

DEVELOPMENT OF A NON-CAFFEINATED COFFEE FROM DATE PALM SEEDS (*Phoenix dactylifera L.*) AND ITS BIO-ACTIVE PROFILE CHARACTERIZATION

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Examination Roll No: 0120/04

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A thesis submitted in the partial fulfillment of the requirements for the degree of Master of Science in Applied Human Nutrition and Dietetics

Department of Applied Food Science and Nutrition Faculty of Food Science & Technology Chattogram Veterinary and Animal Sciences University, Chattogram-4225, Bangladesh

June 2022

I

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This is to certify that we have examined the above Master's thesis and have found that the thesis is complete and satisfactory in all respects and that all revisions required by the thesis examination committee have been made.

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June 2022

PLAGIARISM VERIFICATION

TITLE of THESIS: DEVELOPMENT OF A NON-CAFFEINATED COFFEE FROM DATE PALM SEEDS (*Phoenix dactylifera L.*) AND ITS BIO-ACTIVE PROFILE CHARACTERIZATION

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Shireen Akther

Associate Professor & Head Department of Food Processing and Engineering Faculty of Food Science and Technology Chattogram Veterinary and Animal Sciences University

DEDICATED to MY BELOVED PARENTS

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List of Abbreviations

| Abbreviations | iations Elaboration | | | | |
|---------------|---|--|--|--|--|
| g/L | gram per liter | | | | |
| mg/dl. | Mili gram per deciliter | | | | |
| mmol/L | Mili mol per liter | | | | |
| μg/dl. | Micro gram per deciliter | | | | |
| G | Gram | | | | |
| RDA | Recommended Daily Allowance | | | | |
| USDA | United States Department of Agriculture | | | | |
| cal | Calorie | | | | |
| W | Weight | | | | |
| mg | mili gram | | | | |
| МС | Moisture Content | | | | |
| Abs | Absorbance | | | | |
| RAE | Retinol Activity Equivalents | | | | |
| TTA | Total Titratable Acidity | | | | |
| TSS | Total soluble solids | | | | |
| TPC | Total Phenolic Content | | | | |
| AOAC | Association of Official Analytical Chemists | | | | |

Abstract

Caffeine is an addictive substance whose everyday use may result in major health issues including the induction of skin collagen synthesis, high blood pressure, hypertension in young people, indigestion, and infertility. Therefore, there is a great demand for a natural alternative that tastes, looks, and smells like coffee but has no health risks. Current study shows, coffee was produced through a roasting heat technique by utilizing four different varieties (Ajwa, Maryaam, Zahidi, Safawi) of date seeds profoundly available in the markets of Bangladesh which have no significant value in our country at all. Quality characteristics of coffee-like brew from roasted date seeds (Phoenix dactylifera L.) were determined and compared with those of traditional market coffee. The evaluation of physicochemical character, proximate composition, mineral, and bioactive component analysis of the produced date seed coffee powder was performed. The analysis showed, in the control, ajwa date seed coffee, maryaam date seed coffee and mixed date seed coffee samples, respectively, pH ranged from 4.43-4.56, TSS (°Brix) 1.96-2.76, TTA (% Ascorbic Acid) 1.11-1.21 and nutritional components led to crude protein percentages of 6.95, 7.62, 6.97 and 7.38; crude fiber percentages of 30, 54.72, 54.38 and 55.43; and carbohydrate percentages of 52.10, 24.51, 25.44 and 23.87. Mineral content determination showed that roasted seed powder samples were a source of Zn^{2+} , Cu^{2+} , Na^+ and Fe^{2+} . Additionally, vitamin A and C also identified in substantial amount in every date seed coffee sample which was comparatively higher than control. However, the date seed beverage was found to have higher amount of Antioxidant capacity (DPPH inhibition), Total Phenolic Contents (TPC) and Total Flavonoid Contents (TFC) of 2.74, 180 mg GAE/100g and 210 mg QE/100g which can be considered as strong bioactive characterization than regular coffee. It has been established that the chemical and bioactive profile of dates may change based on the date variety and roasting process. Finally, a reviewing panel of 10 judges decided if the samples were acceptable. The judges appreciated every sample, but the ajwa variety gained the highest marks. Consequently, after considering the chemical components and sensory assessment of roasted date palm seeds, it can be concluded that wasted date seeds may be a significant and excellent source of an economical coffee alternative. (Keywords: Palm date seeds, roasting, physiochemical composition, bioactive compound, coffee like brew, by-product.)

Chapter I: Introduction

1.1 Background

For Asian people, the date palm tree (*Phoenix dactylifera*) performs an essential economical function. Dates are a major agricultural product grown in the majority of Middle Eastern countries, and millions of people in this region use dates as a staple food (El-Nemr et al., 2007). Phoenix dactylifera is a member of the *Arecaceae* family, sometimes known as the *Palmaceae*, and its date seeds make up around 10% to 15% of the fruit mass (Metoui et al., 2018). There are over 2000 distinct types or cultivars in the current world, although only a few numbers are recognized for their fruit quality (Besbes et al., 2004a). The chemical composition of date pits appeared excellent, with substantial levels of polysaccharides (75–80%), proteins (5–6%), and fat (10–12%) (Fikry et al., 2020). Additionally, date pits are high in dietary fibers, polyphenols, antioxidants and flavonoids, all of which may aid to defend against severe serious diseases including cardiovascular disorders and cancer, as well as lowering blood pressure and regulating blood sugar levels (Chiara et al., 2007).

Organic products include health-promoting natural compounds that have a positive impact on the body's physiology. These components, known as nutrients, are found in fruits and vegetables and are responsible for its therapeutic potent (Espin et al.,2007). Typically, equally edible and inedible segments of fruits and vegetables are present where the inedible section is normally the seed or pit that is responsible for the plant's development and reproduction. As a result, it may include more nutritious characteristics than other sections (Al-Farsi et al., 2008). However, it is a common practice in underdeveloped nations to consume edible items and discard inedible elements. The amount of solid and liquid waste produced by the food processing industries quite significant. If this trash is not recycled, it may surely affect the ecosystem. In the fruit industry, fruit seeds are known as organic waste. Numerous experts are interested in valuing organic by-products from the food sector for two main reasons: environmental conservation and economic exploitation of organic wastes (Bouaziz et al., 2010).

Date kernels, on the other hand, are mostly ignored or only partially included into animal feed in most producing nations (Lecheb et al., 2011). In sub-Saharan Africa, date palm by-products (stems, leaves, pedicels, pits, and so on) have a variety of traditional

applications. Furthermore, since it is high in polyphenolic and dietary fibers, which may be employed as medicinal components, therefore, not using these valuable seeds for human nutrition is a significant financial loss (Khali and Boussena, 2015). Recently numerous studies have emphasized the nutritional benefit of date kernels, and their use among both edible and non-food compounds has been recommended (Al-Meqbaali et al., 2017). Flavonoids, sterols, phenolic acids, and tocopherols are among the phytochemicals found in date seeds. Date seeds contain a lot of hesperidin, which is a kind of flavonoid. Hesperidin possesses anti-cancer, anti-atherosclerosis, and anti-bone loss properties. Besides, Phenolic acids include hydroxybenzoic protocatechuic, coumaric, ferulic and caffeic acids (Al-Farsi et al., 2008). Also, antioxidant organic substances in date seeds can lessen the risk of chronic illnesses by preventing oxidative damage induced by increasing dietary consumption (Lourenco et al., 2019). An increasing percentage of studies have been focused to discovering natural antioxidants that may be utilized in food items because certain synthetic antioxidant compounds can be dangerous for human health (Sami et al., 2015).

Some rural communities use roasted, powdered date seeds to make coffee-like food and as a substitute for coffee (Sobia et al., 2017). Coffee consumption is one of the most popular ways to assist our everyday activities across the world. It was commonly used as a tonic to keep people active and healthy. Coffee consumption is often seen as a gesture of friendliness and socializing. It has certain benefits, such as restoring mental health and sleepiness while tired. At the same time, coffee is associated with drinking alcohol and smoking. In addition, caffeine is created from coffee in a form that is more concentrated than that of any other food product (Chitra and Mothil, 2016). Caffeine is a chemical that is consumed on a regular basis, yet it is still an addictive substance. Caffeine is first consumed as chocolate in childhood. They people starts to drink caffeinated Beverage as they get older. Coffee intake has become a routine activity in social contexts such as dates, meetings, and so on as adolescents and young adults, and coffee has thus turned into a typical element of day to day. Caffeine has no constraints on its usage because of its high societal value, therefore the health concerns associated with caffeine intake are unchecked. Caffeine usually provides some advantages for physical performance, such as lowering cognitive impairment, alertness, pain relief, and hydration, as well as reducing the risk of vascular illnesses in the brain and heart (Diego et al., 2008). While simultaneously acknowledging the fact that caffeine has its drawbacks where Caffeine is

a psychoactive substance linked to high blood pressure and panic attacks. The use of coffee has been linked to an increase in stress levels. Kristjansson et al. (2013) found indications of aggressive attitude in caffeine-addicted teenagers. According to Diego et al. (2008), caffeine usage during pregnancy increases the probability of sadness and stress symptoms. Babies born to families who consumed quite so much coffee during pregnancy had issues such as low birthweight and stressful responses including convulsions, and hiccups (Kristjansson et al., 2013).

Individuals who receive a stimulant on a regular basis should be aware of the hazards, and excessive caffeine consumption should be avoided. Individuals who consume a lot of coffee may benefit from using roasted date seeds powder as a substitute without much compromise with taste (Faupel et al., 2013). In this context, the potentials of date seed coffee powder were studied to assess its de-caffeinating properties. Besides, the present study evaluated the physicochemical, proximate composition, mineral contents and phytochemical contents including total phenolic contents (TPC), total flavonoid contents (TFC) and in-vitro antioxidant activity (DPPH assay) of coffee obtained from the roasted date seeds of four variety (Ajwa, Maryaam, Zahidi, Safawi), widely available in Bangladesh.

1.2 Rationale and significance of the study

Evaluating the bioactive component profile of date seeds is the main purpose of this research which is usually an agro-waste, and their use to develop a non-caffeinated coffee. Besides, Agro-wastes that are discarded in the environment are responsible for environmental pollution since it contains hazardous chemicals. On the other hand, it may be used in multiple ways based on its physicochemical characteristics. Currently, there has been a noticeable rise in public awareness regarding functional foods because it contains certain bioactive substances including flavonoids, carotenoids, anthocyanins, fiber, catechin, epicatechin and essential oils. Numerous scientific research conducted over the last several decades have shown that flavonoids and other phenolic components reduce the chance of developing metabolic syndrome, which includes obesity, hypercholesterolemia and hyperglycemia. As a result, the seeds of date require special consideration due to their variety of applications in such forms as substitute of coffee, flour, seed oil and so on.

1.3 Objectives of the study

- To develop non-caffeinated coffee from four different varieties of date (Ajwa, Maryaam, Zahidi, Safawi).
- To compare physicochemical properties of developed date seed coffee
- To investigate proximate composition, vitamin, and mineral contents
- To evaluate the bio-active contents such as total phenolic content (TPC), total flavonoid content (TFC), antioxidant activity (DPPH assay) of the date seed coffee
- To assess the overall acceptability of the developed date seed coffee

Chapter II: Review of Literature

2.1 Botanical description of date seeds

A notable member of the Palmacea family is the date palm, also known by its scientific name Phoenix dactylifera L. Dates come in approximately 2000 different types, each with a unique form, shape and mass. They are typically oblong in form; however, some kinds can be almost spherical. The average weight per fruit is 2.60 g, and the length spans from 1.8 to 11.0 cm and the breadth from 0.8 to 3.2 cm (Zaid, 2002). The features of the seed vary widely depending on type, as well as on the location and growth conditions, similarly as those of the fruit. The seed's weight is between 0.5 and 4 g, its length is between 1.2 and 3.6 cm, and its diameter is between 0.6 and 1.3 cm. The seed typically has an oblong shape, a ventrally grooved surface, a tiny embryo, and a hardendosperm composed of a cellulose deposit on the interior of the cell walls. (Zaid, 2002).



