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FYP JELI

Nutrient analysis and antioxidant activity of dates powder  
(*Phoenix dactylifera*)

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## THESIS DECLARATION


I hereby declare that this thesis entitles “Nutrient analysis and antioxidant activity of dates powder (*Phoenix dactylifera*)” is the results of my own research except as cited in the references. The thesis has not been submitted for a higher degree to any universities or institutions.


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## Nutrient analysis and antioxidant activity of dates powder (*Pheonix dactylifera*)

### ABSTRACT

Dates (*Pheonix dactylifera*) have been known as a fruit that could give energy source to consumers due to their high sugar content especially in fasting month. Turning dates into powder form can help consumer to consume it for a little longer because of the short life of dates and it can be consumed as a sugar substitute. Therefore, the objective was to analyse nutrient content and antioxidant activity of dates powder by performing proximate analysis and determination of antioxidant activity by DPPH assay. The dates were dried by using oven drying method at 60°C temperature for 2 days to completely dry it and blend it to produce dates powder. The nutrient composition of date powder such as ash and moisture content, crude fat, crude protein and total soluble solids was determined and compared with sugarcane as the positive control. Next, the antioxidant activity of date powder was determined by using DPPH (standard 2,2-diphenyl-1-picrylhydrazyl) free radical scavenging assay and also was compared with sugarcane as the control. The nutrient content of date powder obtained were ash content ( $15.84 \pm 5.62\%$ ), moisture content ( $5.12 \pm 0.66\%$ ), crude protein ( $0.52 \pm 0.17\%$ ) and zero crude fat content. The percentage total soluble solids (TSS) of date powder was  $1.2 \pm 0.05$ . Antioxidant activity analysis by using DPPH radical scavenging activity resulted that the date powder has higher antioxidant activity in every concentration (4, 8, 12, 16 and 20 mg/ml) compared to sugarcane sample. The inhibitory concentration 50 (IC<sub>50</sub>) values are a parameter to determine the antioxidant activity of the sample by comparing it with DPPH free radical assay (River-Cruz et al., 2020). The date powder sample has lower IC<sub>50</sub> value which is 4.54 (mg/ml) compared to sugarcane sample which is 48.94 (mg/ml). The lower IC<sub>50</sub> value means the higher antioxidant activity in the sample. In conclusion, date powder has higher nutrient content and antioxidant activity than sugarcane.

**Keywords:** Dates, dates powder, nutrient content, antioxidant activity, DPPH (standard 2,2-diphenyl-1-picrylhydrazyl) assay.

## **Analisis nutrien dan aktiviti antioksidan terhadap serbuk kurma (*Pheonix dactylifera*)**

### **ABSTRAK**

Kurma (*Pheonix dactylifera*) dikenali sebagai buah yang boleh memberikan sumber tenaga kepada pengguna kerana kandungan gulanya yang tinggi terutama sekali di bulan puasa. Menukarkan buah kurma menjadi serbuk dapat membantu pengguna untuk menggunakannya dengan lebih lama kerana jangka masa kurma tersebut sangat singkat dan ianya boleh digunakan sebagai pengganti gula. Oleh itu, objektif kajian ini adalah untuk menganalisis kandungan nutrien dan aktiviti antioksidan terhadap gula serbuk daripada kurma dengan melakukan analisis proksimat dan mengenalpasti aktiviti antioksidan dengan menggunakan ujian DPPH dalam kajian aktiviti ini. Buah kurma telah dikeringkan dengan menggunakan kaedah pengeringan ketuhar pada suhu 60°C selama 2 hari untuk memperoleh buah yang kering sepenuhnya dan kemudian mengisar kurma tersebut untuk menghasilkan gula serbuk. Komposisi nutrient serbuk kurma seperti kandungan abu, kandungan lembapan, lemak kasar, protein kasar dan jumlah pepejal larut telah ditentukan dan dibandingkan gula tebu sebagai sampel kontrol. Seterusnya, aktiviti antioksidan gula serbuk daripada kurma telah ditentukan dengan menggunakan ujian penghapusan radikal bebas DPPH (standard 2,2-diphenyl-1-picrylhydrazyl) dan juga telah dibandingkan dengan gula tebu sebagai sampel kontrol. Kandungan nutrien gula serbuk daripada kurma ialah kandungan abu ( $15.84 \pm 5.62\%$ ), kandungan lembapan ( $5.12 \pm 0.66\%$ ), protein kasar ( $0.52 \pm 0.17\%$ ) dan tiada kandungan lemak kasar. Jumlah peratusan pepejal larut (TSS) yang diperolehi oleh serbuk kurma ialah  $1.2 \pm 0.05$ . Keputusan aktiviti antioksidan menggunakan ujian penghapusan radikal bebas DPPH (standard 2,2-diphenyl-1-picrylhydrazyl) menunjukkan serbuk kurma mempunyai aktiviti antioksidan yang lebih tinggi dalam setiap kepekatan (4, 8, 12, 16 and 20 mg/ml) berbanding dengan sampel gula tebu. Serbuk kurma mempunyai nilai  $IC_{50}$  iaitu 4.54 (mg/ml) berbanding dengan sampel gula tebu iaitu 48.94 (mg/ml). Jika nilai  $IC_{50}$  lebih kurang bermaksa aktiviti antioksidan terhadap sesuatu sampel lebih tinggi. Kesimpulannya, serbuk kurma mengandungi kandungan nutrient dan aktiviti antioksidan yang lebih tinggi berbanding gula tebu.

**Kata kunci:** Kurma, serbuk kurma, kandungan nutrien, aktiviti antioksidan, ujian DPPH (standard 2,2-diphenyl-1-picrylhydrazyl).

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## LIST OF SYMBOLS

%	Percentage
g	Gram
mg	Milligram
°C	Degree Celsius
°Bx	Degree of Brix
% Brix	Sugar Content
mL	Mililiter
mg/mL	Miligram per mililiter
nm	Nanometer
≤	Less than or equal
±	Plus-minus
-	Minus
/	Divide
×	Times
=	Equals

## LIST OF ABBREVIATIONS

IC <sub>50</sub>	Inhibitory Concentration 50
TSS	Total Soluble Solids
DPPH	2,2-diphenyl-1-picrylhydrazyl
ABTS	2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)
FRAP	Ferric Reducing Antioxidant Power

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## CHAPTER 1

### INTRODUCTION

#### 1.1 Research Background

Dates or the scientific name *Pheonix dactylifera* are widely produced in Saudi Arabia, Iraq, Iran and Egypt which are in the regions of hot desert located in Southwest Asia and North Africa (Mohamed & Chang, 2008). Many people have already known about the benefits of dates especially for Muslim. Dates help consumers to stay energized even after fasting for hours. It is because dates are rich in nutrients and antioxidants. Turning dates into powder and identifying nutrient analysis and antioxidant activities has been my interest to study it in more depth. Sugar basically being used in baking and in beverages as a sweetener. Basically, it is made from sugarcane or granulated sugar so this study has determined the dates powder could be as sweet as regular sugar. This study focused on converting the dates into powder because in the previous study, Mohamed and Chang (2008) stated that 70% of the date diets contain carbohydrates and most of it is in

the form of sugar. Dates also consist large amounts of dietary fibre and the minerals found in dates are iron and potassium.

