120^{o}C to 170 ^{o}C cause the increasing in bitter flavour of cocoa beans. This was because of xanthine alkaloids (caffeine) and theobromine.

2.3 By-product of cocoa fruit

2.3.1 Cocoa pulp

The unutilized by-products of cocoa fruit is cocoa pulp or known as cocoa mucilage. (Afolabi et al., 2015). The pulp are embendded with cocoa beans or seed that come from the bean teguments. “The mucilaginous pulp is composed of spongy parenchymtous cells containing cell sap rich in sugars, citric acid and salts.” (Lopez *et al*., 1984). The damage of the pulp during handling and the pressure weight of the mass will produce pulp juice.

2.3.2 Cocoa pod husk

Cocoa pod husk is defined as discarded waste after the wet cocoa beans, sweatings and placenta have been removed. Cocoa pod husk also has various of condensed or polymarized flavanoids sometimes related with glucose.

2.3.3 Cocoa bean shells

During processing of cocoa beans, the seed coat or testa is removed. The main component of cocoa bean shell is a mulch. This mulch have estimately 2.5% nitrogen, 1%

phosphate and 3% potash and natural gum that is initiated when watered. This mulch has been involved in methylxanthine toxicosis among dogs (Ronald *et al*., 2013).

2.3.4. Cocoa butter

Cocoa butter is developed from discarded cocoa beans with small or no economical value has been used in manufacture of toilet soap and body pomade. It also can be used to produce soft soap.

2.3.5 Other by products of cocoa

The other by product are cocoa gum,activated carbon, dietary fibre, pectic enzyme, methylxanthine, theombromine, polyphenols, particle boards, essential oils, compost and biogas. All these by products are derived from cocoa waste (Ronald *et al*., 2013)

2.4 Pyhsiology of cocoa pulp

2.4.1 Nutrient and constituent in cocoa pulp

The cocoa pulp does not contain any alkaloids and other harmful or toxic substances as well as not simply get these substances on heating (Afolabi *et al*., 2015). Although it has a poor source of vitamins, protein and carbohydrate, but it has a good source of calcium, iron, fat and phosphorus. (Opeke, 2005). The pulp consists of 82-87% water, 10-15% sugar (60% is sucrose and 39% a combination of glucose and fructose), 2-3% pentoses, 1-3% citric acid and 1-1.5% pectin. (Claudia *et al*., 2012). Cocoa pulp is sweet, viscous liquid embedded with cocoa beans in the cocoa pod (Afolabi *et al*., 2015). The existence of citric acid results in low pH of cocoa pulp which is 3.0-3.5 (Tagro *et al*., 2010).

2.5. Cocoa processing

2.5.1 Fermentation

Generally, cocoa pods that are still attached to cocoa trees are naturally sterile and immune to any microbial interactions.The cocoa beans are attached to the pulp which are high in sugars content and provides an optimal condition for the growth of microorganism. The microbial activity of a succesion of yeast, lactic acid bacteria (LAB) and acetic acid bacteria (AAB) cause the development of lactic acid and acetic acid. The fermentation is important method to produce a high quality of final product. The pulp are removed from the cocoa fruit and become starter for microbial fermentation to produce a chocolate. During fermentation, the microorganism solubilises the pulp material embendded beans and produces a variety of metabolic end-products such as alcohols and organic acids which enter the beans to cause their death (Tagro *et al*., 2010). This reaction will generate a chocolate flavour, aroma and colour.

2.5.2 Drying

After fementation process, 50% of cocoa protein are estimated to be denatured. Therefore, drying method is important to avoid deterioration of beans by enzymes and microbial activity (Bravo A & McGaw DR, 1974). There are two techniques usually carried out the drying process which are natural sun and artificial hot air. These methods of drying are depends on weather conditions. Drying will results in chemical and biochemical changes to develop flavour and aroma during processing.

2.5.3 Roasting

Roasting was function to give flavour and appearance qualities of cocoa. The selection of roasting rely on the variety and quality of cocoa, the cultivation condition and during harvesting processs. During roasting, there was pyhsical and chemical changes happen such as evaporation of volatile acids which results in decrease in the acidity level thus decrease biterness taste. These reaction cause the favourable chocolate flavour and colour by Mailard reaction (Emmanuel *et al*., 2014)

2.6 Potential use of cocoa pulp

Some had a research on utilization of cocoa pulp extracted before and after fermentation. A fruit wine has been made by fermenting the fresh cocoa pulp by *Saccharomyces cerevisiae* strain (Dias *et al*., 2007). Juice and jellies has been made in Malaysia (Malaysian Cocoa Board, 2004) and in Ghana country, jams have been made from unfermented pulp (Cocoa Research Institute of Ghana, 2010). After fermentation process, the pulp is known as 'cocoa sweating', has been studied (Adams *et al*., 1982) that suitable starter to produce alcohol and vinegar. Furthermore, in Ghana, gin and brandy were commercially in their market.

However, the information about potential health benefits of cocoa pulp is very little and limited. There is maybe due to lot of competition between the cocoa pulp with other known antioxidant-rich commodities. Many research had been done on cocoa bean because of the presence of epicatechin, catechin and procyanidins (Keen *et al*., 2005). The flavonoid in cocoa bean also has been further studies to possess antioxidant activity. Therefore, cocoa pulp also will be investigated for antioxidant and total phenolic content in this project by using common assays in food applications.