Figure 1: Different variants of date seeds

2.2 Chemical composition of date seed

2.2.1. Macronutrients composition of date seed

Numerous studies on the composition of palm seeds from throughout the universe have already been undertaken. According to Saafi et al., (2008), the seeds of a variety of maturing common date species referred to as "Khalti" were found to have 6.88 percent moisture, 8.12 percent total sugars, 8.33 percent fat and 5.31 percent protein. On the other hand, protein 5.56 and 5.17 percent, fat 10.19 and 12.67 percent, ash 1.15 and 1.12

percent, and total carbohydrate 83.1 and 81.0 percent were similarly obtained for Deglet Nour and Allig cultivars, respectively (Besbes et al., 2004a). The seeds of Kabkab and Shahani date types from Bushehr, Iran, included 10.50 percent moisture, 5.56 percent protein, 12.59 percent lipid, 62.18 percent soluble fiber in acidic solutions, and lastly 1.35 percent ash (Amir et al., 2014). However, the changes in seed content across date types might be related to cultivar variability as well as climate circumstances (Saafi et al., 2008). As per Abdul et al., (2013), the fat content of date seeds was affected by variety, provenance, cultivation practices, and manure. Date seeds, on the other hand, are mostly composed of carbohydrate and fat. In compared to date flesh, the protein and fat composition of date seeds is comparatively high (1.5-3 percent protein and 0.1-1.4 percent fat) (Al – Farsi and Lee, 2011).

In comparison to other seed proteins (soybean, cottonseed, peanuts), date seed protein comprises the majority of necessary amino acids (Al-Farisi and Lee, 2014) and has a comparatively larger proportion of sulphur amino acids (methionine, cystine). About half of the total amino acids extracted from the seeds of Ruzeiz and Sifri date cultivars are glutamic acid, aspartic acid, and arginine. The first limiting amino acid in date seed proteins is tryptophan. Additionally, date seeds have more lysine than other foods (Sawaya et al., 1984). Date seeds contain soluble proteins for instance albumin, glutelin and globulin (Abdul et al., 2013).

Seeds of date have a higher fiber content of 77.8–80.2 g/100 g fresh weight (Al-Farsi et al., 2007) or 64.5-80.15 g/100 g fresh weight (Al-Farsi and Lee, 2011). According to Abdul et al., (2013), date seeds contain 58 percent total dietary fiber, with 53 percent of it being insoluble dietary fiber (cellulose, hemicellulose and lignin). As per Hamada et al., (2002), the seeds of three date varieties (Fard, Khalas, and Lulu) grown in the United Arab Emirates contained 46-51 percent acid detergent fiber and 65-69 percent neutral detergent fiber, suggesting a high degree of lignin and resistant starch. Dietary fiber variations are connected to ripening stages and diversity. In compared to cereal grains and oil seeds, the phytic acid content was low (Hamada et al., 2002). Dietary fiber has several health benefits, including the prevention of hypertension, cardiovascular disease, gastrointestinal issues, high blood cholesterol, cancer (Tariq et al., 2000; AlFarsi and Lee, 2011; Hejri Zarifi et al., 2012). As a result, date seed powder may be useful in the treatment of diabetes, obesity, and hyperlipidemia. Date seed, according to Almana and Mahmaud (1994), may contribute significantly to dietary fiber intake.

| | Compositio | on (%) | | | | |
|-----------------------|------------|--------|---------|-------|------------|--------------------|
| Date Varieties | | | | | References | |
| | Moisture | Fat | Protein | Ash | Total | |
| | | | | | СНО | |
| Rajshahi | 5-10 | 7-10 | - | 1-2 | 55-65 | Joardder et |
| (Bangladesh) | | | | | | al.,2012 |
| Fard (UAE) | 10.3 | 9.9 | 5.7 | 1.4 | - | Hamda et al., |
| | | | | | | 2002 |
| Khalas (UAE) | 7.1 | 13.2 | 6.0 | 1.8 | - | Hamda et al., |
| | | | | | | 2002 |
| Lulu (UAE) | 9.9 | 10.5 | 5.2 | 1.0 | - | Hamda et al., |
| | | | | | | 2002 |
| Allig (Tunisia) | - | 12.67 | 5.17 | 1.12 | 72.59 | Besbes et al., |
| | | | | | | 2004a |
| Deglet Nour | - | 10.19 | 5.56 | 1.15 | 81.0 | Besbes et al., |
| (Tunisia) | | | | | | 2004a |
| Khalti (Tunisia) | 6.88 | 8.33 | 5.31 | - | 83.1 | Saafi et al., 2008 |
| Ruzeiz | 5.4 | 10.2 | 6.8 | 1.1 | 61.5 | Sawaya et al., |
| (Soudi Arabia) | | | | | | 1984 |
| Sifri | 5 | 10.4 | 6.5 | 1.1 | 58.5 | Sawaya et al., |
| (Soudi Arabia) | | | | | | 1984 |
| Date Seed | - | 8.08 | 7.08 | 0.98 | 62.3 | Rahman et al., |
| Powder (Oman) | | | | | | 2007 |
| Kabkab & | 10.5 | 12.59 | 5.56 | 1.35 | 80.6 | Amir Azodi et |
| Shahani (Iran) | | | | | | al., 2014 |
| Mabseeli | 3.14 | 5.02 | 3.92 | 1.03 | 86.8 | Al-Farsi et al., |
| (Sun-dried, | | | | | | 2007 |
| Oman) | | | | | | |
| Um-sellah(Sun- | 4.4 | 5.9 | 5.4 | 1.16 | 83.1 | Al-Farsi et al., |
| dried, Oman) | | | | | | 2007 |
| Shahal(Sun-dried, | 5.19 | 5.09 | 2.29 | 0.89- | 86.5 | Al-Farsi et al., |
| Oman) | | | | | | 2007 |

 Table 2.1: Various date varieties' seed composition:

2.1.2. Micronutrient composition of date seed

The concentrations of several minerals, including potassium, sodium, calcium, and magnesium, are significantly greater in date seeds (Al- Hooti et al., 1998; Devshony et al., 1992, Besbes et al., 2004a). Iron, manganese, zinc, and copper are the microelements that are found in higher concentrations (Sawaya et al., 1984). According to Attalla and Harraz (1996), eleven date cultivars in the Qassim region of Saudi Arabia seem to have little amounts of phosphorus in their seeds (0.19 to 0.26 percent). Selenium is a different element that might be present in date seeds. Ten different date species were grown in Suadi Arabia, and their selenium contents ranged from 1.48 to 2.96 mg/g (Al-Showiman et al., 1994). Some date varieties have higher selenium concentrations than others, which may be related to the selenium content of the soil (Al-Farsi and Lee, 2011). Ali-Mohamed and Khamis (2004) compared the mineral ion concentrations of Bahraini date seeds, coffee, and barely. Coffee, date seeds, and hardly have the highest concentrations in date seeds were safe for dietary use.

2.1.3. Bio-active constituents of date seed

Date seeds are high in antioxidants (580-929 µmol trolox equivalents/g fresh weight) and phenolic compounds (3102-4430 mg gallic acid equivalents/100g fresh weight) (Al-Farsi et al., 2007). Except for olive oil, date seed oil has a greater phenol concentration than most other culinary oils (Besbes et al., 2005). When acetone–water, ethanol–water, methanol–water, and water alone were employed as solvents for extraction at temperatures of 22, 45, and 60°C, Guizani et al. (2014) found that polyphenol content in date seeds ranged from 21 to 62 mg gallic acid equivalents/g date seed. Iranian date seed cultivars exhibited a reasonably high antioxidant activity and were powerful radical scavengers, according to Ardekani et al. (2010), and might be exploited for medical and commercial applications. Date seeds include oleic, lauric, linoleic, and palmitic acids, with oleic acid (41.3-47.7 g/100g) having the highest oxidative stability of all. Linoleic acid is also required for good skin health. The primary phenolic acids were phydroxybenzoic acid (9.89 mg/100g), protocatechuic acid (8.84 mg/100g), and mcoumaric acid (8.42 mg/100g) (Al-Farsi and Lee, 2008).

2.2. Date seeds application

2.2.1. Use as animal feeding

Date seeds are thrown or utilized as animal feed in most date-producing countries (Devshony et al., 1992; Saafi et al., 2008; Habib et al., 2013). Animal feed (livestock, sheep, camels, and poultry) and fish feed contain date seed powder. Incorporating date seed into an animal's diet has been shown to stimulate growth, feed efficiency, and meat sensory quality (Elgasim et al., 1995; Hussein et al., 1998; Al-Farsi and Lee, 2011). Additionally, d ate seeds are occasionally used as an organic soil supplement (Guizani et al., 2014). In addition, these seeds are also a fantastic foundation for developing activated carbon (Joardder et al., 2012).

2.2.2. Use as drug

Date seed powder has also been used in conventional medicines and in the manufacture of citric acid and polypeptide by Candida lipolytica, Candida utilis and Apergillus oryzae. Date seeds are said to have antibacterial action, although only an ethanolic date seed extract has been demonstrated to exhibit antimicrobial effect on some microbes for a whole week (Jassim and Naji, 2010). According to Jassim and Naji (2010), date seed extracts may suppress the infectivity of Pseudomonas phage ATCC 14209-B1. Consequently, date seed extracts had the potential to be an antiviral drug agent against pathogenic human viruses.

2.2.3. Use in food industries

Date seeds may be used in a range of foods and can be added to a variety of products. Date paste was enhanced with seed powder by Al-Farisi and Lee (2014) in order to improve nutritional quality and hardness. The best formulation was fortified date paste with 3 percent of dried seed powder, which had no detrimental impact on the product's sensory characteristics (Al-Farisi and Lee, 2014). Date seed powder has been included into wheat flour in purpose of manufacturing bread in a few studies. When compared to control and various amounts of seeds, pan bread with 15% date seed had the highest overall acceptance score, according to Halaby et al. (2014). They also concluded that date seed powder improved the nutritional content of pan bread and had a hypoglycemic impact, lowering the risk of diabetes illnesses. Saudi Mafrood flat loaves using 10% coarse seed powder were shown to have sensory qualities identical to flat breads containing wheat bran. Fine seed powder flat breads scored lower on sensory evaluations

(color, taste, odor, and overall acceptability) than wheat bran controls (Habibi Najafi, 2011).

2.2.5. Use as seed oil

Date seed oil could be used in cosmetics (shaving soap, body creams and shampoos), medicine (as an atherosclerosis preventative), medicines (as a liniment for indolent tumors), and food formulations (Mahmoud Abdalla et al., 2012). Date seed oil, according to Besbes et al. (2005), was resistant to heat treatment for roughly 30- 40 hours and hence may be used in culinary applications (frying and cooking). Date seed oil has a high level of oxidative stability and may be safely kept for a long time. Date seed oil can protect skin against cellular damage produced by UV-A and UV-B radiation, as well as oxidative stress damage induced by hydrogen peroxide (Besbes et al., 2004b). Furthermore, due to its antioxidant action, date seed oil can help to restore human skin. Date seed oil is edible, but due to its low extraction rate, it cannot compete with other oil crops (Boukouada and Yousfi, 2009; Al- Farsi and Lee, 2011). It does, however, contain natural carotenoids (carotene), antioxidants, and phytochemicals, making it a potential essential oil (Habib, 2011).

2.2.6. Use as an alternate of coffee

Nonetheless, Date seed powder has just been introduced to the market as a coffee alternative in both plain and blended versions. The Bouhattam variety's roasted seed powder is a fantastic caffein free beverage that is rich in polyphenols and vitamins and has decreased cytotoxicity while also having a high antioxidant potential (Devshony et al., 1992; Habib et al., 2013).

