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Development of a coffee substitute based on date pits .

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Dedication

I wanna thank **my sunshine mom** for

being with me step by step

And wanna thank **my adorable sisters**

And **my good brother**

And anyone support me to keep going

Thanks to **my sweet friends**

Last but not least thanks to **me** for not stoping

Alhamdulillah

Dedication

All praise is due to Allah, first and foremost. Every success is by His grace, and every step is guided by Him.
To the one who has always been my home and my strength...
To the one who planted hope in me, and whose prayers reached the heavens...
To **my mother**, the symbol of love and sacrifice , I dedicate this achievement with deepest gratitude and love.

To the one who taught me that nothing is impossible with determination...
To **my fathe**r, the guiding light of my path and the source of my strength ,thank you from the heart.

To the heartbeat of my soul and my eternal support,
To my beloved sisters: **Ibtissem, Imane, Sidra** , you are my light in dark times.

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To the ones who walked beside me through the struggle, who fought and persevered with me,
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To my pillar and the partner of my dreams,
Thank you for your patience, and for always ecncouraging me.

Table of Contents

List of abbreviations

TPC: total polyphenols content**.**

RS: reducing sugars.

OM %: Organic Matter.

DM (%): Dry Matter Content.

HPLC: High-Performance Liquid Chromatography.

GPC: Gaz Phase Chromatography.

%: Percentage.

°C: Degrees centigrade.

pH: Potential hydrogen.

C: Concentrations.

g: Grams.

H (%): Moisture content percentage.

DP:date pits .

R:coffe Robusta.

A: Less roasted date pits powder.

F:more roasted date pits powder.

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INTRODUCTION

General introduction

Coffee is the second most consumed product in the world after oil. According to the International Coffee Organization) **[1],** the global annual coffee production is estimated at around 9.68 billion kilograms, equivalent to 159 million 60-kg bags over the past ten years.

Coffee remains one of the most traded commodities, with a steady growth in its commercialization **[2].** However, the waste generated by the coffee industry consists of harmful substances that can cause serious environmental issues, including water pollution, soil degradation, and health risks to humans **[3]**.

These wastes include various by-products such as skin, pulp, mucilage, parchment, silver skin, and defective coffee beans **[4]**.

Caffeine is a substance that promotes temporary alertness and stimulates the central nervous system by providing energy. It is a legal drug, though not strictly regulated, and is available in various forms, consumed daily. It is also the most popular drug worldwide **[5].**

Caffeine is a psychoactive substance that has been associated with negative health outcomes, including high blood pressure **[6]**.

People who consume a substance on a daily basis should be aware of the associated risks, and it is recommended to avoid caffeine. Roasted date seeds can be ground and used as a substitute for those who typically consume more coffee.

The date palm (Phoenix dactylifera) is a crucial plant for the survival of populations in the Saharan regions. Algeria, with its rich and diverse date palm heritage, is home to more than 18 million trees across 940 cultivars, ranking 6th globally with a total date production of 1,040,000 tons.

The date palm produces a variety of by-products, including leaves, pits, trunk, and pedicels, which are utilized in various applications. Date pits, in particular, result from several processing methods such as the production of pitted dates, date paste, date syrup, and date juice **[7].**

Date pits are a rich source of proteins, fats, ashes, and dietary fiber **[8]**. They also contain high levels of phytochemicals such as phenols, sterols, carotenoids, anthocyanins, procyanidins, and flavonoids **[9]** . The nutritional value of these pits is not only determined by their composition but also by the accessibility and digestibility of their

components. Although the hard structure of date pits may pose a challenge to optimizing their nutritional value, they can be ground into powder **[10],** which allows them to be used to enhance the nutritional value of food products in which they are incorporated **[11].**

Roasted and ground date pits are used by some rural communities as a caffeine-free coffee substitute and in coffee-like preparations, particularly in Arab markets **[12].**

In this context, our contribution focuses on the valorization of plant waste, specifically date pits, as a coffee substitute. This valorization involves studying the main physicochemical characteristics of date pit powders and comparing them to those of coffee, as well as examining the effect of roasting on these characteristics. Additionally, we investigate the impact of various roasting processes on the quality of the coffee made from date pits.

The study is organized into three complementary chapters. The first chapter presents a literature review highlighting the characteristics of dates, the date palm, date seeds, and the powdered coffee derived from these seeds. The second chapter outlines the main methods used for physical-chemical, biochemical, microbiological, and sensory analyses. The third chapter presents the key findings of the study, followed by a detailed discussion. Finally, a general conclusion and future perspectives, offering directions for further research, bring the work to a close.