According to Essa et al., (2015) dates fruit contain high phenolics and other antioxidants which is ferulic acid, anthocyanins, caffeic acid and protocatechuic. Antioxidant activities in dates are measured by using DPPH (2,2-diphenyl-1-picrylhydrazyl-hydrate) free radical methods. Dates also contain high sugar content that is suitable to be used as sugar substitute in baking and for drinks. Besides of having high sugar content that could give the same sweet taste as sugar to consumers, the dates powder also provides high nutrition and antioxidant. Nutrient analysis is very important so that a new food product which is powdered sugar from dates is safe to be eaten and commercially viable to the consumers. Antioxidant activities analysis of powdered sugar is to prevent the oxidation of it when being added in foods or in the body at low concentration (Halliwell, 1999).

## **1.2 Problem Statement**

The dates can last between a month or up to 3 months in room temperature depends on different cultivar. The short life of dates can't be kept in longer time. Therefore, by turning dates into powder form can help consumer to consume it for a little longer compare to fresh dates. In the meantime, to help the consumers obtain the same benefits and nutrients of the dates but in the different and more practical way. This is

because the dates powder can be consumed as a sugar substitute in making drinks, baking and others suitable cooks.

### **1.3 Objectives of Study**

1. To develop dates powder.
2. To analyse the nutrient content of dates powder.
3. To determine the antioxidant activity in dates powder.

#### **1.3.1 Research Questions**

1. What was the nutrient content of dates powder?
2. What was the antioxidant activity of dates powder?

### **1.4 Hypothesis of Study**

H<sub>0</sub>: The nutrient content in dates powder had no significantly differences with sugarcane.

H<sub>1</sub>: The nutrient content in dates powder has significantly difference with sugarcane.

H<sub>0</sub>: The antioxidant activity in dates powder had no significant difference with sugarcane.

H<sub>1</sub>: The antioxidant activity in dates powder had significantly difference with sugarcane.

### **1.5 Scope of Study**

This study was focused on the determination of the nutrient content and antioxidant activity of dates powder and compared with sugarcane as a control sample. The dates were converted into powder form and further the nutrient analysis such as ash, moisture, crude fat and crude protein were analysed by using proximate analysis for both samples. Meanwhile, the study also focused on identify the total soluble solids (TSS). Furthermore, the antioxidant activity of date powder and sugarcane was determined using DPPH free radical scavenging assay.

### **1.6 Significance of Study**

From this research project, the creation of dates powder as sugar substitute may offer variety of benefits to the food industry and it also useful for daily basis usage such as for baking or beverages since it can be produce by homemade. There is a potential of dates powder as the new food products since using it as sugar substitute is more beneficial



compare to sugarcane which contain less nutrient than date. Other than that, the dates can be kept in longer time if it was turned into powder form and consumer can use that in more practical way for many cooks.



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## CHAPTER 2

### LITERATURE REVIEW

#### 2.1 Date Fruits (*Phoenix dactylifera*)

Date fruits (*Phoenix dactylifera*) are mostly produced or imported from Arabic country. The first or second largest country that produce dates is Iran (Sahari et al., 2017). In the Middle East, dates have been important food crop in the last 6000 years. Dates also have a wide variety and have been one of the mankind's oldest cultivated plants. The varieties of dates can be detected by their different texture, colour, and flavour (Ghnimi, 2016).

The reason for using dates into being producing into powder as a study because from the previous study by Sahari et al., (2007), they found out that the mean percentages of moisture, protein, lipid, and ash of dates were 29.35, 3.3, 0.42 and 2.25 g/100 g (fresh weight basis), respectively. At the same time, the predominant sugars were fructose (12.62–43.31 g/100 g) and glucose (16.41–54.23 g/100 g) on fresh weight basis.

The high content of sugar and nutritional value discovered by previous researcher have convinced me to investigate either the powdered sugar of dates could be as a sweetener like regular sugar and the nutritional value are the same or not. Furthermore, there were many researchers reported that dates contain bunch of benefits to humans such as high in antioxidant, anticancer, anti-inflammatory, gastroprotective and others.

## 2.2 Maturity Stage of Dates

Meanwhile, Zhen et al., (2013) and Siddiq et al., (2013) stated that the dates have maturity stages which called as *Hababouk*, *Kimri*, *Khalal*, *Rutab* and *Tamr* as shown in Figure 2.2 below. The five stages maturity of dates take six to eight months for the development progress. The development progress of dates are (1) rapidly grow, (2) the characteristics colour is turning, (3) losing water, (4) accumulating sugars, and (5) completely ripen.

The first stage of maturity of dates is called as *Hababouk* which appears after pollination for four until five weeks. The shape of the fruit is round with a colour of whitish-cream and green stripes. The next stage is called *Kimri* and it starts to appear 17 weeks after pollination. The characteristics of fruit in this stage is still young, greenish, elongated, hard-textured and contains 85% of moisture. The weight of the fruit and the concentration of tannin are increasing but it still inedible for direct consuming. In *Khalal* stage, six weeks after *Kimri* stage, the dates turn into maximum size, weight, and hard

texture. The colour of fruit turns into yellow, purplish-pink, or red and it depends on the cultivar. The sugar slowly increases and turns into sucrose.

The next four weeks after *Khalal* stage, dates in *Rutab* stage starts to lose water and half of the dates become soften, sweeter and darker in colour i.e light brown. The sucrose content has been converted to reducing sugars. Then the percentage of protein, fat and ash in the fruit is decreased. The ripening condition has begun in this stage. In the final stage which is *Tamr* or *Tamar/Tamer* usually only lasts for two weeks. The dates become maximum in total solids, have highest sweetness, are dark brown in colour, have a soft texture and their shape becomes wrinkled. *Tamr* stage has a high concentration in reducing sugar such as fructose and glucose. Meanwhile, the percentages of protein, fat and ash is lesser than in *Rutab* stage. However, *Tamr* stage dates have good storage capacity due to low moisture and high sugar content.