Chapter III: Materials and Methods

3.1 Area of study

The whole study has been carried out in the laboratory of the Department of Food Processing and Engineering, Department of Applied Food Science and Nutrition, Department of Applied Chemistry and Quality Assurance, Department of Physiology, Biochemistry and Pharmacology as well as Poultry Research and Training Center (PRTC) of Chattogram Veterinary and Animal Sciences University (CVASU), Chattogram.

3.2 Sample collection

Samples of dates (*Phoenix dactylifera L.*) were obtained from Sholashohor Karnaphuli Market, 2no. Gate, Chattogram. The dates were picked with care in order to get the best possible ripeness. From a grocery store control (Ama Coffee), sugar and milk powder were purchased for preparing coffee. Other necessary supplies for the experiment were obtained from the laboratory's inventory.

Here,

Sample A= Ajwa Date Seed, Sample M= Maryaam Date Seed, Sample X= Mixed Date Seed (Varieties such as Ajwa, Maryaam, Zahidi, Safawi)



Figure 2: Sampling location in Chattogram, Bangladesh (Google Map)

3.3 Preparation of non-caffeinated date seed coffee powder

The date seed powder was prepared in consistent with the method described in Souda et al., (2020) with minor modification. The manufacture of coffee powder was divided into following stages:



Figure 3: Processing steps of the date seed coffee powder

3.4 Physicochemical analysis of date seed coffee powder

The fresh samples of Date Seed Coffee Powder were analyzed for pH, Total Soluble Solid (TSS), Total Titratable Acidity (TTA) determination. These samples were also analyzed for Proximate Analysis, Bio-Active Compounds Analysis and Antioxidant Analysis.

3.4.1 Determination of pH

A pre-calibrated pH meter was used to calculate the pH of the various samples. Prior to use, the pH meter was calibrated using buffers with pH values of 4, 7, and 10. Approximately 50 mL of distilled water were used to extract 5 grams of roasted date seed coffee powder, which was then filtered using Whatman No. 2 filter paper (Souda et al., 2020). A pH meter (pH ORP/CD, bench type, USA) was then used to determine the pH value of the roasted kernel aqueous filtrate. The device was submerged in the suspended solution, and the reading were recorded.

3.4.2 Determination of total soluble solids (TSS)

Total Soluble Solids (TSS) content of the roasted date kernels were found out with the help of hand refractometer by using the same extract (5g roasted date seed powder dissolved in 5oml of distilled water). Total soluble solids (TSS) were directly recorded by digital refract meter (Atago RX 1000) and the results expressed as percent soluble solids (°Brix) as described in ISO (2173:2003).

3.4.3 Determination of total titratable acidity (TTA)

The percentage of acidity was measured in terms of anhydrous citric acid by titrating against the basic solution of 0.1N NaOH using phenolphthalein indicator as described in AOAC (2005). Each time 5g of roasted date coffee powder sample was taken in a 100ml volumetric flask and the volume was made up to 100ml by adding distilled water. Later 10ml diluted filtrate was titrated against 0.1N NaOH where phenolphthalein was the indicator. The appearance of pink hue indicated the endpoint of the titration. Titration was reported thrice at the average value was recorded.

3.5 Proximate analysis determination

According to the AOAC standard procedure, the proximate compositions of the sample date fruit seed coffee powders were examined (2000). By using the dry

ash technique, the oven drying method, Kjeldahl's method, the gravimetric method, and the Soxhlet method, respectively, the contents of moisture, ash, crude protein, crude fiber, and crude fat were measured (AOAC Method 977.11, AOAC Method 923.03, AOAC Method 955.04, AOAC Method 991.43, and AOAC Method 960.39, respectively).

3.5.1 Determination of moisture content

The moisture content was determined by drying the vacant dish and cover at 105°C for three hours, then shifting them to a desiccator for cooling, following the procedure of AOAC (2000). The empty dish and lid had been weighed. containing a sample that weighs roughly 3g in the dish and by using a spatula to spread the sample. Therefore, placing the dish containing the sample within the conventional oven. Lastly, drying operation for 3 hours by setting the temperature at 105°C. After drying, it was necessary to place the dish in the desiccator to cool off with a slightly closed lid. The dish and its dried sample should be weighed again.

3.5.2 Determination of crude protein

Protein content was determined by following AOAC (2000) method 2.049. Following was the course of events:

Reagent required

Concentrated sulphuric acid (nitrogen-free) 20 ml, Digestion mixture such as Potassium sulphate 100gm, Copper sulfate 10 g, Selenium di-oxide 2.5g that was properly homogenized in a mortar and stored in a dry location. Boric acid solution 2% solution in water. Therefore, Alkali solution 400g of sodium hydroxide should be diluted to 1 liter of water. Mixed indicator solution Bromocresol: 0.1g and methyl red: 2g were dissolved in 250 ml ethyl alcohol. Lastly, standard HCl: 0.1N

Procedure

Accurately weighing 5g of the digesting mixture, it was then transferred to a dry, 300ml Kjeldahl's flask. The appropriate amount of sample (1g for each) was added to the flask. Twenty milliliters of sulphuric acid were added, heated slowly until froth subsided, and then simmered fast. After 15-20 minutes, the solution became clear, and another 45 minutes were spent heating it. After cooling, 500 ml—the entire amount—was

quantitatively transferred to a 1-liter round-bottom flask by adding 100 ml of water. A little amount of sodium hydroxide solution was gently added to the side to create cupric hydroxide precipitation. The steam trap and condenser were readily connected to the flask after that. Then, 50 ml of the boric acid solution, 50 ml of distilled water, and 5 drops of the indicator solution were added to a 500 ml conical receiving flask. A 250 ml quantity of distillate was collected after the condenser was placed and the distillation was performed for 4 to 5 minutes. After that, 0.1 N hydrochloric acids were used to titrate the contents of the receiving flask, and a brown coloring served as an indicator of the finish point. Additionally, a reagent blank was found and subtracted from the titration. One gram of nitrogen is equal to one milliliter of hydrochloric acid at a concentration of 0.1N. The percentage of protein from nitrogen was calculated using a protein conversation factor.

3.5.3 Determination of crude fat

Using the gravimetric solvent extraction technique provided in AOAC (2000), this was verified. The sample was wrapped in porous paper and five grams were put in a thimble (Whatman filter paper). A weighted extraction flask containing 200 ml of petroleum ether and a soxhlet reflux flask were used to hold the thimble after mounting it in them. The top of the reflux flask had a water condenser attached to it. After being heated, brought to a boil, and then given time to cool, petroleum ether was condensed into the reflux flask. The solvent was poured over the sample in the thimble and the oil extract was siphoned down to the boiling flask as soon as the reflux flask was fully filled. The defatted sample was removed, the solvent was recovered, and the oil extract was kept in the flask after this process was done four times. To remove any leftover solvent, the flask containing the oil extract was dried in the oven at 60°C for one minute. It was weighed after cooling in a desiccator. Thus, the weight of the oil (fat) extract was calculated as a percentage of the sample's overall weight.

3.5.4 Determination of crude fiber

The AOAC technique (2000) was used to calculate crude fiber. A 5g sample of roasted powder was cooked for 30 minutes at reflux in 150 ml of a 1.25 percent H_2SO_4 solution. The cooked sample was washed several times in hot water with a two-fold towel to capture the particles. It was put back in the flask and cooked for a further 30 minutes under the same conditions in 150 ml of 1.25 percent NaOH. The sample was transferred

quantitatively to a weighted crucible and washed in multiple volumes of hot water before being allowed to drain dry and then dried in an oven at 105°C to a consistent weight. It was then transported to a muffle furnace and burnt until nothing, but ash was left. By using difference, the weight of the fiber was computed as a percentage of the weight of the sample that was being tested.

3.5.5 Determination of ash content

The AOAC (2000)- described furnace gravimetric method was used to achieve this. In brief, 5g of the samples were weighed into a porcelain crucible and measured there. The sample was burnt until it was reduced to ashes in a muffle furnace that was heated to 550°C. After chilling in a desiccator, it was weighed and analyzed.

3.5.6 Determination of carbohydrate

Carbohydrate was determined by the following formula as per AOAC, 2000.

3.6 Minerals determination

This technology utilizes digestion to remove minerals from the food material, as per AOAC, 2010. A sample of date seed roasted powder was digested in an acid solution consisting of HNO₃ and HCIO₄ into a 2:1 ratio. In a conical flask, one gram of sample was weighed. 7 ml HNO₃ and 3 ml HCIO₄ were added, and then the flask was placed on a hot plate at 200W for 3 minutes until complete digestion. The solution was cooled down and filtered through filter paper into a 100 ml standard flask and diluted to the volume with distilled water. This solution was used for mineral content determination by AAS (Humalyzer-3000, Origin Germany). The levels of different minerals including sodium (Na), potassium (K), copper (Cu), chloride (Cl), phosphorus (P), potassium (K), magnesium (Mg), iron (Fe), calcium (Ca) and zinc (Zn) were measured.

3.6.1 Determination of sodium (Na)

Magnesium and uranyl acetate are used to precipitate sodium as a triple salt. A brownish tint results from the reaction of ferrocyanide with excess uranyl ions in an acidic media. The sodium concentration in the sample has an inverse relationship with the color intensity that results. In the precipitation step, a pipette was used to add 0.02 ml of sodium standard and 1 ml of precipitating reagent to the cuvette. In the cuvette for the sample, 0.02 ml sample and 1 ml of precipitating reagent were applied. These were thoroughly

combined, and after 5 minutes of retention time, they were shaken properly. To generate a clean supernatant, these were later centrifuged at 2500 to 3000 RPM. For the blank in the color development phase, use 1 ml of acid reagent. Using a pipette, 0.02 ml of precipitating reagent and 0.1 ml of color reagent were applied to the cuvette. A cuvette was filled with 1 ml of acid reagent, 0.02 ml of supernatant, and 0.1 ml of color reagent for the fabrication of standards and samples. Incubated them at R.T. for 5 minutes after mixing. Within 15 minutes, the absorbance of the blank, the standard, and the sample were measured against distilled water. The sodium concentration was determined in mmol/L by multiplying the sample absorbance by standard absorbance by standard concentration (mmol/L).

3.6.2 Determination of phosphorus (P)

For blank solution preparation only 1 ml phosphorus reagent, for standard 1 ml phosphorus reagent, $10 \,\mu\text{L}$ phosphorus standard and for sample solution 1 ml phosphorus reagent, $10 \,\mu\text{L}$ sample extract were added into cuvette by pipette. Later these were mixed and incubated for 5 minutes. The absorbance of the sample and standard was compared to a blank. The ratio of sample absorbance to standard absorbance is multiplied by standard concentration (mg/dL) and concentration of phosphorus was obtained in mg/dL.

3.6.3 Determination of copper (Cu)

Digestion using a strong acid, such as HNO3, HCl, or H2SO4, is a standard sample preparation technique for solid materials. Samples can be immediately put into flame AAS as well as graphite furnace AAS after dilution of the digested solutions. Standard and sample absorbance were measured against a blank. Concentration of iron was obtained in μ g/dL.

3.6.4 Determination of chloride (Cl)

Mercuric thiocyanate is converted into thiocyanate when chloride ions interact with free mercuric ions. A reddish-brown ferric thiocyanate complex is created when the released thiocyanate mixes with the ferric ions. The amount of chloride in the sample directly relates to how intense the color is. For the preparation of blank solution, 1 ml chloride reagent, 0.01 ml deionized water were added into cuvette. 1 ml chloride reagent and 0.01 ml chloride standard were taken for standard solution preparation. 1 ml chloride reagent and 0.01 ml sample extract for sample solution preparation. After mixing these incubated

at retention time for 2 minutes. The absorbance of standard and sample were measure against blank within 60 minutes. Chloride concentration in mmol/L was found by multiplying standard concentration (mmol/L) with the ratio of sample absorbance to standard absorbance.