2.7 Physical parameter of muffin

The attributes such as physical, chemical, texture, colour and sensory properties are important in order to measure the acceptance of any food product by the consumers. Muffin has sweet taste, high-calorie baked goods that are appreciated by consumers because of their good taste and soft texture. The article written by Rahman *et al*. (2015) study the physicochemical, textural and sensory properties of muffins incorporated with wheat grass powder. The texture of muffin such as hardness, gumminess and chewiness of muffins increased with higher level addition of wheat grass powder whereas the cohesiveness, springiness and colour L*, a*, b* values of muffin showed a reverse trend. In terms of sensory evaluation, the muffins with 7.5% wheat grass powder was rated lower in all sensory attributes and the muffins with 5.0% wheat grass powder was found most acceptable

2.8 Antioxidant

Antioxidant are compound that inhibit oxidation of food and does not include like sugar, cereal, oils, flours, herbs and spices (Khanna, 1987). Oxidation defined as a chemical reaction that can produce free radical. There were some studies said there were strong relation between antioxidant capacity and the phenolic content in fruit and plants. Antioxidant compounds found in food is crucial to the human health. The major sources of antioxidant such as fruits, vegetables and whole grains. Free radical damage can cause to cancer. There were plenty of research in phenolic compound and their antioxidant activity in the consumption of diet high in natural antioxidants with decreased risk of diseases related with oxidative stress such as cancer and cardiovascular diseases (Hatamnia *et al*., 2013). Fruits and vegetables has many different antioxidant components (Hong Wang, 1996). Then, antioxidants play a major role to increase the shelf life as well as preserving the nutritional and organoleptic qualities of baked goods.

2.9 Analysis of Phenolic Compound and Antioxidant Activity

The common method to determine total phenolic is by using Folin-ciocalteu assay (Ainsworth, 2007). Gallic acid is usually used as a standard and the total phenolic content is calculated by using a spectrophotometer with wavelength of 765 nm. Meanwhile, to determine the antioxidant activity, DPPH radical scavenging assay is one of the method to identify antioxidant activity through a DPPH scavenging system. DPPH or known as 2,2-diphenyl-1-picryhydrazyl is a free stable radical which can inhibit oxidation in samples. This method is performed with absorbance 517 nm by using a spectrophotometer machine (Musa *et al*., 2010).

2.10 Importance of Phenolic Compound and Antioxidants

Phenolic compounds are important to sensory, colour and antioxidant characteristics. For example, anthocyanins is mainly for colour to plants and fruits. Besides, flavonoids and non-flavanoids has an antioxidant properties lead to the identification of phenolic compounds in fruits and vegetables that important to high quality of life. Grapes, berries and tomatoes are the examples of phenolic compound. Phenolic compound may increase health benefits by decreasing the chances of metabolic syndrome and the related complications of type 2 diabetes. However, there is still very little and still need for further studies about the mechanism of phenolic that can prevent disease. Plenty of research had proven the benefits of phenolic compounds such as anti-aging, anti-inflammatory, antioxidant and anti-proliferative agents.

2.11 Sensory Evaluation

Sensory evaluation is examined with the human sense as human have five sense such as taste, smell, touch, sight and hearing. It assists to determine the properties of physical matter and enjoyment of products are usually related to the senses. Consumer evaluation can be used to predict whether the product is successful or unsuccessful. Sensory evaluation can be categorized by 2 types of testing which is objective and subjective. Objective testing is a form of questioning which has single correct answer and the sensory attributes are evaluated by selected or trained panel. Meanwhile, subjective testing is a form of questioning which may the answer us more than one or more than one way to express the correct answer (Philip, 2010). Therefore, sensory evaluation can give feedback and acceptance about the product before released to the market.

2.12 Statistical analysis

Statistical method is a study that include planning, designing, collecting data, analyzing, interpretation and reporting the research finding. To compare mean, usually it can be analyzed by using analyze of variance (ANOVA), t-test or other non-parametric method. ANOVA and t-test are one type of statistical analysis to analyze quantitative data. The purpose of using ANOVA is to see if there is any difference between groups of variables. The type of ANOVA such as one-way between groups, one-way repeated measures, two-way between groups and two-way repeated measures. The common used software system to calculate ANOVA such as SPSS, SAS, MS Excel and others (Ali & Bhaskar, 2016).

CHAPTER 3

MATERIALS AND METHODS

3.1 Location

The study was conducted at Food Laboratory and Biology Laboratory. Faculty of Agro Based Industry, Universiti Malaysia Jeli Campus, Kelantan.

3.2 Materials

3.2.1 Raw Materials

Raw materials such as cocoa fruit, wheat flour, sugar, eggs, butter, baking powder, baking soda and milk.

3.2.2 Equipment

Equipment was used in this experiment such as muffin trays, oven, bowl, hand mixer, spoon, micropipette (1000µl), test tube rack, test tube, cuvette rack, cuvette, dropper, media bottle, filter paper, filter funnel, volumetric flask, hot plate, beaker, measuring cylinder, aluminum foil, Brookfield CT-3 Texture Analyzer (USA), NB H3800 Panasonic Electric oven (Panasonic sdn.bhd, Malaysia), CR-400 Chromameter (Konika Minolta Sensing Americas, USA), SES6202 Saffron Precision weighing balance (Saffron Electronic Scales, Varachha Road, Surat, Gujrat, India), Panasonic Blender with Dry Mill and UV Spectrophotometer (Beckman DU640, Japan).

3.2.3 Chemicals and Reagents

2,2-diphenyl-1-picrylhydrazyl (DPPH), ethanol, methanol, ascorbic acid, gallic acid, sodium bicarbonate (Na_2Co_3), Folin-Ciocalteu reagent and distilled water.

3.3 Methods

3.3.1 Cocoa pulp preparation

Five kilogram of ripe cocoa fruit was obtained from Sungai Satan Farm in Ayer Lanas, Kelantan. The fruits was washed and split into half to extract the fresh seeds surrounded by the pulp. The cocoa pulp was dried for 12 hours and 24 hours by using an oven at 60^0 C.