Chapter I
Literature Review

I.General overview of the date palm and dates

I.1.The date palm

Phoenix dactylifera L derives its name from the term "Phoenix," meaning date palm, and "dactylifera," which comes from the Greek word "dactylos," meaning finger, due to the resemblance of its fruit to the shape of a finger **[13]** This species is particularly well-suited to arid and semi-arid climates, where it is widely cultivated, notably in the Algerian Sahara, Saudi Arabia, Iran, and Egypt **[14]** The date palm is a dioecious plant, with male plants (dokkar) and female plants (nekhla) **[15]**The botanical classification of the date palm is shown below:

Clade	Commelinids
Order	Arecales
Family	Arecaceae
Genus	Phoenix
Species	Phoenix dactylifera L

Table 01: The botanical classification of the date **[16]**

The genus Phoenix includes at least twelve species, among which Phoenix dactylifera is the most renowned. Its fruit, the "dates," plays a major role in international trade **[17]**

I.2.The date Fruit

I.2.1.Definition and description of the date fruit

The date is a type of berry fruit, generally elongated or rounded in shape. Its size varies significantly depending on the variety, with lengths ranging from 2 to 8 cm and weights between 2 and 8 grams**[13]** . It consists of an edible portion known as the pulp or flesh, and a hard, inedible pit. The flesh of the date is composed of three main layers:

The epicarp: a thin, cellulose-rich skin.

The mesocarp: a fleshy layer of varying thickness and texture, depending on the variety.

The endocarp: a parchment-like membrane, lighter in color and fibrous in texture that directly surrounds the pit **[13]**

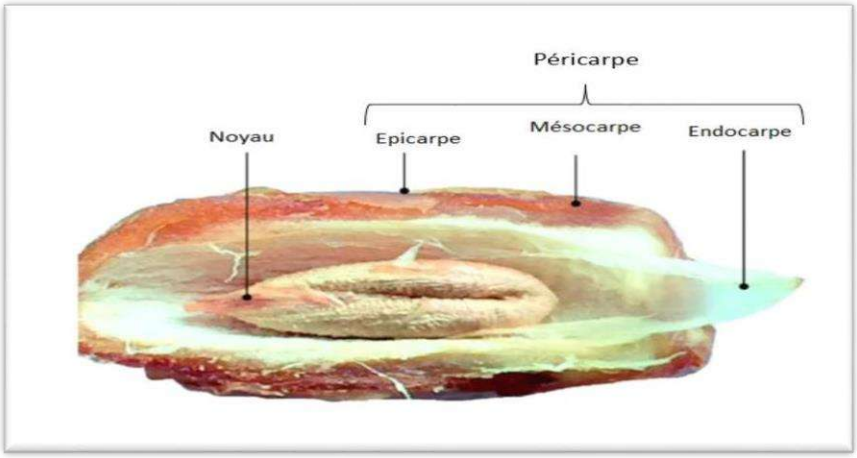


Figure 1: Longitudinal section of a date fruit [18]

Dates exhibit a wide range of colors, influenced by both their stage of ripeness and their variety. In the immature stage, they are typically green, yellow, or orange. As they ripen, their color changes to shades ranging from translucent brown to black, and in some cases, yellowish **[19]**

I.2.2.Botanical and Systematic Classification

Dates are classified into three categories based on their texture: soft, semi-soft, and dry **[13]**

Consist -ency	Characteristics	Varieties	picture
Soft	A moisture content equal to or greater than 30% indicates a high concentration of inverted sugars, particularly fructose and glucose.	Ghars (Algeria)	
Semi-soft	A moisture content ranging from 20% to 30%.	The Deglet-Nour variety (Algeria).	

Dry	They are rich in sucrose when their moisture content is below 20%.	Variety Degla Beida and Mech Degla	

Table02. Classification of dates based on consistency and characteristics **[20]**

I.2.3.Biochemical Composition of the Date Fruit

According to**[21]** , the date fruit is primarily composed of the following elements (Fig. 02): Water, Sugars, Sucrose (non-reducing sugar), Glucose and fructose further to the non-sugar components, which are proteins, lipids, cellulose, ash (mineral salts), vitamins, and enzymes.