3.6.5 Determination of magnesium (Mg)

The strategy is based on a shift in the complex's absorption wavelength caused by the particular binding of calmagite which is a metallochromic indicator, to magnesium at an alkaline pH. The amount of magnesium present in the sample has a direct correlation with the intensity of the chromophores that are produced. One milliliter of the reagent was taken in the cuvette to make the reagent blank solution. The cuvette containing the prepared sample solution received 1 ml of reagent and 10 L of sample extract. One milliliter of reagent and ten milliliters of magnesium standard were placed in the cuvette to prepare the standard solution. After mixing, allow the cuvettes to rest at room temperature for two minutes. In comparison to the reagent blank, the absorbance of the sample and standard at 520 nm was measured. The concentration of magnesium was calculated in mg/dL by multiplying the sample absorbance by the standard concentration (mg/dl).

3.6.6 Determination of potassium (K)

A fine turbidity of potassium tetraphenylboron is generated when potassium and sodium tetraphenylboron combine. The amount of turbidity is inversely correlated with the sample's potassium content. For the preparation of blank solution, 1 ml potassium reagent and 0.02 ml deionized water added into cuvette by pipette. For standard solution, 1 ml potassium reagent and 0.02 ml potassium standard and for sample solution, 1 ml potassium reagent and 0.02 ml sample extract were added into cuvette. After mixing these were incubated at retention time for 5 minutes. Within 15 minutes, the absorbance of the Standard and sample were tested against a blank. The ratio of sample absorbance to standard absorbance is multiplied by standard concentration (mmol/L) and potassium concentration was obtained in mmol/L.

3.6.7 Determination of iron (Fe)

The transferring-iron complex is dissolved by a moderately acidic medium, releasing the iron. Ascorbic acid is used to reduce the liberated Fe to the bivalent form. Ferrozine

forms a colorful compound with ferrous ions. The intensity of the color produced is related to the sample's iron content. For the preparation of blank solution, 1 ml reagent was added into cuvette with the help of pipette. For standard preparation, 200 μ L standard and 1 ml reagent were added. 200 μ L sample extract and 1 ml reagent were added for the preparation of sample solution. These were mixed and then incubated for ten minutes at room temperature. Standard and sample absorbance were measured against a blank. Concentration of iron was obtained in μ g/dL.

3.6.8 Determination of calcium (Ca)

Calcium ions form a violet complex with O-Cresolphthalein in an alkaline medium. For the preparation of reagent blank solution, 25uL distilled water and 1 ml working reagent were added into cuvette. For standard, 25μ L (Ca⁺⁺) standard and 1 ml working reagent were added. 25 μ L sample extract and 1 ml working reagent were added for the preparation of sample solution. The absorbance of both the sample and the standard was determined. The ratio of sample absorbance to standard absorbance is multiplied by standard concentration (mg/dL) and concentration of calcium was obtained in mg/dL.

3.6.9 Determination of zinc (Zn)

Zinc is an alkaline medium reacts with nitro -PAPS to form a purple-colored complex. Intensity of the complex formed is directly proportional to the amount of zinc present in sample. For the preparation of blank solution, 1 ml working reagent, 0.05 ml distilled water were added into cuvette by pipette. For standard solution preparation, 1 ml working reagent and 0.05 ml zinc standard were taken into cuvette. For sample solution preparation, 1 ml working reagent and 0.05 ml zinc standard were taken into cuvette. For sample solution preparation, 1 ml working reagent and 0.05 ml sample extract were taken into cuvette. After mixing well, incubated at retention time for 5 minutes. The absorbance of the standard and sample were measured against the blank within 20 minutes. Concentration of zinc (μ g/dL) was obtained by multiplying 200 with the ratio of sample absorbance to standard absorbance.

3.7. Vitamins determination

3.7.1 Determination of vitamin A

Utilizing a colorimeter, vitamin A was measured. The overall Vitamin A content of a particular meal is calculated using both retinol and betacarotene contributions. Retinol and carotenoids are extracted into light petroleum and combined with alcohol to

precipitate proteins. The light petroleum is evaporated, the carotenoid-induced yellow color intensity is measured, and the residue is then dissolved in chloroform prior to the color reaction. The reaction's contribution from the carotenoid is taken into account (Gibson, 1990). Trifluoroacetic acid interacts with the retinol in the sample (TFA). A blue tint is seen during the sample and TFA reaction, suggesting the presence of retinol in the sample. Since the blue hue is momentary, it must be seen as soon as possible after introducing the reagent-ideally, within two seconds (Guamuch et al., 2007). To prepare each sample Using a vortex mixer, 100 mg of the sample, 1 ml of distilled water, and 2 ml of ethanol were combined in a tube. 1 cc of the supernatant was removed after the tube had been centrifuged for 15 minutes at 3000 rpm. Carotene was discovered first. S2 reagent (6 ml) was used to prepare the blank solution, and standard reagent (6 ml) was pipetted into the cuvette for standard preparation. 1 ml of sample extract, 2 ml of S1 reagent, and 3 ml of S2 reagent were pipetted into a cuvette to prepare the sample solution. For ten minutes, all were thoroughly blended using a vortex mixer and a mechanical shaker. The tubes were centrifuged at 3000 RPM for 10 minutes. The absorbance was then measured at 420 nm in comparison to the blank using 2 ml of sample supernatant, standard, and blank. To stop the solvent from evaporating and the sun from destroying the carotenoids, this was done right away. The retinol was then identified. To make the sample solution, 2 ml of the sample extract used to determine the number of carotenes was collected, and the contents of the sample cuvette were dried out in a water bath heated to 50 °C. The sample cuvette was then filled with 1001 S4 reagent and 1 ml S5 reagent once the solvent had evaporated. 100 ml of S4 reagent and 1 ml of S5 reagent were pipetted into a cuvette to prepare the blank solution. 100 ml of standard reagent and 1 ml of S5 reagent were used to prepare the standard solution. Using a vortex mixer, these were well blended. At precisely 2 seconds after the reagent was added, the absorbance at 620 nm was measured. S5 reagent is a strong acid that emits an irritating vapor, hence.

The carotene, retinol and total vitamin content were measured as follows,

Retinol (mg/L) = $(0.0759 \times \text{Absorbance}) + 0.1023$

Carotene (mg/L) = $(-0.0167 \times \text{Absorbance}) + 0.0091$

Where, 0.0759 and 0.0167 are slope; 0.1023 and 0.0091 are intercept

Total vitamin A (RAE) = μ g of retinol + (μ g of beta-carotene / 6)

3.7.2 Determination of vitamin C

Vitamin C is a vital component for health, but during food preparation, packing, and storage, heat and air quickly reduce or destroy it. The recommended method of analysis for figuring out how much vitamin C is in beverages is the 2, 6-dichloroindophenol titrimetric technique (AOAC, 2010). Here, the color pigment caused the vitamin C to oxidize into dehydroascorbic acid. The dye is also changed into a colorless substance at the same time. Therefore, it is simple to identify the reaction's termination point. As excess may be introduced into plant products by oxidized vitamin C that is partially destroyed during sample and grinding, rapid excretion and filtering are preferred. To prevent oxidation during extraction, metaphosphoric acid is utilized. An extremely acidic solution will get the most accurate result. The titration should be complete in one minute. When the dye is dissolved in water, it turns blue; when dissolved in acid, it turns pink; and when completely reduced, it turns colorless.

Reagent requirement

Dye Solution [260 mg of dye (2,6-dichlorophenol indophenols) and 210 mg of NaHCO₃ dissolved in 100 ml of distilled water], Metaphosphoric acid solution (3%) [15/7.5mg of Metaphosphoric acid and 40/20ml of glacial acetic acid dilutes to make 500/250 ml with distilled water]. Standard ascorbic acid solution [50/25 mg of crystalline ascorbic acid dissolved in 500 ml/250ml of metaphosphoric acid solution].

Procedure

In the burette, dye solution was added up to 0 markings. Then, a conical flask containing 5 ml of Vitamin C solution was in use. Drop by drop, the dye was applied to the conical flask while it was positioned beneath the burette. Titration was finished when pink hue developed and persisted for 20 seconds before disappearing. At least three readings were taken. The same procedure was performed for ascorbic acid solution of unknown concentration. The result was expressed as milligram percentage (mg %).

3.8 Phytochemical content determination

Preparation of Extract

Taking 1gm of sample in Felcon tube and adding 10ml absolute ethanol which left for 72hours. Then, straining the solvent and collection of filtrates after 72 hours. Finally, the ethanoic extract found.

3.8.1 Determination of total polyphenolic content (TPC)

Procedure

The Folin-Ciocalteu reagent technique described for measuring TPC was used to determine TPC of the extracts with a few minor changes (Al-Owaisi et al., 2014). 1.5 ml of FC reagent was added to 1 ml of ethanoic extract in a falconer tube, which was then kept at room temperature for 3 min. The mixture was then given 1.5 ml of 7.5% Na₂CO₃ and was allowed to sit for 60 minutes. Using a UV-VIS Spectrophotometer (UV 2600, Shimadzu Corporation, USA) and C2H5OH as the blank, the absorbance was measured at wavelength 765 nm. According to calculations, TPC is equal to mg of gallic acid equivalents (GAE) for every gram of extracts.

3.8.2 Determination of total flavonoid content (TFC)

With a few minor adjustments, the aluminum chloride colorimetric method described by Chang et al., (2002) was used to measure the samples' total flavonoid content (TFC). Prepared extract stock solution (1 mg/ml) was diluted in aliquots of 0.5 ml with 15 ml of 95 percent C2H5OH in a cuvette. Then, to the liquid in the cuvette, 0.1 ml of 10% AlCl3, 0.1 mil of 1 mol/L potassium acetate, and 2.8 ml of distilled water (D.H2O) were added. For 30 minutes, the mixture was kept at room temperature. An equal amount of 10 percent aluminum chloride replaced with D.H2O was used as the blank. The absorbance was measured at wavelength 415 mm using a UV-visible spectrophotometer (UV-2600, Shimadzu Corporation, USA). By comparing the sample extracts' absorbance to a quercetin standard curve, the total quantity of flavonoid present in the sample was determined. Quercetin equivalents (QE) per gram of extract (mg QE/g) are a measure of the estimated total flavonoids (TFC).

3.8.3 Determination of antioxidant capacity by DPPH scavenging method

Procedure

Antioxidant mobility of the extracts was determined using DPPH assay as the process described by Azlim et al., (2010) with slight modifications. About 6 mg of DPPH was dissolved in100 ml absolute methanol and prepared methanoic DPPH solution.

Then 1 ml methanoic extract was diluted with of 2 ml DPPH solution. Then the mixture was mildly shaken and left for 30 min in dark at room temperature. The absorbance was read at wavelength 517 mm using UV-VIS spectrophotometer (UV-2600, Shimadnu Corporation, USA). Control prepared by mixing 1 mL of methanol with 2 ml of DPPH solution whilst methanol was used like a blank. The scavenging mobility was measured as the decrease in absorbance of the samples in comparison with the DPPH standard solution. Antioxidant capability based on the DPPH free radical scavenging mobility of extracts calculated using the subsequent equation.

Trolox used as standard and TEAC composite (Trolox equivalent antioxidant mobility) was used for the calibration standard curve. The results were revealed in mg/ 100 g of Trolox equivalents per gram of powder on a dry weight (DW) base.

3.9 Caffeination test (FTIR spectroscopy of date seed coffee powder)

Fourier Transformation Analytical methods like infrared spectroscopy may be used to distinguish between organic, polymeric, and occasionally inorganic materials. Infrared light is used in the FTIR analysis procedure to scan test samples and examine chemical characteristics. Here the date seed powder was subjected to sodium hydroxide delignification and sodium chlorite single step bleaching. Further, samples were examined by FTIR spectroscopy using infrared light. Then FTIR spectrum for each sample was done separately.