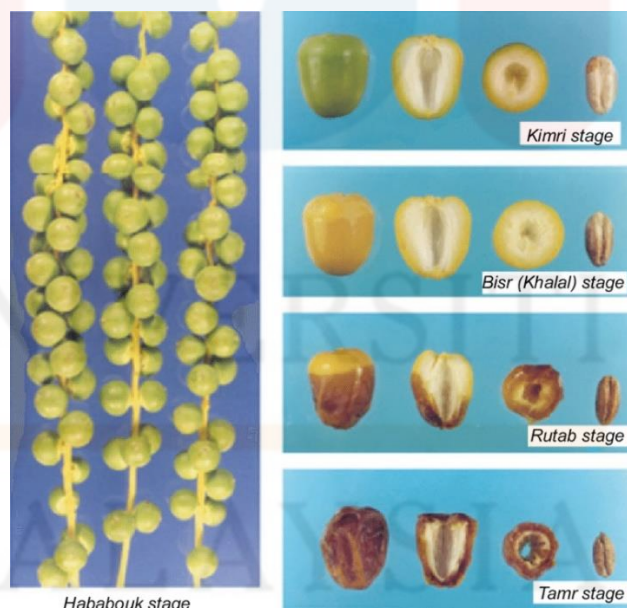


Figure 2.2: The different maturity stage of dates. (Source: Siddiq et al., 2013)

### 2.3 Sugar

Sugar is one of the important ingredients for Malaysians in making food and beverage such as in cooking, baking, making drinks and so on. Usually, Malaysian will only choose either regular sugar or brown sugar as a sweetener in their daily diet. This is because sugar is usually used as a sweetening agent, viscosity-enhancing agent, preservative and also for other usage in food and beverages. Sugar has also been designated as “generally recognized as safe (GRAS) by the U.S. Food and Drug Administration (FDA).

Basically, Malaysians only use regular sugar and brown sugar derived from sugarcane in their food and beverages. We are also aware that if the intake of regular sugar is high, it can lead to obesity among consumers. A study by Larsson et al., (2006) stated that high content of sugar could gives high risk of pancreatic cancer. There are reasons why consumers need to take high content of sugar in their food and beverages. For example, when we were making a pot of tea (let’s say a pot for 5 cups of drink) the volume of sugar needed is four to five tablespoons to make the tea taste sweet. If less spoons are used the tea will taste bland. This is the reasons whys consumers need to consume high content of regular sugar. So, from that situation, it led my study to investigate whether the sweetness of powdered sugar from dates could makes consumer to use less spoon of sugar while satisfying the sweetness in their foods and beverages.

Simple sugars also known as monosaccharides was consists of glucose, fructose and galactose. These simple sugar have six carbon atoms and same molecular formula

( $C_6H_{12}O_6$ ) but they have different characteristics and properties. Sucrose, lactose and maltose belongs to disaccharides. Sucrose ( $C_{12}H_{22}O_{11}$ ) also known as table sugar was obtained from plants (such as sugarcane and sugar beet) industrially. The sucrose was extracted by warm water from the crushed cane in sugar mills to produce sugarcane. The juice obtained was undergo purified and clarified with heat and lime. The precipitate was removed on centrifuges. In the evaporation process, the concentration of the juice in evaporator and the other process was the crystallization in vacuum pans. The syrup was clarified with the addition of lime, phosphoric acid and polymer flocculent. The syrup was filtered and go to vacuum pans for crystallization process.

#### **2.4 Nutritional Value of Dates**

From the previous study, they have found out the mean percent of moisture, protein, lipid and ash of dates were 29.35, 3.3, 0.42 and 2.25 g/100 g (fresh weight basis), respectively (Sahari et al., 2007). Then, the predominant sugars of it were fructose (12.62–43.31 g/100 g) and glucose (16.41–54.23 g/100 g, fresh weight basis) (Sahari et al., 2007). Other than that, Biglari et al., (2008) also stated that the dates have high percentage in carbohydrates (total sugars, 44-88 %), fat (0.2-0.5 %), protein (2.3-5.6 %), 15 kinds of salts and minerals, vitamins, and a high percentage of dietary fibre (6.4-11.5 %). Meanwhile, several studies found out that sugar content in every kind of dates could be different because of the carbohydrate concentration possibly affected by the date cultivars, harvest, and postharvest factors. The growth environment such as fertilizer,

growth temperature and humidity could be one of the factors that attributed the carbohydrate concentration in dates and affect the sugar content.

## **2.5 Proximate Analysis**

Proximate analysis is used to identify the ash and moisture content, crude proteins, crude fats, crude fibre, and carbohydrate content of the sample (Sarah et al., 2015).

The purpose of identifying ash content on food sample is it can assist as an index to the nutritive value of that particular food (Suliman et al., 2012). The ash content also can define the total mineral content in food sample (Nielsen, 2017). If that particular food sample contain high in mineral content then this ash content analysis is important to identify their nutritional, toxicological and food quality. There are two types of ashing that usually being applied to identify the ash content which are dry ashing and wet ashing. In dry ashing, the temperature used to heat the food sample by convectional or microwave heating is 500-600°C while wet ashing used lower temperature compared to dry ashing but it depends on strong acids and chemical oxidizers.

Moisture content in food sample is important in order to preserve the food from spoile (Agboola and Adejump, 2015). In addition, if the food sample contain less moisture it will lead to longer shelf life of the product (Assirey, 2015). The moisture loss was observed when the water evaporates from the sample at higher temperature. According to Hackney et al., (2019) the nutritional analysis of protein in food sample also important since it has been known as the nutritional factor which can decrease and inhibit the loss of strength and mass in the muscle and as energy provider. The crude protein in food

sample can be determine by using Kjeldahl method to get the total nitrogen content in particular sample. Finally, the crude fat in food sample is important since it consider as a major involve in food industry as food labelling regulations (Snow et al., 1997).

## **2.6 Total Soluble Solids (TSS)**

The total soluble solids (TSS) were determined to identify the level of sweetness in fruit sample solution (Kusumiyati et al., 2020). This total soluble solid content can be defined by measuring the index of refraction by refractometer apparatus. The index used in refractometer was related to as degrees of Brix ( $^{\circ}\text{Bx}$ ). The degree Brix can indicate the sugar content of a food sample in grams which is sucrose in an aqueous solution. The refractive index was converted the raw Brix scale measurement into a weight percentage of sucrose content which is displayed as % Brix by refractometer. The TSS value was expressed by % Brix after dropped the solution on the detector of refractometer. The value shows up based on ratio of the speed of light in vacuum and speed of the light go through the sample (Kusumiyati et al., 2020).



## 2.7 Antioxidant Activity of Dates

Naturally, antioxidant activity is higher in plants. In dates, the antioxidant compound that are potentially to be found according to the previous study by F. Biglari et al., (2008) such as phenolics and flavonoids.

The researcher also revealed that the phenolics and flavonoids in dates increased due to the longer storage period of the fruit. Therefore, this is the reason why in this research it is necessary to include the findings of antioxidant activity because it is necessary to discover whether the dates after being converted into date powder have the same antioxidant activity as the dates fruit or not. There was research stated the antioxidant activity is important to prevent the oxidation of the substrate when it presents in foods or in the body at low concentration (Moon and Shibamoto, 2009).

The method used to determine the antioxidant activity in this study was by using DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate) free radical method. DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate) free radical method is an antioxidant assay predicted on electron-transfer that produces a violet solution in methanol (Huang et al., 2005). At room temperature, the free radical is stable and it reduces when the presence of an antioxidant molecule appeared and then it caused colourless to the methanol solution. The most rapid and easy way to estimate antioxidant by spectrophotometry is by using this DPPH assay (Kedare & Singh, 2011).