3.3.2 Drying method

Two kilogram of cocoa pulp was dried using an oven at temperature of 60^0C until moisture content of 7% as stated (Hii *et al*., 2009). The cocoa pulp were dried for 12 hours and 24 hours. After drying, the pulp was grinded by using a blender to make it into powder and the cocoa powder were stored in airtight container.

3.3.3 Muffin formulation

The muffin weight was measured by measuring cup in unit of ml. The substitution was 10% of cocoa pulp.

Table 3.3.3: The muffin formulation of muffin incorporated with commercial cocoa powder, without cocoa powder, 12 hours and 24 hours dried cocoa pulp

Ingredients (ml)	**Control (0%)**	**Commercial cocoa powder**	**12 h**	**24 h**
Wheat flour	500	450	450	450
Cocoa pulp	0	0	50	50
Cocoa powder	0	50	0	0
Butter	125	125	125	125
Milk	125	125	125	125
Egg	79	79	79	79
Sugar	180	180	180	180
Baking powder	10	10	10	10
Baking soda	3	3	3	3
Total (ml)	**1022**	**1022**	**1022**	**1022**

3.3.4 Muffin preparation

The oven was heated at temperature of 180^{o}C. Sugar, egg and butter was mixed in large bowl and beaten using a hand mixer until the batter become smooth. Flour that has been sifted, baking powder, baking soda and milk. Cocoa pulp was dried for 12 hours and 24 hours and was added into the bowl separately. The oven was pre-heated at 10 minutes. Mixture was put into half of the muffin tray then was put in the oven for 20 minutes.

3.3.5 Determination of Texture Profile Analysis in Muffin

The texture profile analysis (TPA) was analyzed using a texture analyzer (Brookfield CT-3 Texture Analyzer). The instrument was set with a 5 kg of load cell and it force sensitivity was 1 g.Then, it was performed at test speed at 5mm s^{-1} at 75% of the original height while the post test speed was 5mm s^{-1} and there was a 5s interval between the two compression cycles. A trigger force was 5g and compression was performed with TA 11/1000 Cylinder 25.4 mm D and 35 mm L. Muffin was tested for springiness, cohesiveness, chewiness and hardness (Baixauli *et al*., 2008; Sanz *et al*., 2009).

3.3.6 Determination of Colour Attributes in Muffin

The colour of the muffin was measured by using Chroma meter. Firstly, the chroma meter was calibrated by using a standard white tile provided. The upper and lower layer of muffin was cut and colour inside the muffin was take. The term L*, a* and b* values were analyzed. L* indicates lightness while a* and b* indicates redness to greeness and yellowness to blueness respectively (Baixauli *et al*., 2009). Each sample was individually measured in triplicates.

3.3.7 Determination of DPPH Free Radical Scavanging Capacity in Muffins

3.3.7.1 Preparation of muffin extract

The samples (Commercial cocoa powder, cocoa powder for 12 hours, cocoa powder for 24 hours and flour) were prepared. 20 g of each cocoa powder were extract in boiling water for 10 minutes. Then, it was filtered and mark up to 100 ml in volumetric flask. 10 g of flour was used to avoid it from sticky.

3.3.7.2 DPPH assay of muffin

The 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging activity on antioxidant properties of muffin was performed by using method of (Yang *et al*., 2008). DPPH solution (0.1mM) was prepared by dissolving 4 mg of DPPH into 100 ml of ethanol. The solution was kept in dark place for 30 minutes to complete the reaction. The solution called as stock solution and was kept in 4^{o}C until used.

The sample extracts was diluted through a serial dilution into a concentration of 6.25, 12.5, 25, 50, 100 and 200 mg/ml. 2 ml of diluted solution were then added with 2 ml of prepared DPPH. The mixed solution was incubated for 30 min at room temperature (29^{o}C) in dark place and the absorbance of the solution was measured at 517 nm. The measurement of sample solution at each concentration was repeated three times. The percentage of inhibition was calculated by using the formula :

$$\text{Percentage(\%) of inhibition} = \frac{\text{A blank- A sample}}{\text{A blank}} \times 100 \qquad \text{------ Equation 1}$$

3.3.7.3 Ascorbic Acid Standard Curve

0.002 g of ascorbic acid was dissolved in 2.5ml of ethanol to perform a serial concentration of 6.25, 12.5, 25, 50,100 and 200 mg/ml. The ascorbic acid solution was determined with DPPH assay to get a standard curve to compare.

3.3.8 Determination of Total Phenolic Content in Muffins

0.005 g of each sample extract was dissolved in 5 ml of distilled water. The test tube was filled with 9ml distilled water and 1 ml was transferred from each tube. Then,1.5 ml of 10% (v/v) Folin-Ciocalteu reagent was added and incubated for 5 minutes followed by the addition of 2 ml of 7.5% (w/v) Na_2CO_3 and was incubated at dark room temperature (29^oC) for 2 hours to allow colour development. Then, the extract and standard sample was transferred into a cuvette and was measured at absorbance of 765 nm on the spectrophotometer three times.

3.3.8.1 Gallic Acid Calibration Standard

0.001 g gallic acid was dissolved in 9ml of methanol to create concentration at 100, 10, 1, 0.1 and 0.01 mg/liter respectively. According to Waterhouse (2002), the gallic acid solution was covered, labeled and stored at 4^oC when not in use. The gallic acid solution were analyzed by Folin-Ciocalteu method. The total phenolic content was calculated as gallic acid equivalent (GAE) by using the equation T= C x V/M using formula in Equation 2.