Compostions	**Content** (g/100g)
Water	7.2-50.4
Total sugars	52.6-88.6
Glucose	17.6-41.4
Fructose	13.6-36.8
Sucrose	0.5-33.9
Lipids	0.1-1.4
Proteine	1.1-2.6
Fiber	3.53-10.9

Table03: Chemical Composition of Dates ***[22]***

I.2.4.Mineral Elements

According to ***[22]*** dates are rich in mineral elements. The main minerals they contain include potassium, phosphorus, calcium, and magnesium (see Table 2).

Mineral Elements	**Content (mg/100g)**	**Mineral Elements**	**Content (mg/100g)**
Potassium	354-1287	Copper	0.01-0.8
Sodium	1-261	Iron	0.10-1.5
Calcium	5-206	Zinc	0.02-0.6
Magnesium	31-105	Manganése	0.01-0.4
Phosphorus	35-74		

Table 04: Mineral Element Content of Dates ***[22]***

I.3.Date pits

I.3.1.Definition and description of the date pits

The date pit, also known as the seed of the date palm, is the hard, inedible part of the fruit, accounting for approximately 7% to 30% of the total weight of the date ***[17]*** It is elongated in shape and varies in size **(Figure 2)**. The pit is enclosed by a parchment-like endocarp, which may be smooth or have lateral ridges or wing-like projections, with a ventral groove (**Figure3)**.

Inside the pit is a hard, horny albumen (endosperm), while the dorsal embryo, which is always very small compared to the albumen, measures between 2 and 3 mm Date pits (DP) have no distinct odor, range in color from light to dark brown, and have a neutral taste with a slightly bitter note ***[22]***

Figure 2: The Different Shapes of Date Pits***[23]***

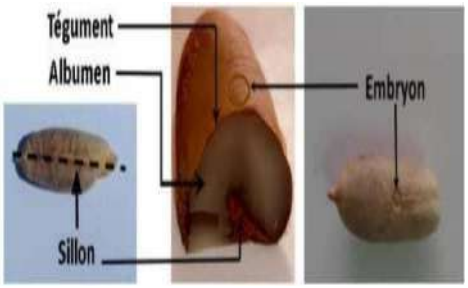


Figure3:Date palm seed ***[24]***

I.3.2.Chemical Composition of Date Pits

-The moisture content: According to ***[25] ,*** the moisture content of the date pit, which remains internally humid, ranges between 7% and 19%.

-The ash: According to several authors, the ash content of date pits remains low, ranging from 0.89% to 1.30% of dry matter ***[21]***

-Fat: Date pits are recognized for their richness in fats, encompassing a diverse profile of saturated and unsaturated fatty acids. Their lipid content varies between 3.01% and 13.5%.

-Fiber: According to results obtained from various cultivars, date pits have a very high fiber content (both crude and dietary), ranging from 71% to 94% . Moreover, their levels of soluble pectin (0.67%), crude pectic acid (3.12%), protopectin (1.43%), and total pectin (3.21%) exceed those found in date pulp, which are reported at 0.51%, 2.65%, 1.02%, and 2.77% respectively ***[17]***

-Sugar Content: Date pits contain both reducing and non-reducing sugars. Several studies have highlighted the carbohydrate richness of date by-products The total sugar content, as well as the proportions of reducing sugars and sucrose, vary depending on the variety, with total sugars ranging from 4.4% to 4.6% and reducing sugars around 2.2% Date pits represent a local, easily accessible, and low-cost biomass. "Their potential uses are substantial and could appeal to various sectors, such as the food, cosmetic, and pharmaceutical industries**.**

-Protein: Date pits contain proteins in varying amounts, influenced by the region of cultivation and the cultivar. ***[21]*** reported protein contents ranging from 2.3% to 6.0%. Overall, these findings confirm that date pits are a valuable source of protein, regarded as important biomolecules.

-Polyphenols: Date pits contain polyphenols, which are major secondary metabolites.Their content varies by variety, ranging from 4.34% to 33.97% according to different studies ***[21]*** identified several phenolic acids, including gallic, caffeic, and vanillic acids. The concentration of phenolic compounds also depends on the maturation stage, region, sun exposure, and storage conditions. These compounds contribute to stress resistance through the production of phytoalexins (stilbenoids) ***[15]*** Preliminary phytochemical analysis revealed significant amounts of total phenolic compounds (38.8 mg gallic acid equivalents) and total flavonoids (87.86 mg rutin equivalents) in date pit powder

I.4.Valorization of date pits

Date pits represent a local, easily accessible, and low-cost biomass. "Their potential uses are substantial and could appeal to various sectors, such as the food, cosmetic, and pharmaceutical industries", date pits can be incorporated into human food: after roasting, they can replace coffee, providing an infusion with a pleasant taste and aroma (similar to decaffeinated coffee). They are mainly used as animal feed, with a nutritional value comparable to that of a kilogram of barley. Therefore, they represent a highly valuable by-product.