Mechanism in DPPH antioxidant assay is based on the measurement of the scavenging capacity of antioxidant towards it. DPPH is a stable free radical by virtue of

the delocalisation of the spare electron over the molecule so that the molecules do not dimerise. The delocalisation will cause the deep violet colour when the absorption with ethanol or methanol solution at around 517 nm. When DPPH solution with a substance that can donate a hydrogen atom on mixing, it will cause to the reduced form with the loss of violet colour. The DPPH radical and hydrogen atom reduced from the substance then free radical produced and it will undergo further reactions.

## CHAPTER 3

### METHODOLOGY

#### 3.1 Materials and Apparatus

##### 3.1.1 Chemical and Reagents

Sulphuric acid. Sodium hydroxide. Boric acid. Kjeltabs Cu/3,5. 3.7% Hydrochloric acid from HmbG Chemicals, petroleum ether from R&M Chemicals, Methanol was purchased from Merck. 2,2-diphenyl-1-picrylhydrazyl (DPPH)

##### 3.1.2 Apparatus

Muffle furnace PROTHERM, crucible with lid used for ash content analysis. 105°C oven drying. Blender. Moisture analyzer (AND MX-50).

Kjedahl digestion machine Vapodest C. Gerhardt and Foss Kjelttec 8200 Auto Distillation. Soxtec Foss 2055 and dessicator. Apparatus for antioxidant activity analysis were spectrophotometer from Biochrom, shaking waterbath from MEMMERT and centrifuge 5810 R from Eppendorf. ATAGO Digital Hand-held "Pocket" Refractometer PAL- $\alpha$ .

## **3.2 Methods**

### **3.2.1 Sample Preparation**

The dates (*P. dactylifera*) (kurma tangkai Utika Azewa, Tunisia) and sugarcane (CSR, Selangor) from Pantai Timur Tanah Merah. The treatment process was started with cleaning the fruits under running tap water to discharge any dirt on it. After that, the excess water on the fruit was removed by wiping the fruit on tissue paper to keep the samples dry. Then, the samples were deseed and chopped with knife into smaller pieces and the sample was weighed and recorded. Lastly, the sample was put in the A3 size zip lock plastic bag to avoid with any environment contamination before undergo drying treatment.

### **3.2.2 Oven Drying Method**

The fresh date was heated in oven drying and the temperature used was 60°C (Manaa et al., 2013). The previous studies reported that the time taken to dry the dates is in 7 to 10 hours (Manaa et al., 2013) and 18 hours in oven dryer to obtain date powder (Saikiran et al., 2018) but the fresh date sample was heated for 2 days to completely dry it.

### **3.2.3 Blend Dried Dates into Powder**

The dried dates was grinded using blender and the powdered dates was placed in the A3 size zip lock plastic bag to secure it for further treatment which was nutrient analysis and antioxidant activity.

### **3.2.4 Determination of Nutrient Content**

Proximate analysis used to analyse nutrient content were ash, moisture content, crude protein, crude fat, and total soluble solids in dates powder and sugarcane as the control.

### 3.2.4.1 Ash Content

Dry ashing method was applied to determine the ash content of the date powder and sugarcane. The dry ashing is the standard method to determine the ash content in food sample which at 550-600°C of temperature to completely oxidized the organic materials in th sample without burning it. By following Nollet (2004) the date powder and sugarcane sample was placed in muffle furnace at 550°C for 12 hours.

% Ash:

$$\frac{w_2 - w_1}{w_s} \times 100$$

where;

$W_1$  = weight of crucible (g)

$W_2$  = weight of crucible + ash (g)

$W_s$  = weight of sample (g)

### 3.2.4.2 Moisture Content

The date powder and sugarcane sample was weighed in the provided pan and then analysed the moisture by using moisture analyzer (AND MX-50) by following the operating procedure. The moisture analyzer was showed the reading of percentage of moisture automatically.

### 3.2.4.3 Crude Protein

The protein content in foods was determined according to the total of nitrogen content. The nitrogen content was determined by using the classical Kjeldahl method. The analysis consists of three steps which is digestion, distillation and titration step. According to Mæhre et al. (2018), the Kjeldahl method was performed by hydrolyzed 1 gram of both sample with 12 mL concentrated sulfuric acid ( $H_2SO_4$ ) and two tablets of kjeltabs Cu/3,5 (potassium sulfate,  $K_2SO_4$ ) then heated in Kjeldahl digestion machine Vapodest C. Gerhardt and Foss Kjeltac 8200 Auto Distillation at  $400^\circ C$  for 1 hours. In digestion step, the polypeptides bond were break down and converted into simpler chemicals (such as ammonia, carbon dioxide and water) by catalyst kjeltabs Cu/3,5 (potassium sulfate,  $K_2SO_4$ ) to speed up the reaction. The ammonia was separated from the digestion mixture in distillation step. Then, in titration step, the ammonia trapped was determined by titration with a standard solution and the amount of total nitrogen multiplied with conversion factor of 6.25 to identify the total of crude protein content. You don't mention here about the equation below.

%Kjedahl Nitrogen:

$$\frac{(V_s - V_b) \times N \times 14.01}{w \times 10}$$

where;

$V_s$  = standardized hydrochloric acid used to titrate a sample (mL)

$V_b$  = standardized hydrochloric acid used to titrate a reagent blank (mL)

$N$  = normality of standard (0.1)

14.01 = atomic weight of nitrogen

$W$  = weight, in grams, of the sample or standard

10 = factor to convert mg/gram to percent

% Crude protein:

$$\% \text{ Kjeldahl Nitrogen} \times F$$

$F$  = factor to convert nitrogen to protein; 6.25 (Phillips et al., 2021)

#### 3.2.4.4 Crude Fat

The crude fat was determined by using Soxhlet extraction method (Holman et al., 2019). The crude fat of each sample was extracted by Soctex™ FOSS 2055. The aluminium cup was heated in 105°C oven drying for 30 minutes and cool it down in the desiccator for 20 minutes. Then, 1 gram of both samples was placed and prepared in the thimble and placed 80 mL petroleum ether in the



aluminium cup. Placed thimbles and aluminium cups in the machine and run the analysis. After finish steps in the extraction machine, the aluminium cup was heated again in 105°C oven drying for 30 minutes and cool it down in the desiccator for 20 minutes. Finally, was weighed the aluminium cup. The crude fat content was measured by followed the equation below.

% Crude fat:

$$\frac{R_2 - R_1}{w} \times 100$$

R<sub>1</sub> = Empty thimble (g)

R<sub>2</sub> = Thimble with sample (g)

W = Weight of sample (g)

#### 3.2.4.5 Total Soluble Solids (TSS)

The total soluble solids for both date powder and sugarcane was determined by using brix analysis. 1 gram of both date powder and sugarcane sample was diluted in 100 ml of distilled water. The concentration of both solution is 10 mg/ml. Then, the degree of Brix was measured by ATAGO Digital Hand-held "Pocket" Refractometer PAL- $\alpha$ .