3.10 Sensory evaluation of finished product

Samples were assessed for their sensory qualities both as drink and as powder. Roasted date seed powder samples were judged on their overall acceptability, color, flavor, and texture. Consequently, a second round of testing was conducted on powdered roasted date seed samples using the criteria of appearance, flavor, taste, and general approval. A 1–9 point hedonic rating scale was used to assess the samples' acceptability. Ten panelists
were recruited from among the lecturers, students, and employees of the Chattogram Veterinary and Animal Sciences University's Applied Food Science and Nutrition Department, Food Processing and Engineering Department, and they were all briefed on the review procedure before it began. Each of the ten panelists was given a portion of each sample. The taste panelists were asked to score the sample on a scale of 1 to 9, where 1- dislike extremely, 2 dislike very much, 3 dislikes moderately 4 dislikes slightly; 5-neither like nor dislike; 6-like slightly, 7- like moderately, 8 like very much, 9 like extremely (Amerine et al., 2013).

3.11 Costing of produced date seed coffee powder

The production cost of the developed date seed coffee powder has been calculated depending on the following factors: Cost of raw materials (date seeds), electricity cost for roasting and grinding operation, packaging cost in glass bottle and manpower.

3.12 Statistical analysis

Data were collected and kept in a Microsoft Excel 2019 spread sheet for statistical analytical evaluation. For the proximate composition and sensory assessment of date seed coffee samples, descriptive statistics (mean and standard deviation) were performed. MINITAB 21 was used to sort, code, and record the data. Following that, statistical analysis was done. One-way ANOVA techniques were used to examine the physiochemical and proximate composition, phytochemical contents, and sensory assessment data to determine the amount of significant variance at a 95% confidence interval. The statistical analysis was performed with a significance threshold of 5% (p 0.05).

Chapter IV: Results

4.1 Physicochemical analysis of date seed coffee samples

Different samples of date seed were subjected to physicochemical investigation in the experiment. The lab test outcomes for the control (Ama Coffee), ajwa date seed powder, maryaam date seed powder, and mixed date seed powder samples were displayed in Table 2, accordingly. To determine the overall mean difference of values for various parameters of the roasted date seed powder samples, a one-way ANOVA (Analysis of Variance) test was conducted. The findings indicated that there was a substantial mean difference in the values of the several date seed powder properties in comparison to the Control.

Table 2: Physicochemical analysis test results for Control, Ajwa date seed powder,Maryaam date seed powder and Mixed date seed powder.

| | Control | Ajwa date | Maryaam Date | Mixed date |
|--------------------|------------------------|-------------------------|--------------------------|------------------------|
| Parameters | (Ama | Seed coffee | Seed Coffee | seed coffee |
| | coffee) | | | |
| р ^н | 4.56±0.05 ^a | 4.53±0.05 ^{ab} | 4.56±0.05 ^a | 4.43±0.05 ^b |
| | | | | |
| TSS(°Brix) | 2.76±0.05 ^a | 1.96±0.05 ^b | 1.96±0.0577 ^b | 2.00 ± 0.00^{b} |
| | | | | |
| Titratable acidity | 1.21 ± 0.03^{a} | 1.12±0.01 ^b | 1.21±0.03 ^a | 1.11 ± 0.00^{b} |
| (% Ascorbic Acid) | | | | |

Legends: The table contained all values as (ME \pm SD), where ME stands for Mean and SD for Standard Deviation. The superscripts a, b, c, and d indicate significant differences (p \leq .05) across samples.

The values of pH, TSS(°Brix) and TTA (% Ascorbic Acid) of date seed coffee are presented in Table 4.2. All these parameters showed significant differences (p<0.05). The pH, TSS(°Brix) and TTA (% Ascorbic Acid) varied respectively from 4.43 to 4.56, 1.76 to 2.00 and 1.11 to 1.21.

4.2 Proximate analysis

Accordingly, Table 3 illustrated that the results of the lab tests performed on the control (Ama Coffee), ajwa date seed coffee, maryaam date seed coffee and mixed date seed coffee samples.

| | Control | Ajwa Date | Maryaam | Mixed Date |
|-------------------|-------------------------|-------------------------|--------------------------|------------------------|
| Parameters | (Ama | Seed | Date | Seed Coffee |
| | Coffee) | Coffee | Seed Coffee | |
| Moisture (%) | 5.49±0.03 ^a | 2.09±0.01 ^d | 2.16±0.01° | 2.22 ± 0.01^{b} |
| | | | | |
| Crude Protein (%) | 6.95±0.02 ^c | 7.62±0.03 ^a | $6.97 \pm 0.02^{\circ}$ | 7.38±0.02 ^b |
| Ash content (%) | 0.98 ± 0.02^{b} | 1.18±0.01 ^a | 1.16±0.02 ^a | 1.16±0.02 ^a |
| Crude Fat (%) | 4.57±0.08° | 9.91±0.02 ^{ab} | 9.96±0.02 ^a | 9.85±0.01 ^b |
| Crude Fiber (%) | 30±0.00 ^d | 54.72 ± 0.05^{b} | $54.38 \pm 0.06^{\circ}$ | 55.43 ± 0.06^{a} |
| Carbohydrate (%) | 52.10±0.20 ^a | 24.51±0.06 ^c | 25.44 ± 0.04^{b} | 23.87 ± 0.29^{d} |
| | | | | |

Table 3: Proximate analysis test results for Control (Ama Coffee), Ajwa date seed coffee,

 Maryaam date seed coffee, and Mixed date seed coffee samples.

Legends: The table contained all values as (ME \pm SD), where ME stands for Mean and SD for Standard Deviation. The superscripts a, b, c, and d indicate significant differences (p \leq .05) across samples.

It was observed from the above tables obtained from one way ANOVA analysis that there is a significant difference in moisture content (%) of every sample, ranged from 2.22 to 5.49. Highest percentage of moisture found in Control (5.49%) and lowest percentage found in maryaam date seed coffee (2.09%).

For crude protein percentage, there is no significant difference between Control and Maryaam date seed coffee. But there is a significant difference in Ajwaa date seed coffee which is higher than the other samples.

There is no crude fiber found in the control. On the other hand, all the date seed coffee samples have shown an enriched profile of fiber percentage varied from 54.72% to 55.43%.

Significant differences were observed among the crude fat (%) content irrespective of different samples. Highest percentage of fat percentage (9.96±0.02) found in maryaam date seed coffee and the lowest value in the control (4.57±0.08).

For ash content (%), there is no significant difference among the date seed coffee samples. Ash content result of ajwa date seed coffee, maryaam date seed coffee and mixed date seed coffee is 1.18%, 1.16% and 1.16% respectively whereas control has slightly less ash content of 0.98% thus showing a significant difference.

4.3 Minerals determination result

Minerals were analyzed by AAS in the laboratory. Result of the date seed powder samples are shown in Table 4.

Table 4: Minerals test results for Control (Ama Coffee), Ajwa date seed coffee,Maryaam date seed coffee, and Mixed date seed coffee samples.

| Parameters | Control | Ajwa date | Maryaam Date | Date Mixed Date | | | |
|----------------|--------------------------|---------------------------|--------------------------|--------------------------|--|--|--|
| | | Seed coffee | Seed Coffee | Seed Coffee | | | |
| Sodium (Na) in | 21.25±0.44 ^c | 47.51±0.08 ^a | 46.06±0.18 ^b | 46.54±0.29 ^b | | | |
| mmol/L | | | | | | | |
| Phosphorus (P) | 0.11 ± 0.00^{d} | 2.5167±0.07 ^a | 2.1233±0.02° | 2.4000 ± 0.05^{b} | | | |
| in mg/dL | | | | | | | |
| Copper (Cu) in | 8.07±0.126 ^d | $108.57 \pm 0.34^{\circ}$ | 112.62±0.19 ^a | 111.45±0.19 ^b | | | |
| µg/dL | | | | | | | |
| Chloride (Cl) | 0.15±0.00 ^c | 3.42±0.04 ^b | 3.42 ± 0.04^{b} | 3.31 ± 0.13^{a} | | | |
| in mmol/L | | | | | | | |
| Magnesium | 0.32±0.41 ^b | 0.77 ± 0.028^{a} | 0.69 ± 0.01^{ab} | 0.70 ± 0.01^{ab} | | | |
| (Mg) in mg/dL | | | | | | | |
| Potassium (K) | 2.83±0.05 ^a | 1.32 ± 0.02^{b} | $1.21\pm 0.03^{\circ}$ | 1.11 ± 0.02^{d} | | | |
| in mmol/L | | | | | | | |
| Iron (Fe) in | $20.10 \pm 0.07^{\circ}$ | 44.62 ± 0.09^{b} | 44.77 ± 0.07^{a} | 44.62 ± 0.05^{ab} | | | |
| µg/dL | | | | | | | |
| Calcium (Ca) | 0.44±0.03 ^a | 0.01 ± 0.00^{b} | 0.01±0.00 ^b | 0.01 ± 0.00^{b} | | | |
| in mg/dL | | | | | | | |
| Zinc (Zn) in | 31.19 ± 0.06^{d} | 141.50±0.18° | 145.46±0.08 ^a | 144.85±0.05 ^b | | | |
| μg/dL μg/dL | | | | | | | |

Legends: The table contained all values as (ME \pm SD), where ME stands for Mean and SD for Standard Deviation. The superscripts a, b, c, and d indicate significant differences (p \leq .05) across samples.

Among the analyzed macro-nutrients, Zinc (Zn), Copper (Cu), Iron (Fe) and sodium (Na) were the most abundant elements with 145.50 μ g/dL, 111.45 μ g/dL, 44.77 μ g/dL and

47.51 mmol/L respectively in the developed date seed coffees. Besides, significant differences were observed in Magnesium (Mg), Potassium (K), Chloride (Cl) and Calcium (Ca) contents irrespective of different samples ($p \le 0.05$)

4.4 Vitamins determination result

The results of the laboratory bio-chemical analysis of vitamins are displayed below:

Table 5: Vitamins test results for Control (Ama Coffee), Ajwa date seed coffee,

 Maryaam date seed coffee, and Mixed date seed coffee samples.

| Parameters | Control | Ajwa Date | Maryaam | Mixed Date | | |
|-----------------------|------------------------|-------------------------|-------------------------|-------------------------|--|--|
| | | Seed | Date | Seed Coffee | | |
| | | Coffee | Seed Coffee | | | |
| Carotene (mg/L) | 00 ± 0.00^{d} | 2.77±0.03 ^a | 2.59±0.01° | 2.69±0.05 ^b | | |
| Ratinol (mg/L) | 00 ± 0.00^{d} | 99.87±0.07 ^a | 99.08±0.05 ^c | 99.23±0.03 ^b | | |
| Total Vitamin-A (RAE) | 00 ± 0.00^{d} | 17.12±0.03 ^a | 16.56±0.02 ^c | 16.96±0.03 ^b | | |
| Vitamin-C (mg/100g) | 2.83±0.04 ^a | 9.94±0.05 ^b | 9.69±0.02 ^c | 9.77±0.02 ^b | | |

Legends: The table contained all values as (ME \pm SD), where ME stands for Mean and SD for Standard Deviation. The superscripts a, b, c, and d indicate significant differences (p \leq .05) across samples.

Significant difference can be found among the values not only in Vitamin A (RAE)as well as in Vitamin C (mg/100g) through observing from the above tables obtained from one way ANOVA analysis. Total vitamin- A (RAE) varied from 16.96 to 17.12 and amount of Vitamin-C (mg/100g) ranged from 9.69 to 9.944 among the date seed samples where Ajwa date seed coffee found with the highest of vitamins.

4.5 Phytochemical content analysis

Antioxidant capacity and bio-active contents were analyzed by UV-Visible spectrophotometer in the laboratory. Result is shown in Table 6.