I.5.Health Benefits of Date Pits

I.5.1.Antioxidant Properties

Thanks to their richness in antioxidants and strong free radical scavenging activity, date pits help reduce damage caused by oxidative stress. They can be used as a functional food ingredient to support and improve metabolic functions ***[22]***

I.5.2.Reduction of Blood Sugar Levels

Blood glucose-related disorders, particularly diabetes and its complications, can be alleviated with date pit treatment. Recent studies have shown protective effects against early diabetic complications affecting the liver and kidneys ***[22]***

I.5.3.Prevention of DNA Damage

the antioxidant and free radical-scavenging properties of date pits help protect the liver by preventing chemical-induced hepatic lesions and oxidative DNA damage, thereby aiding in the prevention of liver intoxication***[22]***

I.5.4.Antiviral Agents

Date pits possess antiviral properties that assist in the treatment and prevention of various viral infections. Date extracts have demonstrated a strong ability to enhance the efficacy of the Pseudomonas phage and completely prevent bacterial lysis ***[22]***

I.5.5Protection of the Kidneys and Liver

Due to their high content of proanthocyanidins, date pits have the ability to protect both kidneys and liver from damage. Extracts of these compounds have been shown to prevent nephrotoxicity and hepatotoxicity caused by chemical agents ***[22]***.

I.6 Date pit coffee substitute

I.6.1 Definition and description

Roasted date pit powder is a fine, brown-colored powder that is caffeine-free and rich in phenolic compounds. Its flavor profile largely depends on the date variety used. It offers a mild taste and a pleasant aroma**.** The beverage made from date pits is generally well accepted, although it is slightly lower in quality compared to traditional Arabic coffee. It is characterized by reduced bitterness, a lighter color, and a milder coffee aroma ***[18].***

I.6.2 Transformation process

Cleaning the pits: Date pits are carefully cleaned to remove any impurities or residues. ***[23].*** **Drying:** The pits are then dried at room temperature. ***[23].*** They are dried to remove excess moisture before proceeding to roasting. ***[18].***

Roasting: The date pits are roasted at a temperature of 200 °C for 20 minutes ***[18].*** This process also helps to release the aromatic compounds.

Grinding: After roasting, the date pits are ground using an electric grinder ***[21].***

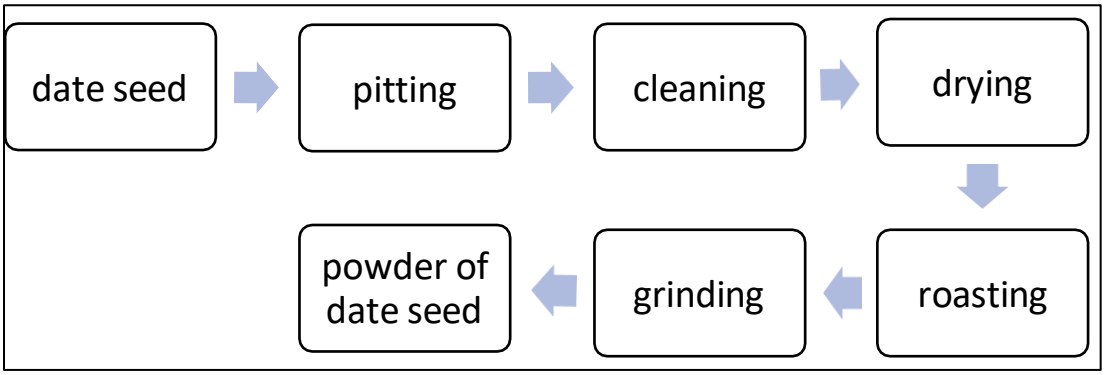


Figure 4: Preparation diagram of Ghars date kernel powder.