### 3.2.5 Determination of Antioxidant Activity by DPPH

DPPH (standard 2,2-diphenyl-1-picrylhydrazyl) act as free radical scavengers or hydrogen atoms. The DPPH solution was prepared by dissolve 0.004 g DPPH in 100 mL of methanol (Bhuiyan et al., 2009). The stock solution for both samples was extracted by added 1 gram of sample into 25 mL methanol and placed in shaking waterbath at room temperature for 2 hours and 30 minutes After that, 3 ml of freshly prepared DPPH methanol solution (0.004%) were added to all stock solution sample with different concentrations (4, 8, 12, 16 and 20 mg/ml) then being incubated in the dark for 30 minutes. The absorbance was measured at 517 nm by using spectrophotometer.

% Scavenging of the DPPH free radical formulation:

% Inhibition:

$$\frac{A_0 - A_1}{A_0} \times 100$$

where;

$A_0$  = absorbance control

$A_1$  = absorbance sample

The IC<sub>50</sub> values was a parameter to determine the antioxidant activity of the sample by compared it with DPPH free radical assay (River-Cruz et al., 2020). IC<sub>50</sub> value was measured from % Inhibition vs. Concentration graphs by using linear regression analysis.

### **3.3 Statistical Analysis**

The data analysis was expressed as mean  $\pm$  standard deviation of 3 independent results (n=3). Statistical analyses were determined by T-Test (Two-tailed) and One-Way Analysis of Variance (ANOVA). The significant for nutrient analysis and antioxidant activity of powdered sugar from dates compared to sugarcane was considered at 5% significant level ( $p \leq 0.05$ ) by using Statistical Package for Social Science, IBM SPSS Statistical 26.

## CHAPTER 4

### RESULT AND DISCUSSION

#### 4.1 Nutrient Content of Date Powder

The analysis of nutrient content of produced date powder was performed by using proximate analysis methods. In this proximate analysis, the process was focused on to determine ash, moisture content, crude protein and crude fat of date powder by comparing with control sample which is sugarcane.

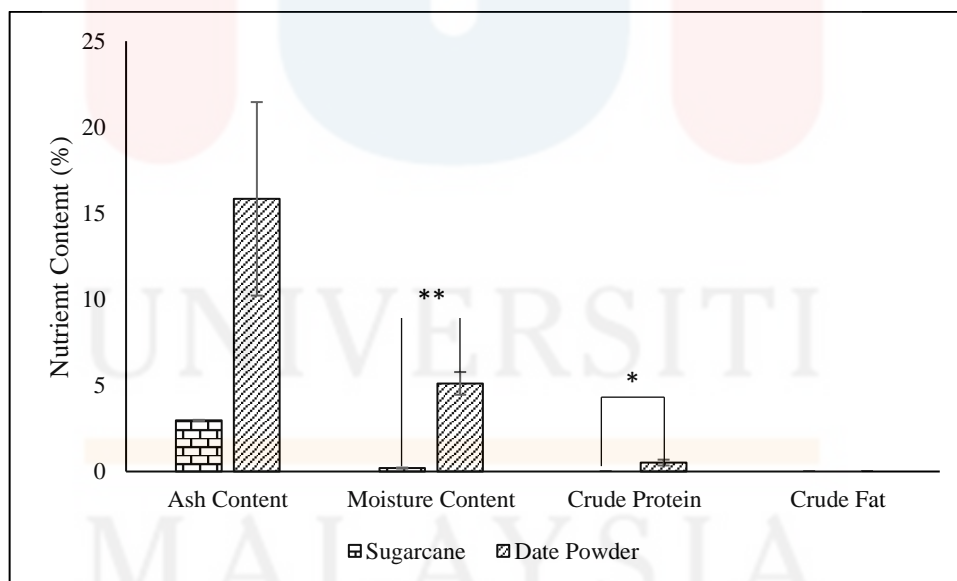
The result in the Figure 4.1 shows the percentage of proximate analysis of sugarcane and date powder in ash, moisture content, crude protein, and crude fat. From the figure, it shows that the percentage of ash content in date powder recorded a value of  $15.84 \pm 5.62$  which is higher than the ash content in sugarcane with a value of  $2.97 \pm 0.03$ . There was no significant difference ( $p \leq 0.05$ ) in terms of ash content between the

sugarcane and date powder. The ash content is very important in food nutrient analysis it is because the total mineral content in date powder can be analysed by amount of ash content.

The moisture content in Figure 4.1 shows that the date powder has higher moisture content than sugarcane with the value of  $5.12 \pm 0.66$  and  $0.20 \pm 0.03$ , respectively. Then, there was significant difference ( $p \leq 0.05$ ) of moisture content between the sugarcane and date powder. According to Assirey (2015), if the food sample contains a low moisture content, then it tends to have a longer shelf life. However, since the date powder contains higher moisture level than sugarcane, thus it also tends to have lesser shelf life compared to sugarcane. The higher moisture level might easily expose the date powder to microbial spoilage and according to Agboola and Adejumo (2015) will affect the consumers' preferences to consume the food product.

The crude protein represents the amount of protein in a food sample and its content is very important to evaluate the economic value of a product (Miao, 2011). The protein content in food functions to slow down and prevent the loss of strength and mass in muscle (Hackney et al., 2019). From Table 4.1, the result shows that crude protein of date powder with the value of  $0.52 \pm 0.17$  recorded the significant higher ( $p \leq 0.05$ ) compared to crude protein in sugarcane. However, according to Agboola and Adejumo (2015), the dates were not reported to be a good protein source. It might be caused by low crude protein analysed during the analysis. Meanwhile there is no result for crude protein of sugarcane detected. According to the nutritional fact of the control (CSR sugar product) there was no protein content showed on the packaging. The Picture 4.1 shows the nutrient content on the packaging of the sugar.

The crude fat was not detected in both sugarcane and date powder. The result of cup weight before and after analysis showed no change. It may be due to no crude fat content in both control and food samples. Thus, no result can be shown in terms of mean and standard deviation therefore no significant value either. Suleiman et al. (2011) and Sarah et al. (2015) has stated that most of varieties of dates were classified as fruit that have low fat content. In fact, the date powder is suitable to be consumed as a substitute to the sugarcane in daily basis since the fat content is zero. It proved that low fat content in food is good for consumer health because according to Bhandari and Sapra (2021), the lowered fat content in food could decrease the risk of cardiovascular diseases such as heart attack and stroke.



All values are mean $\pm$ SD ( $n=3$ ); Significant difference at  $*p<0.05$  and  $***p<0.01$ , respectively

Figure 4.1: The percentage of nutrient content of sugarcane and date powder by proximate analysis (%).



Picture 4.1: The nutrient content of the CSR sugar.