Table 6: Phytochemical content analysis test results for Control (Ama Coffee), Ajwa

 date seed coffee, Maryaam date seed coffee, and Mixed date seed coffee samples.

| | Control | Ajwa Date | Maryaam Date | Mixed Date |
|------------------|----------------------|--------------------------|--------------------------|-------------------------|
| Parameters | (Ama | Seed Coffee | Seed Coffee | Seed Coffee |
| | Coffee) | | | |
| Total phenolic | | | | |
| content (TPC) | 16.50 ± 0.01^{d} | $180.07 {\pm} 0.04^{a}$ | $90.79 \pm 0.16^{\circ}$ | 130.32 ± 0.08^{b} |
| (mg QE/100 g) | | | | |
| Total flavonoids | | | | |
| content (TFC) | 51.11 ± 0.86^{d} | 210.72±0.19 ^a | 161.66±0.28 ^c | 197.82 ± 0.04^{b} |
| (mg QE/100 g) | | | | |
| Antioxidant | | | | |
| capacity (DPPH) | 0.11 ± 0.00^{d} | $2.74{\pm}0.04^{a}$ | $1.58{\pm}~0.08^{\rm b}$ | $0.43 \pm 0.02^{\circ}$ |
| (% inhibition) | | | | |

Legends: The table contained all values as (ME \pm SD), where ME stands for Mean and SD for Standard Deviation. The superscripts a, b, c, and d indicate significant differences (p \leq .05) across samples.

The total phenolic content (TPC) of date seed coffee samples ranged from 16.50 to 180.07 with significant differences (p<0.05) between the control, ajwa date seed coffee, maryaam date seed coffee and mixed date seed coffee. The highest phenolic content was reported in Ajwa date seed coffee (180.07 mg GAE/100g) and the least was reported in the control (16.50 mgGAE/100 g). Similarly, Sample of Ajwa date seed coffee had the highest total flavonoid content (210.72 mg QE/100 g), followed by sample Mixed date seed coffee (197.82 mg QE/100 g), sample Maryaam date seed coffee (161.66 mg QE/100 g) and control (51.11mg QE/100g). Moreover, Antioxidant capacity (% inhibition) also showed the similar result where the value ranged from 0.11 to 2.74 where Ajwa date seed coffee had highest value of 2.74.

4.6 Caffeination test (FTIR analysis)

Caffeine makes around 20–40% of typical coffee bean powder. By utilizing FTIR spectroscopic analysis, this has been verified. The disappearance of peaks at 1600 and 1800 cm⁻¹, which demonstrate that this roasted date seed coffee powder contains zero percent caffeine.

4.7 Sensory quality evaluation

The sensory evaluation had been performed by ten semi-trained panelists. The panelists comprised of female and male members who had previous a few experiences on food products evaluation.







Figure 5: Sensory Quality Evaluation of Date Seed Coffee

This method does not, of course, reflect actual consumer perception, but it does strongly indicate attributes which a good quality product should possess. One way ANOVA (Analysis of variance) test was performed to see the overall mean difference of sensory parameter for scores provided by the panelists Figure 4.7.1 and Figure 4.7.2 showed significant difference of mean of different samples.

It was observed from the above tables obtained from one way ANOVA analysis that, there is no significant difference in flavor and mouthfeel of the jellies.

There is a significant difference in taste of the jelly samples. Ajwa date seed coffee got the highest value while maryaam date seed coffee got the lowest.

There is a significant difference in appearance of the seed coffee samples. Control got the highest value comparing with the score of date seed coffee samples.

There is a slightly difference in overall acceptability of the jelly samples. All the jelly samples were positively accepted by the panelist, but the Ajwa date seed coffee was the most preferable.

4.8 Calculation of cost in production of date seed coffee

The production cost of the developed Ajwa date seed coffee has been calculated and Table 7 showed total cost for 500g ajwa seed coffee is approximately Tk 37 in comparison to Control (Ama Coffee) which is charged in market 250tk for 500g.

| Raw materials | Quantity | Price in BDT |
|------------------------|-----------|--------------|
| Date Seed (By Product) | 700g | 00 |
| Electricity Cost for | 1500 watt | 12 |
| Roasting and Grinding | | |
| Bottling | 1pc | 25 |
| Manpower | | 00 |
| Total | | 37 |

Table 7: Production cost of Date Seed Coffee

Therefore, one bottle of 500g date seed coffee would cost only 37taka only.

Chapter V: Discussions

5.1 Physicochemical Analysis of date seed coffee

5.1.1 pH

As pH affects aspects of food including texture, flavor, and scent, among others, it is a crucial factor. The pH of a food affects the growth of bacteria, yeasts, and molds. Microbial development will be inhibited by extremely low or extremely high pH levels. According to Table 2, pH value of Control, Ajwa date seed coffee, Maryaam date seed coffee and Mixed date seed coffee are 4.56, 4.53, 4.56 and 4.43 respectively which reveals the date seed coffee samples can be considered as acidic. Fikry et al.,2019 remarked that the pH of the brew decreased as roasting temperature and duration increased. It is interesting that throughout the roasting process, the pH value of 5.67 dramatically decreased to around 4.56. Therefore, it can be justified that roasting operation playing a big role for pH profile of the developed date seed coffee samples.

5.2.2 Total soluble solids (TSS)

Total Suspended Solids (TSS) is the portion of fine particulate matter that remains in suspension in water. As per Table 2, the control has shown highest value of TSS (°Brix) 2.76, but among the date seed coffee samples there has no significant differences identified. Similar findings were also reported by Ghnimi et al.,2016. On the other hand, according to Youssif et al., (1990), they concluded that the increase of TSS might be attributed to the degradation of polysaccharides in the presence of acid within the date seeds.

5.2.3 Total titratable acidity (TTA)

Since a particular level of acidity prevents the growth of germs in the food goods, acidity is one of the physicochemical features that contributes to the food products' longer shelf life (Tifani et al., 2018). The most noteworthy and least titratable acidity of Mixed date seed coffee and Ajwa date seed coffee was recorded as 1.11 and 1.12 TTA (% Ascorbic Acid). Both control and Maryaam date seed coffee had shown same value of 1.21 TTA (% Ascorbic Acid) (Table 2). According to Youssif et al., (1990), the acidity of dates increased with prolong storage. According to Agarwal and Mangaraj, (2005), increase in acidity was due to the formation of acids by degradation of

polysaccharides and oxidation of reducing sugar or by break down numerous substances. The reduction in acidity may be incompletely because of co-polymerization of natural acids.

5.2 Proximate Analysis of Date Seed Coffee

5.2.1 Moisture content

The most often assessed aspect of food products to determine their shelf life is moisture content (Niazi et al., 2017). In evaluation, the moisture content for Maryaam date seed coffee, Ajwa date seed coffee and Mixed date seed coffee was discovered 2.16, 2.09 and 2.22 percent separately (Table 3). The value of moisture content recorded in this study was much higher when compared to the values (5-10) recorded by Joadder et al.,2012. They concluded, a food's moisture level may be used as an indication for how long it will last and therefore, the low moisture obtained in the study shows that the date seed coffee would have highest possibility to longer shelf life.

5.2.2 Crude protein

The body uses protein for a variety of purposes. It promotes metabolic responses, supports tissue growth and repair, and synchronizes biological processes. The values of protein of Ajwa date seed coffee, Maryaam date seed coffee, and Mixed date seed coffee were 7.62, 6.97, 7.38 respectively which is greater than the control's value of 6.95 (Table 3). According to Abdul et al. 2013, the protein percentage was recorded 7.08. Another study on date pits discovered protein value of 6.5% as well (Saafi et al., 2008). Thus, it can be said that depending on the cultivar of the date the protein content can be verified.

5.2.3 Ash content

The quantity of ash present in food items can be used to estimate the overall mineral content. Ash content serves as an indicator for food nutrients. From Table 3, it can be easily proved the date seed coffee samples contain higher ash profile (Ajwa date seed coffee, Maryaam date seed coffee, and Mixed date seed coffee 1.18%, 1.16% and 1.16% of Ash respectively) in comparison to the control coffee sample (0.98%). However, the ash value of date seed coffee samples from this present study is similar with the ash content of date seeds 1.18% studied by Nehdi et al., 2010.

5.2.4. Crude fat

Fat aids in providing our bodies with energy, safeguards our organs, promotes cell development, lowers blood pressure and cholesterol, and aids in the body's absorption of essential nutrients. As per laboratory test the values shown in Table 3, it can be depicted that the date seed coffees fat percentage value ranges from 9.85 to 9.96, which is almost higher than Coffee sample (only 4.57%). According to the Hamda et al. 2002, the fat percentage was reported 9% in the date verities found in UAE (Fard). However, a higher percentage of fat (almost 13%) was found in Allig variety (Tunisian) (Besbes et al., 2004a). Abdul et al., 2013 reported that the fat content in date seeds depended on variety, origin, harvesting time and fertilizer.

5.2.5. Crude fiber

Consuming enough fiber might help the body eliminate waste more easily by preventing or treating constipation. Additionally, it promotes a balanced intestinal microbiome. A 2007 review found that dietary fiber improves stool volume, encourages regular bowel movements, and shortens the time waste spends in the intestines (A1-Farsi et al., 2007). According to Abdul et al., 2013, date seeds contain 58 percent total dietary fiber, of which 53 percent is insoluble dietary fiber namely hemicellulose, cellulose, and lignin. Hamada et al., 2002 found a high concentration of lignin and resistant starch in the seeds of three date types (Fard, Khalas, and Lulu) grown in the United Arab Emirates, which included 46–51 percent acid detergent fiber and 65–69 percent neutral detergent fiber. In this study, according to Table 3, the fiber% were found 55.43% which highest in Mixed date seed coffee, followed by 54.72% in Ajwa date seed coffee and 54.38% in Maryaam date seed coffee. The variety and maturation stage are too responsible for these variances in dietary fiber (Hamada et al., 2002).

5.2.6. Carbohydrates

The body receives glucose from carbohydrates, which is then transformed into energy for use during physical activity and maintaining biological processes. However, overabundance of simple carbohydrates might cause us to put on weight. They can also make people more susceptible to high cholesterol, diabetes, and heart disease. Form Table 3, significant difference can be noticed in the quantities of control coffee sample and date seed coffee samples where simple coffee had almost more than two times of total carbohydrates (52.10% CHO) in comparison of the total sugar of palm seeds

(24.51% CHO in Ajwa date seed coffee, 25.44% CHO in Maryaam date seed coffee and 23.87% CHO in Mixed date seed coffee). Similar trend had been reported by Joadder et al., 2007. But a higher range of Carbohydrate content of 35% had been noticed in the study of Amir et al., 2014. Total sugar percentage value largely depends on the maturation stage and variety of dates (Hamada et al., 2002).

5.3 Minerals of Date Seed Coffee

Our bodies require minerals to remain healthy and utilize minerals for a variety of purposes, including maintaining healthy bones, muscles, hearts, and brains. The synthesis of hormones and enzymes depends on minerals. Numerous minerals, including sodium, potassium, magnesium, calcium, phosphorus, iron, manganese, zinc, copper, nickel, cobalt, chromium, lead, and cadmium are shown to be present in date seeds. (Abdillah and Andriani, 2012; Abdul et al., 2013). According to Table 4 of the current study, the date seed coffee samples are more abundant in essential minerals as zinc, iron, copper, and sodium when compared to the control coffee samples.

Mixed date seed coffee has a greater Zinc (Zn) concentration (144.85 μ g/dL) than other coffees. Copper and iron, with values of 112.62 μ g/dL and 44.77 μ g/dL respectively, can be found in Maryaam date seed coffee at the highest concentrations. Among all the samples used in this investigation, Ajwa date seed coffee had the greatest ranges of sodium (47.51 mmol/L) and potassium (1.32 mmol/L).

In earlier investigations, it had been stated that date seeds contain more potassium, phosphorus, magnesium, calcium, and salt than other foods (Al- Hooti et al., 1998; Devshony et al., 1992, Besbes et al., 2004a). Among the microelements, iron is present in higher amounts than manganese, zinc, and copper (Sawaya et al., 1984). The seeds of 11 date cultivars in the Qassim area of Saudi Arabia contained trace quantities (0.19-0.26 percent) of phosphorus, according to Attalla and Harraz, 1996).