I.6.3.Description of seed powder

Category	Details
Type of food	Flour ***[18].***
Part used	Roasted and ground date seeds ***[22].***
Sensory analysis	**Texture**: smooth, homogeneous, non-granular ***[23].*** **Aroma**: slightly caramelized **Taste**: mild, slightly bitter **Dominant flavor**: slightly sweet ***[22].*** **Color**: light to dark browen ***[18].***
Chemical state	Rich in fiber and antioxidants ***[22].***
Nutritional value	Fibres, antioxidants, cafeine **free** ***[23].***
Usage	Beverage praparation (decaffeinated coffee subtitute) ***[24].***
Physical form	Fine, dry powder, brown color ***[18].***

I.6.4.Coffee Coffee is a beverage prepared by infusing roasted seeds from the coffee plant. Global coffee production mainly relies on two species: Coffea arabica and Coffea robusta Chemically, coffee contains over 1,000 bioactive compounds, including caffeine, chlorogenic acids, diterpenes, and trigonelline. These compounds are responsible for its unique aroma, characteristic taste, and physiological effects such as stimulating and antioxidant properties

Figure 5: Types of Coffee Beans: Arabica and Robusta ***[26].***

Chapter II

Material & methods

II.Material & methods

1.Sampling

In this study, the dates used belong to the **Ghars** variety. They were purchased from the local market in Touggourt during the 2024/2025 season. The coffee sample, used as a reference standard, is of the *Robusta* variety originating from Indonesia. It was purchased from the local market of Ouargla in the 2025 season.

Figure 6: Date palm variety: Ghars

2.Transformation process

To achieve the objectives of this study, three distinct samples were selected based on their type or production conditions, as described below:

-The **reference sample** is a Robusta coffee originating from Indonesia.

Date pits were divided into two samples according to roasting time :

Sample 1: roasted at 200 °C for 25 to 30 minutes.

Sample 2: roasted at 200 °C for 10 to 15 minutes.

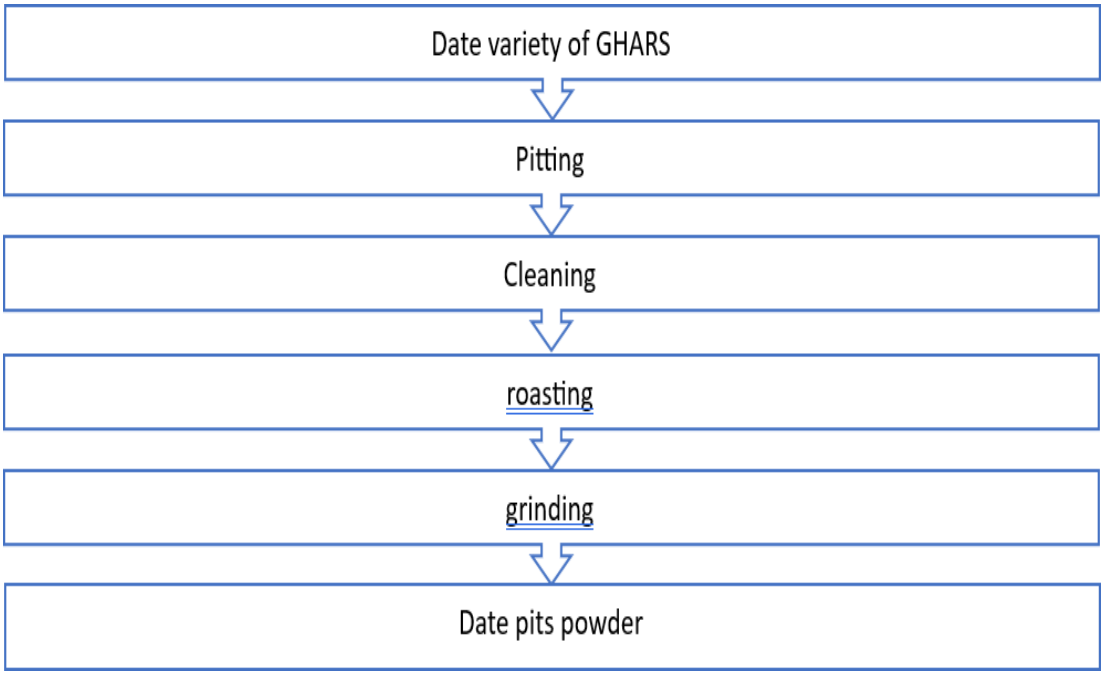


Figure 7: Manufacturing diagram of Ghars date kernel powder

Figure 9: less date pits before grinding

Figure 8: date pits before roasting

Figure 10: more date pits before grinding

Figure 11: Less roasted date pits powder

Figure 12: More roasted date pits powder

3.Physical-chemical Analyses

3.1Determination of pH (Annex 6)