#### 4.2 Total Soluble Solids (TSS) of Date Powder

The total soluble solid of date powder and sugarcane was performed by using Brix analysis. The degree of Brix have been measure by using ATAGO Digital Hand-held "Pocket" Refractometer PAL- $\alpha$ . The concentration (mg/ml) involved in the analysis is by 1 gram of each sample was diluted in 100 ml of distilled water, so the concentration of the solution is 10 mg/ml.

Table 4.2 shows the total soluble solids of both food samples by using brix analysis. The result stated that the total soluble solids of sugarcane is higher compared to the total soluble solids of date powder, which is  $1.3 \pm 0.26$  and  $1.2 \pm 0.05$  respectively. There was no significant difference ( $p \leq 0.05$ ) between total soluble solids of sugarcane and date powder. Then, the meaning of degree brix ( $^{\circ}\text{Bx}$ ) is the scale that represents the sugar content in an aqueous solution therefore from the result on Table 4.2 above shows that the percentage of Brix of the date powder has slightly contain less sugar content compared to the sugarcane. In that case, the sweetness of the date powder couldn't be compared same as the sweetness of the sugarcane.



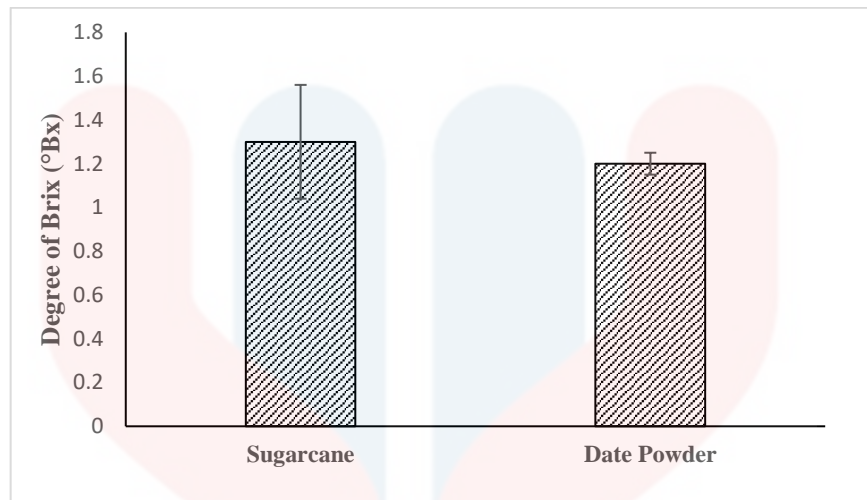


Figure 4.2: The percentage of degree of Brix (°Bx) of sugarcane and date powder.

### 4.3 Antioxidant Activity of Date Powder

The antioxidant activities of date powder were performed by using DPPH radical scavenging activity. The DPPH assay free radical is a method that can predict antioxidant assay on electron-transfer that produce a violet solution in methanol (Huang et al., 2005). The antioxidant molecules appear after the methanol solution and violet solution from DPPH assay become colourless due to the free radical in the solution become stable and reducing.

Figure 4.3 shows the percentage of radical scavenging activity of sugarcane and date powder at different concentration which are 4, 8, 12, 16 and 20 mg/ml. The result determined that the DPPH free radical scavenging activity of both sugarcane and date powder increased with increasing concentration of the samples. In the meantime, the date powder defined as higher antioxidant activity in every concentration compared to sugarcane sample. The trend of the DPPH free radical scavenging activity in sugarcane sample shows that the percentage is increasing but slightly different in every concentration. There was significant difference ( $p \leq 0.05$ ) between different concentration of sugarcane sample. Meanwhile, for date powder sample it shows that the DPPH free radical scavenging activity increasing but the trend is highly different at every concentration. There was significant difference ( $p \leq 0.05$ ) between different concentration of date powder sample. The date powder defined as highest value of free radical scavenging inhibition at 20 mg/ml concentration which is  $95.87 \pm 1.07$  meanwhile the

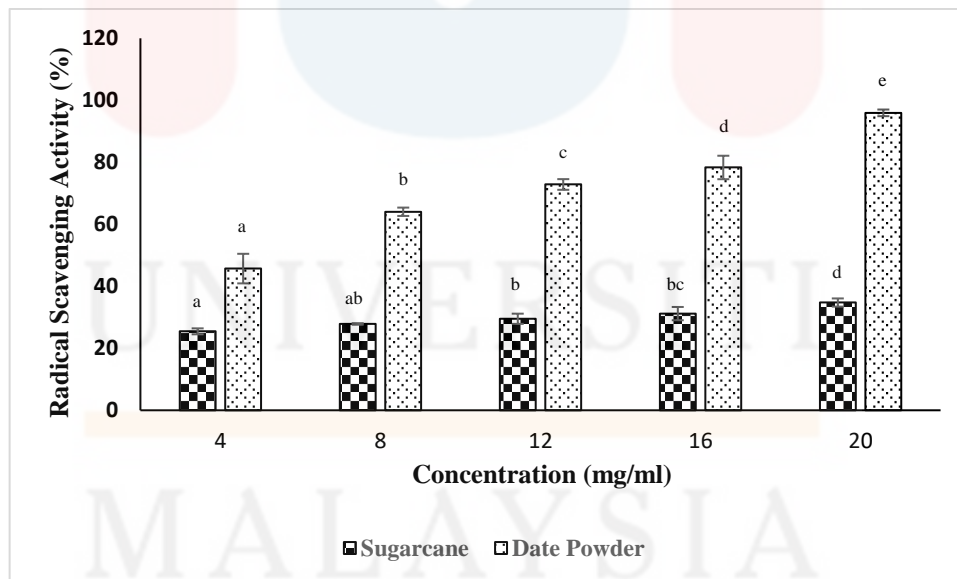
sugarcane sample radical scavenging activity value at 20 mg/ml is  $34.76 \pm 1.32$ . Hence, there was significant difference ( $p \leq 0.05$ ) of free radical scavenging inhibition at different concentration 4, 8, 12, 16 and 20 mg/ml between sugarcane and date powder sample.

The  $IC_{50}$  values is a parameter to determine the antioxidant activity of the sample by comparing it with DPPH free radical assay (River-Cruz et al., 2020). The concentration of antioxidants needs to decrease the initial DPPH concentration by 50% in order to calculate the value of  $IC_{50}$ . According to River-Cruz et al. (2020), if the value of  $IC_{50}$  is low meaning that the antioxidant activity in that particular sample is higher. The 50 percent of scavenging activity of sugarcane and date powder sample was determined from the curves plotted (Refer to Appendix A) and then the  $IC_{50}$  value obtained by the linear regression equations (Refer to Appendix A).

The result in Table 4.3 shows that the date powder sample has lower  $IC_{50}$  value which is 4.54 (mg/ml) compared to sugarcane sample which is 48.94 (mg/ml). In consideration of the fact that the low  $IC_{50}$  value has higher antioxidant activity, therefore from the result above shows that date powder sample has higher antioxidant activity compared to the sugarcane. In addition, the preparation as low as 4.54 mg/ml concentration in date powdered sample could cause 50 percent of scavenging activity.