TPC (mg GAE/g) = (CxV) /M ---------- Equation 2

C = concentration of unknown from standard curve (µG/mL)

V= volume of extrct used (mL)

M= weight of crude extrac

3.3.9 Sensory analysis

The sensory test was conducted to determine the consumer likeness towards muffin formulation. Muffin incorporated with cocoa pulp was evaluated in terms of colour, aroma, hardness, chewiness, taste apperance and overall acceptability (Sanz *et al*., 2009) by thirty random (n=30) utrained students from Universiti Malaysia Kelantan Jeli Campus. The samples were evaluated on 7-point hedonic scale (1= extreamly dislike and 7= extreamly like) (Meilgaard *et al*., 2000). Sensory panelists received a cup of water and a sensory evaluation form. Panelists was given one sample at a time.

3.3.10 Statistical analysis

The data in the experiment was reported as mean ± SD by using one-way analysis of variance (ANOVA) by using IBM SPSS Statistic 21. The mean comparison from 3 replicates was performed using the Tukey post-hoc multiple comparison test at 95% confidence level ($P < 0.05$) except for sensory analysis was use Duncan procedure.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Influences of Different Drying Time of Cocoa Pulp on Colour Attributes of Muffins

Table 4.1 shown the colour attributes of muffin with cocoa powder, without cocoa powder, 12 hours and 24 hours dried cocoa pulp. The lightness (L*), redness (a*) and yellowness (b*) of muffin with commercial cocoa powder and muffin without cocoa powder showed significant difference ($p \leq 0.05$). All the colour attributes on muffins for both 12 hours and 24 hours dried cocoa pulp was not significant difference to each other ($p \geq 0.05$). Muffin with 12 hours dried cocoa pulp was 41.07 ± 0.03, 17.99 ± 2.18 and 35.07 ± 2.19 respectively while 24 hours dried cocoa pulp was 41.54 ± 2.12, 16.49 ±.2.21 and 36.06 ± 1.56 respectively. Muffin with 12 hours and 24 hours dried cocoa pulp was lighter compared to commercial cocoa powder but a little darker compared to muffin without cocoa powder.

Colour was an important criterion in baked goods product because it can reflect the first impression towards consumers. The muffin without cocoa powder which using a 100 %

wheat flour exhibited the lightest colour (highest L* reading), 48.62 ± 0.71 in comparison with commercial cocoa powder and dried cocoa pulp incorporated muffin. It was observed that baked muffins incorporated with cocoa powder were darker than control. As reported by Rashida *et al* (2015) the L* value of control muffin was 49.42 ± 2.31 compared to the muffin with wheat grass powder where it value showed not more difference with this project.

Furthermore, a* (red component) and b* (yellow component) value of control muffin was still has the highest reading compared to other formulations. This could be due to uneven exposure of muffin's surface area to high baking temperature and effect colour from compounds such as process of caramelization and Millard reaction. The Millard reaction, non-enzymatic browning reacts with reducing sugars and protein that result in reddish brown colour (Lathia, 2011). The other contributing factor is increasing in density. The density of bakery products was affected by the trapping of gas cells within the structure with denser products having a lower volume fraction of air cells. The higher density causes a dark colour by increasing the number of pigment molecules per square area.

The commercial cocoa powder shows the lowest L* value, 20.92 ± 0.00 indicate that it has the darkest colour. The darker colour may due to the alkalization treatment during cocoa processing that aim for browning, wettability, dispersability flavor as well as to increase the nutritional and biological qualities of cocoa (Serra *et al*., 2002).

Table 4.1: Comparison of colour attributes of muffin incorporated with commecial cocoa powder, without cocoa powder to 12 hours and 24 hours dried cocoa pulp

Muffin with cocoa powder	Colour attributes		
	L*	***a****	***b****
Commercial cocoa powder	20.92 ± 0.00 [c]	8.26 ± 0.54 [b]	12.26 ± 0.45 [c]
Without cocoa powder	48.62 ± 0.71 [a]	19.37 ± 0.61 [a]	41.79 ± 1.2 [a]
12 hours dried cocoa pulp	41.07 ± 0.03 [b]	17.99 ± 2.18 [a]	35.07 ± 2.19 [b]
24 hours dried cocoa pulp	41.54 ± 2.12 [b]	16.49 ± 2.21 [a]	36.06 ± 1.56 [b]

[a-b] The means with different superscripts letter in the same row indicates significance difference (p≤0.05). The value was mean ± SD

4.2 Influences of Different Drying Time of Cocoa Pulp on Texture Properties of Muffins

According to Table 4.2, a texture profile analysis of muffin was assessed in term of hardness, springiness, chewiness and cohesiveness. It illustrated that the incorporation of dried cocoa pulp reduced the springiness and chewiness of the muffin. The chewiness of 12 hours dried cocoa pulp was found to be the lowest (106 ± 89.47) compared to 24 hours dried cocoa pulp (257 ± 59.63), without cocoa powder (350 ± 57.15) and commercial cocoa powder (378 ± 92.08). Meanwhile, in terms of hardness of dried cocoa pulp muffins sample has no significant affect but muffin with 12 hours dried cocoa pulp shown the lowest hardness (5021 ± 15.62) compared to all the muffin formulation.

Hardness means as the highest peak force during the first compression cycle. The table 4.2 show that the muffin with commercial cocoa powder has the highest hardness, 5054.67 ± 21.57 compared with other muffins. The hardness increase because of the high volume that cause the gluten net with high strength and gas retention (Kaak & Pedersen, 2005). As studied by Grigelmo-Miguel *et al* (1999) about substitution of wheat flour with peach dietary fibre in muffin shown an increasing in hardness is because of density and decrease number of air pockets.

Springiness is a quality attributes of muffin that indicates the ability of food recovers during the time that elapses between the end of the first compression and the start of the second compression. Springiness is related with freshness in a product with a high quality muffin having higher springiness reading. Muffin with commercial cocoa powder has the highest springiness while the lowest is muffin with 12 hours dried cocoa pulp. As stated by Sanz *et al*. (2009) reducing of springiness in muffin is associated with the decreasing number of muffin air bubbles and existence of a denser matrix.