pH represents the activity of hydrogen ions (H^+) in a solution and thus indicates the acidity or alkalinity of a medium. It is measured using a pH meter. In food products, the pH depends both on the concentration of ionizable acids (which release H^+ ions) and on the presence of mineral compounds, such as hydroxyl ions (OH^-), which can influence the chemical balance**.** The pH of the sample is measured using a pH meter, following the procedure described by [27]. A portion of the sample is first diluted with distilled water, then the mixture is filtered using filter paper prior to analysis.

3.2.Moisture Content Determination (Annex 6)

The moisture content of date pits is assessed using the standardized method, in accordance with as described by This analysis aims to quantify the amount of moisture present in the pits. Moisture plays a significant role in lipid degradation, particularly by promoting the hydrolysis of fatty acids during storage. Therefore, determining this parameter helps to anticipate potential deterioration of the sample due to moisture.The moisture content of the dates was assessed by drying a 5-gram sample in an oven at 105 °C until a constant weight was achieved, in accordance with the method described by [28] The moisture content was then calculated based on the weight loss observed during the drying process.

$$H\ (\%) = 100 \times (M1 - M2)/P$$

Moisture Content Calculation Variables:

H (%): Moisture content percentage.

M_1: Mass of the container plus the fresh sample before drying (in grams).

M_2: Mass of the container plus the sample after drying (in grams).

P: Mass of the test sample (5 gram).

3.3.Determination of Dry Matter Content (Annex 6)

The dry matter content of a food product refers to the residual mass remaining after the removal of moisture. This is achieved by drying the sample in an oven at 105 °C until a constant weight is obtained.

The dry matter content is calculated using the following formula:

$$\text{DM (\%)} = [(\text{Dry weight}) / (\text{Fresh weight})] \times 100$$

3.4.Ash Content (Annex 6)

Ash refers to the mineral fraction remaining after the incineration of the dried residue of the product. It reflects the mineral richness of date pits. To determine this, the date pits was incinerated in a muffle furnace at **600** °C for **3** hours, until a white or grey coloration appeared, indicating complete combustion [28].

The ash content is then calculated using the following formula:

$$\text{Ash} = 100 - \text{OM \%}$$

$$\text{OM \%} = 100 \times (M1 - M2) / P$$

Where:

OM %: Organic Matter

M_1: Mass of the crucible + test sample (g)

M_2: Mass of the crucible + ash (g)

P: Mass of the test sample (g)

3.5.Titratable Acidity

Titratable acidity is determined by titration with a 0.1 N NaOH solution in the presence of phenolphthalein as an indicator**.** The analysis is carried out on a suspension prepared by dissolving 10 g of date pits powder in 100 ml of distilled water. The acidity is expressed in grams of citric acid per 100 g of product and serves as an indicator of date pits quality [10].

The total acidity is calculated using the following formula:

Where:

$$\text{Acidity (\%)} = (250 \times V1 \times 100 \times 0.07) / (V0 \times M \times 10)$$

M is the mass in grams of the sample taken,

V0 is the volume in milliliters of the sample used,

V1 is the volume in milliliters of the 0.1 N NaOH solution,

0.07 is the conversion factor for titratable acidity to citric acid equivalent.

3.6.Fibers: (Annex 6-7)

The Van Soest method

Principle: Based on the use of neutral and acid detergents to fractionate plant cell wall constituents into different fiber fractions [29].

NDF (Neutral Detergent Fiber):

Contains cellulose + hemicellulose + lignin

Allows the separation of cell walls (total fibers) from soluble cell contents.

ADF (Acid Detergent Fiber):

Contains cellulose + lignin

Hemicellulose = NDF – ADF

Lignin (ADL):

Obtained by treating the ADF residue with concentrated sulfuric acid (72% H_2SO_4).

Cellulose = ADF – ADL

Method for determining pectin (gravimetric method by alcoholic precipitation)

Principle: Pectin is extracted with hot acidified water and then precipitated with alcohol. The precipitate is filtered, dried at moderate temperature, and then weighed to determine its content [30].