Antioxidant was known as a molecule that capable to prevent or slow the oxidation of another molecule (Moon and Shibamoto, 2009; İlhami Gülçin, 2012). Higher antioxidant activity was important for consumer to inhibit the oxidation of other

molecules even at a low concentration. It is because the oxidation can cause variety diseases for consumer such as cancer, aging, arthritis, diabetes and others (Moon and Shibamoto, 2009). In fact, a study by Clarke and Armitage (2002) also stated that the dietary substances with antioxidant activity plays an important function to prevent a few illnesses by defend human body from any free radicals. Other than that, antioxidant also important in terms of food. Since the role of antioxidant is to prevent or delay the oxidation of substrate so it usually being added into food product to inhibit the radical chain reactions, delay rancidity development and flavour deterioration caused by oxidation in food (İlhami Gülçin, 2012). Furthermore, antioxidant properties also can scavenge free radicals in food and increase its shelf life during processing and storage (İlhami Gülçin, 2012).



\*a-e All values are mean $\pm$ SD ( $n=3$ ), Different letters in same row indicates significant differences at  $p\leq 0.05$

Figure 4.3: Percentage of scavenging of sugarcane and date powder at different concentration (mg/ml)

Table 4.3: Percentage inhibition and inhibitory concentration 50 (IC<sub>50</sub>) value of Sugarcane and Date Powder at different concentration (mg/ml)

Treatment	IC <sub>50</sub> Value (mg/ml)
Sugarcane	48.94
Date Powder	4.54

## CHAPTER 5

### CONCLUSION

#### 5.1 Conclusion

In conclusion, the dates can be turned into powder form by using oven drying to dry the fruit without losing its nutrients. In fact, the nutritional value that was determined using proximate analysis showed that the nutritional value in terms of ash and moisture content, and crude protein of the date powder was higher than that of sugarcane (the control). Meanwhile, the crude fat analysis for both date powder and sugarcane identified that both samples contained zero fat content. The total soluble solids (TSS) were determined to represent the sugar content (sucrose) of both date powder and sugarcane by diluting in 100 ml of distilled water. The % Brix showed that the sugarcane had higher sugar content compared than date powder. Antioxidant was known as a molecule that capable to prevent or slow the oxidation of another molecule (Moon and Shibamoto, 2009; İlhami Gülçin, 2012). Higher antioxidant activity was important for consumer to

inhibit the oxidation of other molecules even at a low concentration. Analysis of antioxidant activity using DPPH radical scavenging activity resulted in date powder having higher antioxidant activity in each concentration compared to sugarcane sample. Since that the date powder contains higher antioxidant activity then the IC<sub>50</sub> value of the sample resulting was lower than that of sugarcane.

## **5.2 Recommendation**

The date powder has high nutritional value and antioxidant activity. Therefore, the production of date powder should be conducted by using freeze drying to dry the dates instead of oven drying. Comparison in terms of the nutritional properties and antioxidant activity between date powder from freeze drying and oven drying method should also be conducted. The purpose is to discover whether different drying methods of dates would affect the nutritional properties and content of antioxidant activity. The antioxidant activity of date powder can be analysed by different method such as using 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) assay and Ferric Reducing Antioxidant Power (FRAP) assay method to find consistency in determining the antioxidant properties of the food sample produced.

## **5.3 Limitation of the study**

Due to the unavailable spray dryer machine for drying dates, the drying procedure of date has been conducted by using oven drying which is took longer time to prepare the food sample. The freeze-drying method also need more time compared to oven drying to dry the dates.