Next, no significant different ($p \geq 0.05$) was recorded in cohesiveness of muffin with commercial cocoa powder, without cocoa powder and 24 hours dried cocoa pulp but it increases significantly ($p \leq 0.05$) in 12 hours dried cocoa pulp muffin. Cohesiveness indicates the sensory crumbliness and perceptions associated to denseness of muffin and energy required to bite the food pieces. 12 Hours dried cocoa pulp muffin has the higher cohesiveness compared to the other muffin. This show that it need more energy to compress.

Based on Jauharah *et al*. (2014), studied on incorporation of young corn powder in muffin observed that higher substitution was less cohesive than control where muffin crumbled easier during handling thus show that it has a good quality muffin.

Chewiness is measured as the product of hardness, cohesiveness and springiness. Commercial cocoa powder has the highest chewiness and muffin without cocoa has the lowest chewiness. Even though the incorporation with 12 hours and 24 hours dried cocoa pulp into muffin had reduced some of the textural attributes quality but it has highest hedonic score by sensorial evaluation for chewiness which were 3.7 ± 1.53 and 3.53 ± 1.66 respectively compared to muffin without cocoa powder, 3.31 ± 1.62.

Table 4.2: Comparison of texture properties of muffin incorporated with cocoa powder,without cocoa powder to 12 hours and 24 hours dried cocoa pulp (n=3)

Muffin with cocoa powder	**Texture properties**			
	Hardness	**Cohesiveness**	**Springiness**	**Chewiness**
Commercial cocoa powder	5054 ± 21.57 [a]	0.66 ± 0.08 [a]	15.12 ± 8.11 [a]	378 ± 92.08 [a]
Without cocoa powder	5037 ± 24.54 [a]	0.65 ± 0.07 [a]	11.85 ± 3.64 [a]	350 ± 57.15 [a]
12 hours dried cocoa pulp	5021 ± 15.62 [a]	0.84 ± 0.05 [a]	5.54 ± 1.38 [a]	106 ± 89.47 [b]
24 hours dried cocoa pulp	5038 ± 19.04 [a]	0.61 ± 0.07 [a]	9.94 ± 4.02 [a]	257 ± 59.63 [ab]

[a-b] The means with different superscripts letter in the same row indicates significance difference ($p \leq 0.05$). The value was mean ± SD

4.3 Influences of Different Drying Time Total Phenolic Content in Muffins

Standard calibration curve was used to determine total phenolic content by using gallic acid. The advantages of using gallic acid was because of its price, stability and availability (Waterhouse, 2002). Based on Figure 4.3.1 the calibration curve showed a linear graph for gallic acid in range of concentration 0.1 µg/ml. The correlation coefficient (R^2) for the calibration curve is 0.981.

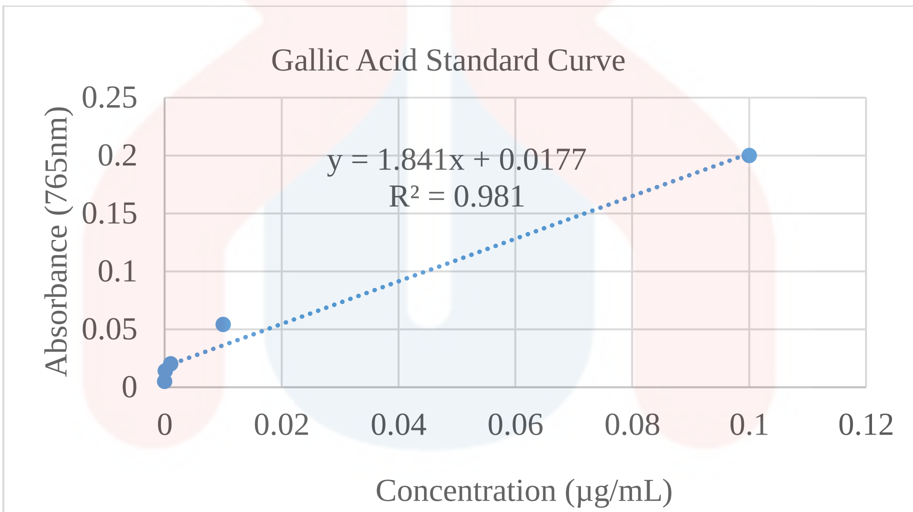

Figure 4.1 : Standard calibration curve of absorbance against concentration with gallic acid at concentration of 0.01, 0.1, 1, 50, 100 µg/ml

Total phenolic content of muffin with cocoa powder was listed in Table 4.3.1. Commercial cocoa had a significant ($p \leq 0.05$) total phenolic content (20.06 mg GAE/g) than muffin without cocoa powder (2.23mg GAE/g). Total phenolic content in 12 hours dried cocoa pulp was higher (14.92 ± 0.94) than 24 hours dried cocoa pulp (10.39 ± 0.62) but the lowest phenolic content was found in muffin without cocoa powder. According to Martinez *et al.* (2012) that determined the total phenolic content of cocoa mucilage sample by extraction with methanol was ranged in 173.67 and 182.63 mg GAE/100 g. The difference of total phenolic content with current study might because of the difference in sample preparation and extraction methods. However, there were no research found regarding on total phenol content in food incorporated with cocoa pulp in different drying time which could be compare results for this study.