5.4 Vitamins of Date Seed Coffee

5.4.1 Vitamin- A

Many foods naturally contain vitamin A, a fat-soluble vitamin. Normal growth and development, the immune system, reproduction, and eyesight ultimately require on vitamin A. Heart, lungs, and other organs keep functioning properly with the aid of vitamin A. Habib et al., (2013) examined the profiles of fatty acids, carotenoids, and fat-

soluble vitamins in the seed oil of 18 date types grown in the United Arab Emirates. The main carotenoid in all 18 date seed oils, ranging from 1.18 mg to 2.68 mg/100g, was beta-carotene, which is like the result (2.59 - 2.77 mg/L of carotene) of this study. Caffeine has found to be interfere the quantity of vitamin A (Wolde, 2014). However, date seed coffee is non-caffeinated, and it contains substantial amount of vitamin A.

5.4.2 Vitamin-C

Ascorbic acid is water-soluble compound that is fundamental for life. Vitamin C plays a crucial role in the cellular chemistry that produces energy, aids in the production of sperm, and is necessary to produce the collagen protein, which is essential for the development and health of cartilage, joints, skin, and blood vessels. Vitamin C is also a significant antioxidant and helps protect against cancers, heart disease, and stress. Date fruits are also good sources of vitamin and contain at least six vitamins (Al-Shahib and Marshall, 2003b). In this present study, the vitamin C content in Control, Ajwa date seed coffee, Maryaam date seed coffee and Mixed date seed coffee were found 2.83 mg/100g, 9.94 mg/100g, 9.69 mg/100g and 9.77 mg/100g respectively. According to the study of Wahini (2016), the flour developed by utilizing the date seeds with sun dried process found vitamin C 23mg/100mg which is much higher. As we know vitamin C is highly heat-sensitive, therefore, it can be said that roasting operation effected the concentration of vitamin C in the date seed coffee samples.

5.5 Phytochemical content

5.5.1 Bio-active contents

Date seeds are an excellent source of phenolic compounds. According to research, polyphenols can maintain healthy blood vessels and assist control blood pressure, which will improve circulation. Another risk factor for heart disease, chronic inflammation, is also decreased by them. Again, our blood sugar levels can be lowered and better controlled by polyphenols (Al-Farsi et al., 2007). Findings of the bio-active ingredient content identified in date seed coffee samples are presented in Table 4.6 where it can be illustrated that Ajwa date seed coffee contains highest among all the samples 210.72 mg QE/100 g of TFC and 180.07 mg GAE/100 g of TPC. Second highest value of TFC and TPC could be reported for the Mixed date seed coffee (197.82 QE/100 g and 130.32 mg GAE/100 g respectively), followed by Mixed date seed coffee (161.66 mg QE/100 g of

TFC and 90.79 mg GAE/100 g of TPC. And lastly, the control coffee scored the lowest with 51.11 QE/100 g of TFC and 16.50 mg GAE/100 g of TPC. Therefore, it can be concluded that amount of Total Polyphenolic Content and Total Flavonoid Content largely depend on the variety of the date. According to Guizani et al. (2014), the polyphenol content of date seeds varied from 21 to 62 mg GAE/g date seed which is much lower than the findings of this study. Distinct date cultivars, date types, and date origins have different bioactive components (Hamada et al., 2002).

5.5.2 Antioxidant capacity

By using the anti-free radical test, coffee extracts made from roasted date seed varieties kernels were assessed for their antioxidant activity (DPPH). Antioxidant capacity (% inhibition) results of Ajwa date seed coffee, Maryaam date seed coffee and Mixed date seed coffee found with values such as 2.74, 1.58 and 1.98 respectively, which were much higher if compared to value of control coffee sample (only 0.11). Therefore, date seed coffees can be recommended as a good source of antioxidants.

A similar finding of higher antioxidant activity in polar extract was reported by Prasad et al., 2009. Although dates are good source of antioxidant like tannins, carotenoids, sterols, and polyphenols had been reported. Studies show that they are good for the heart and may protect against osteoporosis and cancer. Antioxidant potentials varies with different dates cultivars, date type and origin (Hamada et al., 2002).

5.6 Caffeination test by FTIR spectroscopy

Caffeine content in commercial coffee powder usually varied from 20-40%. Here, a caffeine-free coffee substitute was developed using date seeds. *P. dactyliferais* date seed was processed into coffee powder in six steps: washing, draining, drying, roasting, grinding, and sieving. A suitable process for making seed powder is roasting. Before being ground for this investigation, the seeds were roasted at 180°C for 20-30 minutes. The date seed powder's quality is improved during the sieving process, which is also employed for better extraction.

The absence of peaks at 1600 and 1800 cm⁻¹, which demonstrate that this roasted date seed coffee powder contains zero percent caffeine, is used to prove the absence of caffeine using FTIR spectroscopic analysis (Anusree and Kousalya, 2021). People who are sensitive to caffeine who would rather enjoy the distinctive flavor and scent of

caffeine-free coffee without compromising the negative consequences of caffeine and thereby can consume date seed coffee. Our findings demonstrate that date seed powder may be highly recommended as a powerful motivator for people who want to enjoy coffee without increasing their caffeine intake.

5.7 Consumer acceptability of date seed coffee

The three samples of roasted date seed coffee extracts were assessed for their sensory qualities and compared with regular coffee. By boiling 45 g of the powders in 100 mL of water for 2 minutes and adding milk powder corresponding to the powder, beverage preparations were prepared from roasted date seed powders and control coffee. Each panelist's sample presentation was given in a different sequence.

Date seed powder's sensory quality was assessed based on color, flavor, texture, taste, and overall acceptance (Figure 4). Mean sensory score of flavor control's results were highest with7.29, followed by 6.37, 6.9, 6.2 for Ajwa date seed powder, Maryaam date seed powder and Mixed date seed powder respectively. Ajwa date seed powder dominated over Control coffee in terms of mean score of colour and texture. Mean score of colour and texture varied from 7.26 to 7.99 and 6.9 to 7.83 respectively (highest score Ajwa date seed powder). Nonetheless, Ajwa date seed powder also came out on top in terms of overall acceptability for public acceptance.

Sensory quality of date seed coffee based on appearance, taste, flavor and overall acceptability were evaluated (Figure 5). Mean sensory score of taste results were 7.36, 7.73, 6.5 and 6.9 for Control Coffee, Ajwa date seed sample, Maryaam date seed sample and Mixed date seed sample respectively. Highest taste score was observed for Ajwa date jelly.

Mean score of flavors was varied from 6.2 - 7.23 coffee samples. Results of one-way ANOVA revealed that it was statistically significant (p \leq .05) differences in flavor acceptability. Control had highest score in flavour section. Commercially available coffee powder come with additional flavoring agents, whereas the date seed coffee samples were purely maintained throughout the whole procedure. Finally, Overall acceptability score was found the highest for Ajwa date seed coffee (7.83) in relation to control commercial coffee and other remaining date seed coffee samples. According to the hedonic rating scale, the Ajwa date seed coffee has the highest mean score of 7.83,

which means "Like moderately." As a result, it is suitable for widespread consumption as a coffee replacement.

5.8 Cost effectiveness of produced date seed coffee

The manufactured date seed coffee, which has a 500gm serving size, only cost 37 taka (Table 7). If compared to the regular coffee, it would cost an individual about 250 taka (a standard 500gm serving). However, 100 grams of decaffeinated coffee from well-known brands costs around 500tk which in very expensive. Because the procedure to remove caffeine from coffee beans is time- and money-consuming, decaffeinated coffee is more expensive than ordinary coffee. Costly chemicals are frequently used in decaffeination process. By utilizing the date seeds, therefore, people can save money without significant compromising with the taste of regular coffee.

Chapter VI: Conclusion

The food sector currently produces a lot of waste that is frequently not recognized, including date kernels from varieties with limited commercial potential. The ability to use this residue through biotechnological methods, however, represents the best option for using agricultural byproducts and marketing them locally and internationally. The current study involved the production of caffeine - free powder from date seeds and analyzed the caffeine content of those seed powder samples. A coffee-like drink made from powdered update seeds after drying, roasting, and grinding which can be regarded as a bioactive beverage with therapeutic potential. Based on the chemical makeup and sensory assessment of the roasted date palm seeds, it is possible to draw the conclusion that the waste date seeds may be a useful and outstanding source of functional food ingredients and affordable cocoa alternative beverage.

Chapter VII: Recommendations and Future Perspectives

These investigations have produced promising results for the development of date seed coffee. Based on the findings of the current investigation, the following recommendations and research directions are provided for future study. Modern food companies can use the method from medium to large scale production.

- a) It is simple to prepare therefore date seeds can be consumed as coffee substitute.
- b) More revenue may be generated by enhancing the product's taste and therapeutic capabilities.
- c) Since date seeds are an abundant by-product of the date fruit processing business, using them in different food and animal feed would be financially advantageous.
- d) Date seeds are also enriched with various fatty acids. Thus, these seeds can be utilized for producing seed oil.
- e) Adequate actions should be taken to improve the nutritional content of commercially accessible drinks by adding date seeds.

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Appendices

Appendix-A: Preparation of date seed coffee



Date Seeds Samples Collection



Drying of the date seeds before roasting



Date Seed Coffee Powder

Appendix-B: Laboratory Works



Roasting of the seeds



Laboratory testing



Grinding of roasted seeds

Appendix-C: Sensory evaluation



Pictures of the panelists



Appendix-D: Sensory evaluation of date seed powder (hedonic rating test)

| Name: | Product: |
|---------------|----------|
| Panelist No.: | Date: |
| | |

Instructions:

Taste the given samples. Then place an $\sqrt{\text{mark}}$ on the point in the scale which best describes your feelings.

| 5- | | | | | | | | | | | | | | | | | | | | | |
|----|--------------|--------|---|---|-----|-----|---|---|-----|------|---|---|-----|-----|---|---|----|-------|-----|-------|-----|
| | | Colour | | | Fla | vor | • | | Tex | ture | 9 | | Tas | ste | | | Ov | erall | | | |
| | | | | | | | | | | | | | | | | | | ac | cep | tabil | ity |
| | SCORE | 1 | 2 | 3 | Δ | 1 | 2 | 3 | 4 | 1 | 2 | 3 | 4 | 1 | 2 | 3 | 4 | 1 | 2 | 3 | 4 |
| | | 1 | | | | 1 | 2 | 5 | | | 2 | 5 | | 1 | 2 | 5 | | 1 | 2 | 5 | |
| 9 | Like | | | | | | | | | | | | | | | | | | | | |
| | extremely | | | | | | | | | | | | | | | | | | | | |
| 8 | Like very | | | | | | | | | | | | | | | | | | | | |
| | much | | | | | | | | | | | | | | | | | | | | |
| 7 | Like | | | | | | | | | | | | | | | | | | | | |
| | moderately | | | | | | | | | | | | | | | | | | | | |
| 6 | Like | | | | | | | | | | | | | | | | | | | | |
| | slightly | | | | | | | | | | | | | | | | | | | | |
| 5 | Neither like | | | | | | | | | | | | | | | | | | | | |
| | nor dislike | | | | | | | | | | | | | | | | | | | | |
| 4 | Dislike | | | | | | | | | | | | | | | | | | | | |
| | lightly | | | | | | | | | | | | | | | | | | | | |
| 3 | Dislike | | | | | | | | | | | | | | | | | | | | |
| | moderately | | | | | | | | | | | | | | | | | | | | |
| 2 | Dislike very | | | | | | | | | | | | | | | | | | | | |
| | much | | | | | | | | | | | | | | | | | | | | |
| 1 | Dislike | | | | | | | | | | | | | | | | | | | | |
| | extremely | | | | | | | | | | | | | | | | | | | | |

Here, 1 = Control, 2 = Ajwa Date Seed Powder, 3 = Maryaam Date Seed Powder, 4 = Mixed Date Jell Seed Powder.