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## APPENDIX A

### LINEAR REGRESSION EQUATION TO DETERMINE IC<sub>50</sub>

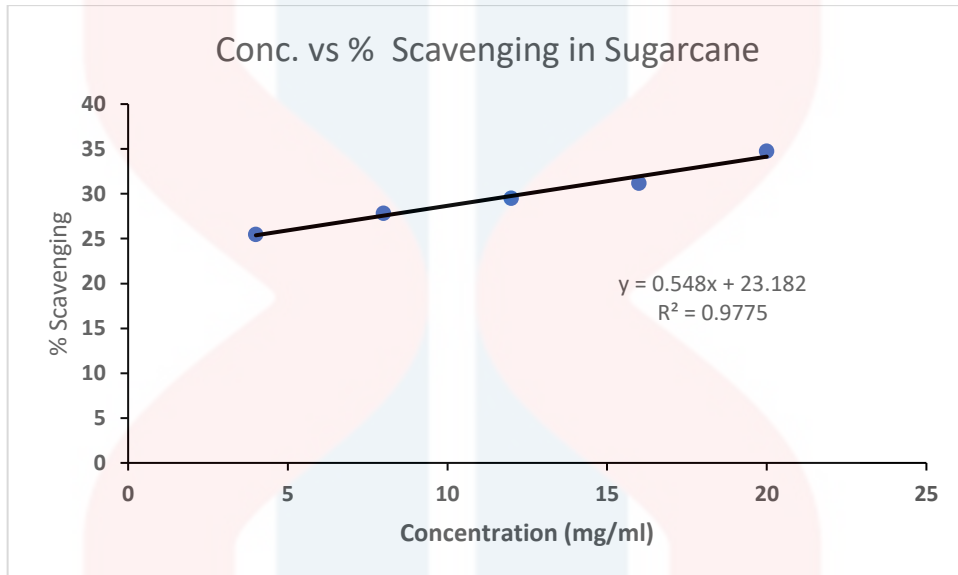


Figure A. 1: IC<sub>50</sub> for Sugarcane

$$y = ax + b$$

$$y = 50$$

$$y = 0.548x + 23.182$$

$$x = \text{IC}_{50}$$

$$\text{IC}_{50} = (50 - b/a)$$

$$= (50 - 23.182)/0.548$$

$$= 48.94 \text{ mg/ml}$$

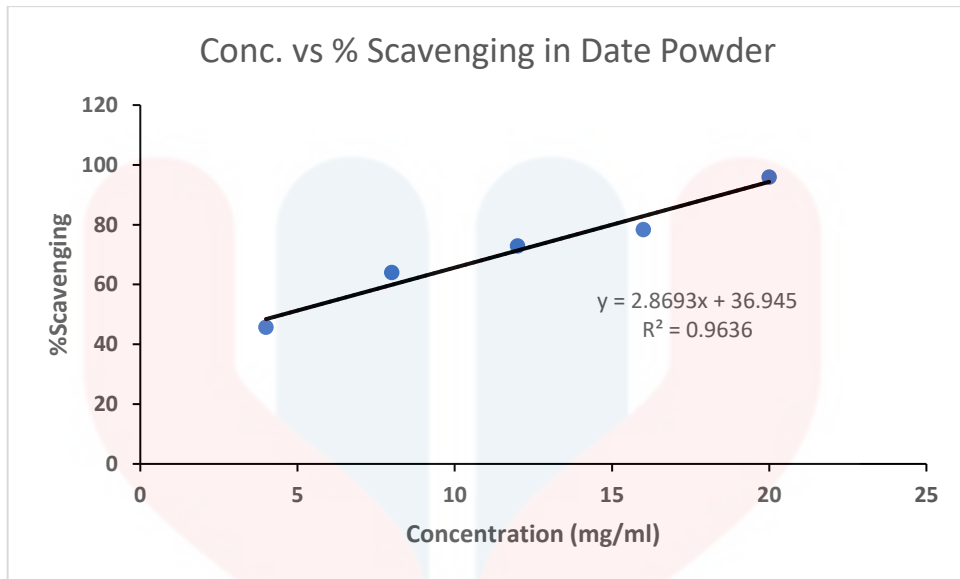


Figure A. 2: IC<sub>50</sub> for Date Powder

$$y = ax + b$$

$$y = 50$$

$$y = 2.8693x + 36.945$$

$$x = IC_{50}$$

$$IC_{50} = (50 - b/a)$$

$$= (50 - 36.945)/2.8693$$

$$= 4.54 \text{ mg/ml}$$

## APPENDIX B

Table B.1: Statistical analysis of percentage of ash content in sugarcane and date powder

### T-TEST

Ash Content

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Sugarcane	2.9700	3	.03000	.01732
	Date Powder	15.8433	3	5.62616	3.24826

Ash Content

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Sugarcane & Date Powder	3	-.988	.098

Ash Content

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Sugarcane – Date Powder	-12.87333	5.65581	3.26538	-26.92313	1.17647	-3.942	2	.059

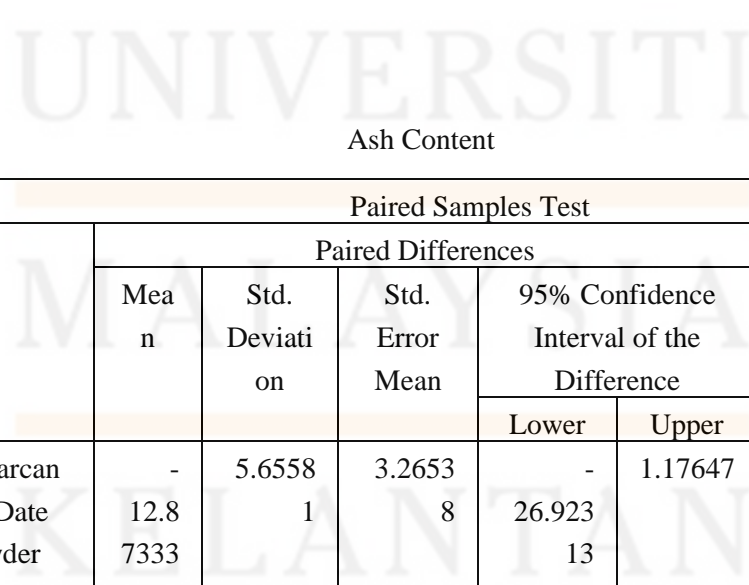


Table B.2: Statistical analysis of percentage of moisture content in sugarcane and date powder

**T-TEST**

Moisture Content

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Sugarcane	.2067	3	.03786	.02186
	Date Powder	5.1200	3	.66910	.38631

Moisture Content

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Sugarcane & Date powder	3	.456	.699

Moisture Content

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Sugarcane - Date Powder	-4.91333	.65271	.37684	-6.53476	-3.29191	-13.038	2	.006



Table B.3: Statistical analysis of percentage of crude protein in sugarcane and date powder

**T-TEST**

Crude Protein

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Sugarcane	.0000	3	.00000	.00000
	Date Powder	.5233	3	.17502	.10105

Crude Protein

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Sugarcane & Date Powder	3	.907	.006

Crude Protein

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Sugarcane - Date Powder	-.51667	.17559	.10138	-.95287	-.08047	-5.096	2	.036

Table B.4: Statistical analysis of percentage of crude fat in sugarcane and date powder

**T-TEST**

Crude Fat

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Sugarcane	.0000 <sup>a</sup>	3	.00000	.00000
	Date Powder	.0000 <sup>a</sup>	3	.00000	.00000
a. The correlation and t cannot be computed because the standard error of the difference is 0.					

Table B.5: Statistical analysis of percentage of total soluble solids (TSS) in sugarcane and date powder

**T-TEST**

Total Soluble Solids (TSS)

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Sugarcane	1.3000	3	.26458	.15275
	Date Powder	1.2333	3	.05774	.03333

Total Soluble Solids (TSS)

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Sugarcane & Date Powder	3	.655	.546

Total Soluble Solids (TSS)

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Sugarcane – Date Powder	.06667	.23094	.13333	-.50702	.64035	.500	2	.667

Table B. 6: Statistical analysis of percentage of radical scavenging activity in sugarcane and date powder

**T-TEST**

Radical Scavenging Activity

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Sugarcane	29.7580	5	3.50552	1.56772
	Date Powder	71.3760	5	18.48630	8.26732

Radical Scavenging Activity

Paired Samples Correlations				
		N	Correlation	Sig.
Pair 1	Sugarcane & Date Powder	5	.990	.001

Radical Scavenging Activity

Paired Samples Test									
		Paired Differences					t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
					Lower	Upper			
Pair 1	Sugarcane – Date Powder	-41.61800	15.02472	6.71926	-60.27365	-22.96235	-6.194	4	.003

Table B. 7: Statistical analysis of percentage of radical scavenging activity in sugarcane (4, 8, 12, 16 and 20 mg/ml)

**ANOVA**

Radical Scavenging Activity

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	149.948	4	37.487	18.586	.000
Within Groups	20.169	10	2.017		
Total	170.117	14			

**Radical Scavenging Activity**

Duncan<sup>a</sup>

Concentration	N	Subset for alpha = 0.05			
		1	2	3	4
4 mg/ml	3	25.3200			
8 mg/ml	3	27.8600	27.8600		
12 mg/ml	3		29.5233	29.5233	
16 mg/ml	3			31.0333	
20 mg/ml	3				34.7600
Sig.		.053	.182	.222	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Table B. 8: Statistical analysis of percentage of radical scavenging activity in date powder (4, 8, 12, 16 and 20 mg/ml)

**ANOVA**

**Radical Scavenging Activity**

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	4112.174	4	1028.043	119.167	.000
Within Groups	86.269	10	8.627		
Total	4198.443	14			

**Radical Scavenging Activity**

Duncan<sup>a</sup>

Concentration	N	Subset for alpha = 0.05				
		1	2	3	4	5
4 mg/ml	3	45.5567				
8 mg/ml	3		63.8900			
12 mg/ml	3			72.7767		
16 mg/ml	3				78.1733	
20 mg/ml	3					95.8700
Sig.		1.000	1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.