According to Madrau *et al*. (2009), drying at low drying temperature (55 to 60^{0}C) in availability of oxygen, resulting to a polyphenoloxydese (PPO) enzymatic activity. Since low drying temperature need longer drying time compared to high temperature , it cause polyphenols content decrease. Kyi *et al*. (2005) studied on the oxidation reaction kinetic of polyphenols in cocoa beans drying at temperature of 40^{0}C and 60^{0}C. The author stated that higher temperature caused to the higher polyphenols in cocoa. The thermal processing can effect the phytochemical of fruit. It breakdown the cell structure which resulted in changes of component. As a result, it losses by breakdown some chemicals reaction such as enzymes, light and oxygen. Low total phenolic content during drying also can be caused by the binding of polyphenols with other component or alterations in chemical structure of polyphenols which cannot be determined by available methods. Polyphenolic compound can be degraded depends on it reaction towards polyphenol oxidase activity, organic acid content, sugar concentration and pH.

Drying of cocoa beans showed higher theobromine (19.44 ± 0.34 mg g^{-1}) and caffein (2.74 ± 0.03 mg g^{-1}) value that indicates there is a methylxanthine content reduction through drying. The study was similar with Belscak *et al*. (2009) findings that showed the theobromine (16 mg g^{-1}) and caffeine (2 mg g^{-1}) in the dry cocoa analysis. The high total phenolic content is because of the method which was Folin-Ciocalteu asay used based on the oxidation and reduction content measured by the presence of other reducing non-phenolic component like carbohydrate, pigments and Maillard process thus formation reductones and melanoidins in Maillard reaction with amino acids and low sugars resulting to high total phenolic content upon heat condition.

Table 4.3: The total phenolic content of muffin incorporated with commercial cocoa powder, without cocoa powder, 12 hours and 24 hours dried cocoa pulp

Muffin with cocoa powder	Total Phenolic Content (Mean ± SD)
Commercial cocoa powder	20.06 ± 0.06^{a}
Without cocoa powder	2.23 ± 0.06^{d}
12 hours dried cocoa pulp	14.92 ± 0.94^{b}
24 hours dried cocoa pulp	10.39 ± 0.62^{c}

$^{a\text{-}b}$ The means with different superscripts letter in the same row indicates significance difference ($p \leq 0.05$). The value was mean ± SD

4.4 Influences of Different Drying Time on Scavenging Activity of Muffins

The antioxidant activity of each muffin was performed by using DPPH radical scavenging assay. Each samples performed in a concentration of 200,100,50,25,12.5,6.25 mg/ml. According to the table 4.3.1 the highest antioxidant activity was muffin with 12 hours dried cocoa pulp ,48.58 ± 0.00. The lowest percentage (%) of DPPH inhibition was muffin without cocoa powder, 8.77 ± 0.12. As longer time of drying, the percentage of inhibition decrease as shown in table, 13.71 ± 0.24.

Muffin without cocoa powder showed the lowest antioxidant activity. Wheat is known to have hydroxycinnamic acid derivatives which demonstrated antioxidant activity (Andresean *et al*., 2001). As stated by Zhou *et al*. (2004) ferulic acid was the main phenolic acid which contained around 57-77% of total phenolic acids in wheat. This acids cause lower antioxidative properties than rutin based on the structural components (Cook & Samman, 1996). Then, muffin with commercial cocoa powder was higher than muffin without cocoa powder. According to Namiki (1990), the inhibition was higher because of high reactive nature of hydoxyl radicals. Previous study showed that cocoa oil and cake have the ability to scavange more highly reactive hydroxyl radicals and stopping the chain reactions in biological macromolecues (Lugasi *et al*., 1999).

Next, since the cocoa pulp was dried this may lead to heat-sensitive compounds as a factor to the antioxidant in muffin formulations of dried cocoa pulp destroyed during the baking where only certain phenolic compounds remain in muffin which was heat resistant (Rupasinghe *et al*., 2008). The other reason for the loss of phenolic content was because of binding of phenols to proteins, changes in chemical compound or method of extraction. The decreased in phenolic compounds decreased with the high temperature and storage time may because of oxidation and polymerization of phenolic compounds. This can be concluded that drying process at low temperatures which result to long drying times can decrease the antioxidant activity (Vega-Galvez *et al*., 2009).

Table 4.4 : The % DPPH inhibition of muffin incorporated with commercial cocoa powder, without cocoa powder, 12 hours and 24 hours dried cocoa pulp

Muffin with cocoa powder	Percentage of Inhibition (Mean ± SD)
Commercial cocoa powder	43.2 ± 0.00 [b]
Without cocoa powder	8.81 ± 0.12 [d]
12 hours dried cocoa pulp	48.58 ± 0.00 [a]
24 hours dried cocoa pulp	13.71 ± 0.24 [c]

[a-b] The means with different superscripts letter in the same row indicates significance difference (p≤0.05). The value was mean ± SD