Hedonic scale scoring test results for Control, Ajwa date seed powder, Maryaam date seed powder and Mixed date seed powder:

| Parameters | Control | Ajwa Date | Maryaam Date | Mixed Date |
|-----------------------|---------------------------------|---------------------------------|-------------------------|---------------------------------|
| | | Seed Powder | Seed Powder | Seed Powder |
| Colour | 7.29 <u>+</u> 0.58 ^b | 7.99 <u>+</u> 0.40 ^a | 7.27±0.39 ^b | 7.26 ±0.32 ^b |
| Flavour | 7.29±0.66 ^a | 6.37 <u>±</u> 0.49 ^c | 6.90 ± 0.30^{b} | 6.20 ± 0.36^{d} |
| Texture | 7.33 ±.30 ^b | 7.83 <u>+</u> 0.15 ^a | $7.00 \pm 0.40^{\circ}$ | 6.90 ± 0.25^{d} |
| Taste | 7.16±0.20 ^c | 7.93±0.30 ^a | 7.5 ± 0.50^{b} | $7.20 \pm 0.29^{\circ}$ |
| Overall acceptability | 7.23 ± 0.30^{b} | 7.73 <u>±</u> 0.15 ^a | $7.09 \pm 0.40^{\circ}$ | 6.98 <u>±</u> 0.25 ^d |

Legends: The table contained all values as (ME \pm SD), where ME stands for Mean and SD for Standard Deviation. The superscripts a, b, c, and d indicate significant differences (p \leq .05) across samples.

Appendix-E: Sensory evaluation of date seed coffee (hedonic rating test)

| Na | Name: | | | | | | | | Product: | | | | | | | | |
|----|------------------|-----|--------|------|-------|------|-----|-------|----------|------|---------|-------|-------|-------|--------|-------|-------|
| Pa | nelist No.: | | | | | | | | | Da | te: | | | | | | |
| In | Instructions: | | | | | | | | | | | | | | | | |
| Та | ste the given sa | amp | les. T | Then | place | e an | √ma | ark o | n the | poir | nt in t | he sc | ale v | vhich | n best | desci | ribes |
| yo | ur feelings. | | | | | | | | | | | | | | | | |
| | | A | ppe | aran | ce | | Fla | avor | | | Та | ste | | | Ove | erall | |
| | | | | | | | | | | | | | | a | ccept | abili | ty |
| | SCORE | 1 | 2 | 3 | 4 | 1 | 2 | 3 | 4 | 1 | 2 | 3 | 4 | 1 | 2 | 3 | 4 |
| | - 11 | | | | | | | | | | | | | | | | |
| 9 | Like | | | | | | | | | | | | | | | | |
| | extremely | | | | | | | | | | | | | | | | |
| 8 | Like very | | | | | | | | | | | | | | | | |
| | much | | | | | | | | | | | | | | | | |
| 7 | Like | | | | | | | | | | | | | | | | |
| | moderately | | | | | | | | | | | | | | | | |
| 6 | Like | | | | | | | | | | | | | | | | |
| | slightly | | | | | | | | | | | | | | | | |
| 5 | Neither | | | | | | | | | | | | | | | | |
| | like nor | | | | | | | | | | | | | | | | |
| | dislike | | | | | | | | | | | | | | | | |
| 4 | Dislike | | | | | | | | | | | | | | | | |
| | lightly | | | | | | | | | | | | | | | | |
| 3 | Dislike | | | | | | | | | | | | | | | | |
| | moderately | | | | | | | | | | | | | | | | |
| 2 | Dislike | | | | | | | | | | | | | | | | |
| | very much | | | | | | | | | | | | | | | | |
| 1 | Dislike | | | | | | | | | | | | | | | | |
| | extremely | | | | | | | | | | | | | | | | |

Here, 1 = Control, 2 = Ajwa Date Seed Coffee, 3 = Maryaam Date Seed Coffee,

4 = Mixed Date Seed Coffee.

Hedonic scale scoring test results for Control, Ajwa date seed coffee, Maryaam date seed coffee and Mixed date seed coffee:

| Parameters | Control | Ajwa Date | Ajwa Date Maryaam Date | | |
|-----------------------|---------------------------------|---------------------------------|---------------------------------|-------------------------|--|
| | | Seed Coffee | Seed Coffee | Seed Coffee | |
| Appearance | 7.33 <u>±</u> 0.15 ^a | 7.12 <u>±</u> 0.40 ^b | 6.43 <u>+</u> 0.25 ^d | $6.90 \pm 0.35^{\circ}$ | |
| Flavour | 7.23 <u>±</u> 0.50 ^a | 6.50 ±0.40 ^c | 6.90 ± 0.30^{b} | 6.20 ±0.36 ^d | |
| Taste | 7.36 <u>+</u> 0.20 ^b | 7.73 <u>+</u> 0.30 ^a | 6.50 ± 0.50^{d} | $6.90 \pm 0.29^{\circ}$ | |
| Overall acceptability | 7.33 ± 0.30^{b} | 7.83 <u>+</u> 0.15 ^a | $7.00 \pm 0.40^{\circ}$ | 6.90 ± 0.25^{d} | |

Legends: The table contained all values as (ME \pm SD), where ME stands for Mean and SD for Standard Deviation. The superscripts a, b, c, and d indicate significant differences (p \leq .05) across samples.

Appendix F: Experimental data

A) Bio-active compounds data:

| Parameters | Control | Ajwa Date | Maryaam Date | Mixed Date |
|------------------|---------|-------------|--------------|-------------|
| | | Seed Powder | Seed Powder | Seed Powder |
| | | | | |
| Total phenolic | 16.504 | 180.030 | 90.765 | 130.402 |
| content (TPC) | 16.663 | 180.090 | 90.754 | 130.556 |
| | 16.455 | 180.112 | 90.458 | 130.331 |
| | 51.668 | 210.835 | 161.992 | 197.830 |
| Total flavonoids | 50.114 | 210.554 | 161.448 | 197.776 |
| content (TFC) | 51.554 | 210.778 | 161.559 | 197.866 |
| Antioxidant | 0.115 | 2.771 | 1.579 | 0.454 |
| capacity (DPPH) | 0.112 | 2.696 | 1.678 | 0.424 |
| | 0.119 | 2.779 | 1.508 | 0.415 |

B) Physicochemical and Proximate Analysis Data:

| | Control | Ajwa Date | Maryaam | Mixed Date |
|-----------------------|---------|-----------|-------------|-------------|
| Parameters | (Ama | Seed | Date | Seed Powder |
| | Coffee) | Powder | Seed Powder | |
| p ^H | 4.6 | 4.5 | 4.4 | 4.4 |
| | 4.5 | 4.6 | 4.6 | 4.4 |
| | 4.6 | 4.5 | 4.5 | 4.3 |
| TSS(°Brix) | 1.8 | 2 | 2 | 2 |
| | 1.7 | 2 | 1.9 | 2 |
| | 1.8 | 1.9 | 2 | 2 |
| Titratable acidity (% | 1.25 | 1.11 | 1.12 | 1.12 |
| Ascorbic Acid) | 1.19 | 1.13 | 1.09 | 1.11 |
| | 1.21 | 1.14 | 1.11 | 1.12 |
| | 0 | 17.15 | 16.59 | 16.99 |
| Vitamin-A(RAE) | 0 | 17.09 | 16.55 | 16.93 |
| | 0 | 17.12 | 16.54 | 16.96 |
| Vitamin-C | 2.87 | 9.91 | 9.70 | 9.79 |

| (mg/100g) | 2.79 | 9.97 | 9.68 | 9.78 |
|------------------|------|-------|-------|-------|
| | 2.85 | 9.95 | 9.70 | 9.77 |
| Moisture (%) | 5.46 | 2.09 | 2.17 | 2.22 |
| | 5.53 | 2.11 | 2.15 | 2.21 |
| | 5.49 | 2.08 | 2.16 | 2.24 |
| Protein (%) | 6.95 | 7.59 | 6.98 | 7.35 |
| | 6.93 | 7.65 | 6.99 | 7.39 |
| | 6.98 | 7.62 | 6.94 | 7.40 |
| Ash content (%) | 0.99 | 1.18 | 1.16 | 1.15 |
| | 1.01 | 1.17 | 1.19 | 1.19 |
| | 0.96 | 1.19 | 1.15 | 1.16 |
| Crude Fat (%) | 4.50 | 9.94 | 9.97 | 9.84 |
| | 4.67 | 9.91 | 9.98 | 9.87 |
| | 4.55 | 9.89 | 9.94 | 9.86 |
| Crude Fiber (%) | 0 | 54.67 | 54.32 | 55.44 |
| | 0 | 54.78 | 54.45 | 55.37 |
| | 0 | 54.71 | 54.37 | 55.49 |
| Carbohydrate (%) | 82.1 | 24.53 | 25.40 | 24.00 |
| | 81.9 | 24.57 | 25.49 | 24.09 |
| | 82.3 | 24.45 | 25.45 | 23.54 |
| | | | | |

C) Minerals Data:

| Parameters | Control | Ajwa Date | Maryaam Date | Mixed Date |
|----------------------------|---------|-------------|--------------|-------------|
| | | Seed Powder | Seed Powder | Seed Powder |
| Sodium (Na) in mmol/L | 21 | 47.5 | 46.1 | 46.6 |
| | 20.99 | 47.44 | 45.87 | 46.81 |
| | 21.76 | 47.60 | 46.23 | 46.22 |
| Phosphorus (P) in mg/dL | 0.12 | 2.50 | 2.10 | 2.40 |
| | 0.11 | 2.45 | 2.15 | 2.35 |
| | 0.12 | 2.60 | 2.12 | 2.45 |
| Copper (Cu) in | 8.03 | 108.97 | 112.47 | 111.30 |

| µg/dL | 8.22 | 108.31 | 112.56 | 111.67 |
|--------------------|-------|--------|--------|--------|
| | 7.98 | 108.45 | 112.85 | 111.38 |
| Chloride (Cl) in | 0.16 | 3.32 | 3.41 | 3.30 |
| mmol/L | 0.15 | 3.18 | 3.47 | 3.45 |
| | 0.16 | 3.26 | 3.39 | 3.19 |
| Magnesium (Mg) | 0.09 | 0.78 | 0.69 | 0.70 |
| m mg/aL | 0.8 | 0.79 | 0.70 | 0.69 |
| | 0.09 | 0.75 | 0.68 | 0.71 |
| Potassium (K) in | 2.84 | 1.31 | 1.20 | 1.10 |
| mmol/L | 2.78 | 1.35 | 1.26 | 1.11 |
| | 2.88 | 1.32 | 1.19 | 1.14 |
| Iron (Fe) in μg/dL | 20.08 | 44.59 | 44.84 | 44.60 |
| | 20.19 | 44.73 | 44.78 | 44.69 |
| | 20.04 | 44.54 | 44.69 | 44.58 |
| Calcium (Ca) in | 0.44 | 0.01 | 0.01 | 0.01 |
| mg/dL | 0.48 | 0.01 | 0.01 | 0.01 |
| | 0.41 | 0.01 | 0.01 | 0.01 |
| Zinc (Zn) in | 31.13 | 141.30 | 145.45 | 144.90 |
| μg/dL μg/dL | 31.25 | 141.67 | 145.39 | 144.87 |
| | 31.19 | 141.54 | 145.55 | 144.79 |

Appendix G: FTIR Spectrum

FTIR Spectrum of Control, Ajwa Date Seed Coffee, Maryaam Date Seed Coffee and Mixed Date Seed Coffee respectively.



Brief Biography

Sowmik Roy Rathi graduated with a B.Sc. (Hons.) in Food Science and Technology from the Chattogram Veterinary and Animal Sciences University (CVASU), in Chattogram, Bangladesh, with a 3.50 cumulative grade point average on a 4.00 scale. He previously completed the Higher Secondary Certificate (HSC) Examination in 2013 with a GPA of 4.80/5.00 after passing the Secondary School Certificate (SSC) Examinations in 2011 with a Grade Point Average (GPA) of 5.00/5.00. He briefly declared himself a candidate for the MS in Applied Human Nutrition and Dietetics degree under Department of Applied Food Science and Nutrition, the Chattogram Veterinary and Animal Sciences University (CVASU), Bangladesh. He has a strong passion for sharing information and academic research, and he would appreciate any further opportunity to do so.