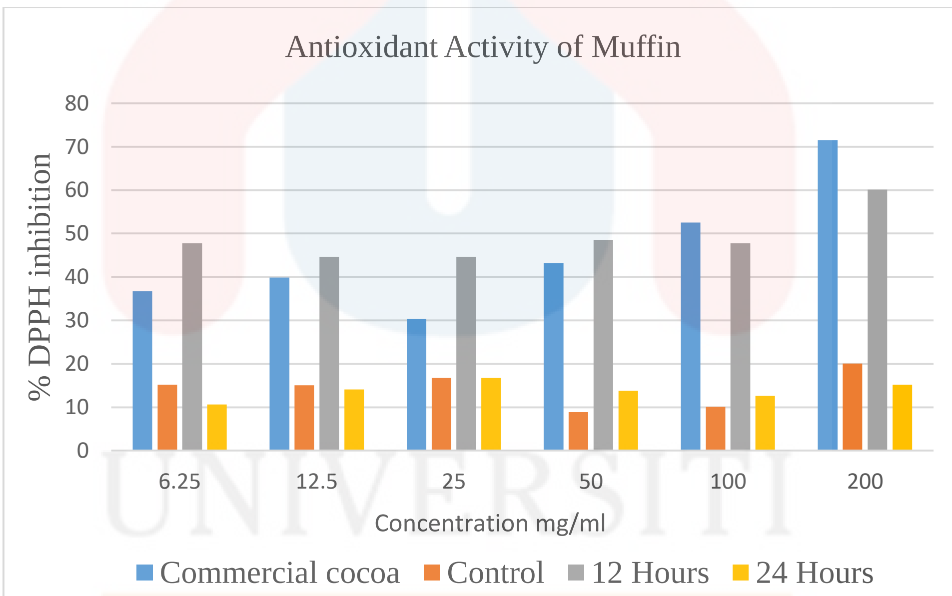

Figure 4.2: The antioxidant activity of muffin with commercial cocoa powder, without cocoa powder, 12 hours and 24 hours dried cocoa pulp. 12 hours dried cocoa pulp muffin follow the trend of commercial cocoa powder

4.5 Influences of Different Drying Time of Cocoa Pulp on Sensory Acceptability of Muffins

According to Table 4.5, the average aroma attribute ratings of muffin with commercial cocoa powder, without cocoa powder, 12 hours dried cocoa pulp and 24 hours dried cocoa pulp. The statistical result showed the sensorial attributes data of muffins were not significantly affected with 12 hours and 24 hours dried cocoa pulp. For colour attribute of muffin, commercial cocoa powder has the highest score among the formulations but not significant with control. Control muffin significantly less in chewiness and aroma whereas 12 hours dried cocoa pulp has the highest hedonic score in term of aroma and appearance.

The sensory panelist rated commercial cocoa powder the highest mean score, 5.03 ± 1.50 in term of aroma followed by 12 hours dried cocoa pulp, 5 ± 1.64 and control, 4.58 ± 1.19. The lowest is 24 hours dried cocoa pulp with mean 4.33 ± 1.69. Aroma means smell that help consumers to discover and enjoy food as much as taste. 12 hours dried cocoa pulp has not much significant difference of mean with commercial cocoa powder. This shows that the aroma of 12 dried cocoa pulp can be accepted among consumers. The aroma development of cocoa seed was developed during the fermentation process by migration of aroma compound to pulp into the seed tissue thus develop a flavor cocoa notes.(Eskes, A. B *et al.*, 2007).

Besides, it can be concluded that most panels prefer darker colour of muffin crumb. The data recorded showed that commercial cocoa powder has the highest acceptability compared to other formulations in terms of colour but has no significant difference with control muffin. Based on the colour analysis, the lowest lightness was found in commercial cocoa powder muffin which gave the darker crumb while the higher intensity was found in 12 hours dried cocoa pulp which obtained lower acceptability score than control and muffin with commercial cocoa powder.

According to the results obtained from sensory analysis, highest score was recorded for muffin with 12 hours dried cocoa pulp in term of aroma and appearance. Therefore, it can be concluded that the panelist prefers a light cocoa aroma muffin with a mild brown colour appearance compared to the commercial cocoa powder muffin.

Lastly, based on the data obtained, muffin incorporated with 12 hours dried cocoa pulp received higher overall acceptability compared to muffin incorporated with 24 hours dried cocoa pulp. Although muffin without cocoa powder received higher acceptance, 5.14 ± 0.88 than muffin with 12 hours dried cocoa pulp but it showed no significant difference ($p>0.05$) which most probably some of the panelist might not like the flavor of cocoa. Muffin with 12 hours dried cocoa pulp had the highest hedonic score ,4.83 ± 1.08 for overall acceptance which indicate higher acceptance level by the sensory panelists. The lowest rating was observed in muffin with 24 hours dried cocoa pulp in all of the attributes.

4.5 Sensory Acceptability

Table 4.5: Sensory acceptability of muffin incorporated with commercial cocoa powder, without cocoa powder and 12 hours and 24 hours dried cocoa pulp based on 7-hedonic scale (n=3)

Muffin with cocoa powder	**Colour**	**Aroma**	**Hardness**	**Chewiness**	**Taste**	**Appearance**	**Overall Acceptance**
Commercial cocoa	5.40 ± 1.00 [a]	5.03 ± 1.50 [a]	5.17 ± 0.99 [a]	5.1 ± 1.03 [a]	5.33 ± 1.30 [a]	5.4 ± 1.04 [a]	5.33 ± 0.92 [a]
Without cocoa powder	5.27 ± 1.02 [a]	4.63 ± 1.19 [a]	4.13 ± 1.41 [b]	3.31 ± 1.62 [b]	4.63 ± 1.03 [b]	5.18 ± 0.88 [b]	5.14 ± 0.88 [a]
12 hours dried cocoa pulp	4.97 ± 0.84 [ab]	4.90 ± 1.64 [a]	3.8 ± 1.42 [b]	3.7 ± 1.53 [b]	4.7 ± 1.26 [b]	5.17 ± 0.95 [ab]	4.83 ± 1.08 [a]
24 hours dried cocoa pulp	4.60 ± 1.10 [a]	4.33 ± 1.69 [a]	3.7 ± 1.66 [b]	3.53 ± 1.66 [b]	4.13 ± 1.38 [b]	4.33 ± 0.96 [b]	4.27 ± 1.14 [b]

[a-b] The means with different superscripts letter in the same row indicates significance difference ($p \leq 0.05$). The value was mean ± SD

CHAPTER 5

CONCLUSION AND RECOMMENDATION

5.1 Conclusion

From this study, muffin incorporated with 12 hours dried cocoa pulp had improved the physicochemical in term of colour and texture, had higher in antioxidative and had a higher overall acceptance in sensory acceptability among sensory panelist. There were no significant differences between muffin with 12 hours dried cocoa pulp and 24 hours dried cocoa pulp in term of L*, a* and b* attributes. However, muffin with 12 hours dried cocoa pulp exhibit lighter colour compared to muffin with 24 hours dried cocoa pulp. Muffin incorporated with 12 hours dried cocoa pulp had higher antioxidative activity than muffin incorporated than 24 hours dried cocoa pulp. This was because the longer drying time cause polyphenols content degrade. For sensory acceptability, the overall acceptance of 12 hours dried cocoa pulp received highest score than muffin with 24 hours dried cocoa pulp. Although panelists were rated higher for muffin without cocoa powder for overall acceptance but it had no significant difference with muffin with 12 hours dried cocoa pulp. The higher score might because some of the sensory panelist do not like the flavor of cocoa. It can be concluded that formulations of muffin with different drying time of cocoa pulp was successfully developed where 12 hours dried cocoa pulp was the best formulation to incorporate with muffin.

Therefore, cocoa pulp is a by-product of cocoa fruit that has potential to be added into bakery product to improve the physicochemical, antioxidative capacity and sensory properties among consumers.

5.2 Recommendation

Further research on investigational on nutritional composition, potential health benefit compound shall be performed on cocoa pulp to clarify their functional properties in bakery products. Cocoa pulp has a great potential to be developed as a value-added that promoting health and act as a natural preservative. The cocoa pulp can be dried thus it preserve the fruit by lowering the water and microbial activity as well as increase fruit stability and maintain their chemical characteristics. However, to obtain a good quality of dehydrated product, drying process should be that allow effective retention of colour appearance, flavor, taste and nutritive value. The oldest drying technique method of preservation can be applied. In conclusion, an extensive literature research and updated information, low known high value fruit is necessary to help develop underutilized by product of fruit and prevent wastage thus safe the environment in Malaysia.

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APPENDIX A

Table A.1: Statistical analysis of colour attributes of muffin

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
L	Between Groups	1279.597	3	426.532	339.593	.000
	Within Groups	10.048	8	1.256		
	Total	1289.645	11			
a	Between Groups	223.649	3	74.550	29.068	.000
	Within Groups	20.517	8	2.565		
	Total	244.166	11			
b	Between Groups	1528.050	3	509.350	235.361	.000
	Within Groups	17.313	8	2.164		
	Total	1545.363	11			

Table A.2: Statistical analysis of texture properties of muffin

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Hardness of muffin	Between Groups	1701.667	3	567.222	1.355	.324
	Within Groups	3348.000	8	418.500		
	Total	5049.667	11			
Springiness of muffin	Between Groups	110220.062	3	36740.021	1.007	.438
	Within Groups	291865.030	8	36483.129		
	Total	402085.093	11			
Cohesiveness of muffin	Between Groups	.094	3	.031	.564	.654
	Within Groups	.446	8	.056		
	Total	.540	11			
Chewiness of muffin	Between Groups	134760.982	3	44920.327	7.709	.010
	Within Groups	46613.195	8	5826.649		
	Total	181374.177	11			

Table A.3: Statistical analysis of percentage of DPPH inhibition of muffin

ANOVA

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	3677.240	3	1225.747	74100.562	.000
Within Groups	.132	8	.017		
Total	3677.373	11			

Table A.4: Statistical analysis of Total Phenolic Content of muffins

ANOVA

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	514.128	3	171.376	539.357	.000
Within Groups	2.542	8	.318		
Total	516.670	11			

Table A.5: Statistical Analysis of Sensory Properties of muffins

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
colour of muffin	Between Groups	11.358	3	3.786	3.879	.011
	Within Groups	113.233	116	.976		
	Total	124.592	119			
Aroma of muffin	Between Groups	8.625	3	2.875	1.248	.296
	Within Groups	267.300	116	2.304		
	Total	275.925	119			
Hardness of muffin	Between Groups	39.692	3	13.231	6.728	.000
	Within Groups	228.100	116	1.966		
	Total	267.792	119			
chewiness of muffin	Between Groups	44.558	3	14.853	6.852	.000
	Within Groups	251.433	116	2.168		
	Total	295.992	119			
taste of muffin	Between Groups	21.800	3	7.267	4.647	.004
	Within Groups	181.400	116	1.564		
	Total	203.200	119			
appearance of muffin	Between Groups	21.625	3	7.208	7.966	.000
	Within Groups	104.967	116	.905		
	Total	126.592	119			
overall acceptance of muffin	Between Groups	17.267	3	5.756	5.592	.001
	Within Groups	119.400	116	1.029		
	Total	136.667	119			

APPENDIX B

SENSORY EVALUATION TEST OF MUFFIN INCORPORATED WITH DRIED COCOA PULP

Age: ___ years   Panel No:

Race: Malay / Chinese / Indian / Others   Sample No:

Gender: Male / Female   Date:

Directions: Please circle your degree of likeness based on the characteristics below.

Do rinse your mouth before and after testing each sample.

1. Colour

1 2 3 4 5 6 7

Extremely dislike   Neither like nor dislike   Extremely like

2. Aroma

1 2 3 4 5 6 7

Extremely dislike   Neither like nor dislike   Extremely like

3. Hardness

1 2 3 4 5 6 7

Extremely dislike   Neither like nor dislike   Extremely like

4. Chewiness

1 2 3 4 5 6 7

Extremely dislike | Neither like nor dislike | Extremely like

5. Taste

1 2 3 4 5 6 7

Extremely dislike | Neither like nor dislike | Extremely like

6. Appearance

1 2 3 4 5 6 7

Extremely dislike | Neither like nor dislike | Extremely like

7. Overall acceptance

1 2 3 4 5 6 7

Extremely dislike | Neither like nor dislike | Extremely like

Figure B.1: Sensory evaluation form