Pectin content (g/100 g sample) = (Mass of dried pectin (g)/Mass of initial sample (g)) ×100

4.Biochemical Analysis

4.1. Lipid Determination: (Annex 6-7)

Soxhlet extraction, which has been used for a long time, is a standard technique and the primary reference for evaluating the performance of other solid-liquid extraction methods [31] This method was first developed by Franz von Soxhlet in 1879, originally for determining lipid content. After extraction, the lipids undergo transesterification with methanol, converting the lipid extracts into fatty acid methyl esters (FAMEs). A final analysis is then conducted using gas chromatography with flame ionization detection (GC-FID) for quantification.

4.2. Protein Determination: Kjeldahl Method (Annex 6-7)

This reference method involves the complete mineralization of organic molecules, converting the nitrogen they contain into ammonia, which can then be quantified using various techniques. The subsequent steps include titration, colorimetry, and potentiometry [32].

4.3. Determination of Total Polyphenol Content (Annex 6-2)

The Folin-Ciocalteu method is used to quantify the total polyphenol content in the extracts. This reagent, composed of a mixture of phosphomolybdic and phosphotungstic acids, is reduced by phenolic compounds present in the sample. The resulting chemical reaction produces a blue coloration due to the formation of tungsten and molybdenum oxides. The

intensity of this color, measured at 760 nm, is directly proportional to the polyphenol concentration [33]. An aliquot of 100 µl of the extract is added to 500 µl of Folin-Ciocalteu reagent diluted 1:10, then supplemented with 2 ml of a 20% sodium carbonate solution. The mixture is incubated away from light for 30 minutes. Absorbance is measured at 760 nm with a UV-visible spectrophotometer. The phenolic content is calculated based on a calibration curve established with gallic acid, and results are expressed in mg of gallic acid equivalents per 100 g of date seed powder.

4.3.1 .Flavonoid Quantification (Annex 6-2)

The aluminum chloride method is used to determine the flavonoid content in date pits extracts. This method is based on the formation of a stable yellow complex between flavonoids and aluminum chloride, which can be measured using UV-visible spectrophotometry at 410 nm.

One milliliter of the extract is mixed with 1 ml of a 2% aluminum chloride solution. After 10 minutes of incubation, the absorbance is recorded at 410 nm [34]. The flavonoid concentration is then calculated from a calibration curve prepared with rutin and expressed as milligrams of rutin equivalents per 100 g of date seed powder.

4.4.Quantitative Sugar Determination by the Dubois Method (1956) (Annex 6-4)

Total sugars were measured according to the method developed by. This technique is based on the reaction of neutral sugars with a phenol-sulfuric acid reagent. 1 mL of the sample is mixed with 1 mL of 5% phenol and 5 mL of concentrated sulfuric acid. The absorbance is then measured at 485 nm. The total sugar concentration was determined using a calibration curve established with glucose as the standard solution

4.5 Determination of reducing sugars by High-Performance Liquid Chromatography (HPLC): (Annex 5)

Sample Preparation:

Three samples were prepared at the laboratory of Kasdi Merbah University – Ouargla. The preparation procedure was as follows:

Weigh 1 g of coffee and date pits powder.

Add 100 mL of hot distilled water.

Stir for 10 minutes.

Filter the supernatant through a 0.45 µm filter.

HPLC Analysis:

Sugar analysis was performed using High-Performance Liquid Chromatography (HPLC) at the technical platform for physical-chemical analyses of Kasdi Merbah University – Ouargla.

6.Microbiological analyses (Annex 6)

6..1 Preparation of Dilutions

Dilution is a crucial step used to reduce the microbial concentration in the coffee, allowing for more accurate colony counting. After incubation. this step must be carried out under sterile conditions. It involves taking 1 mL of the stock solution and mixing it with 9 mL of a sterile diluent (physiological saline and Buffered Peptone Water ISO) in a test tube. After shaking, the first dilution is obtained. Then, 1 mL of this dilution is taken and transferred into a second test tube containing 9 mL of diluent, followed by another round of shaking. This process is repeated sequentially to obtain a series of dilutions**.**

6.2.Detection and Enumeration of Yeasts and molds

The presence of yeasts in food products can lead to their deterioration, which is often manifested by visual turbidity and the development of unpleasant odors. Molds, in addition to altering the organoleptic and nutritional properties of food, may also produce toxic compounds, notably mycotoxins.

For their detection, Sabouraud agar medium is used. A volume of 0.1 ml from each 10^{-1} dilution is spread onto the surface using a sterile spreader. The plates are then incubated at 25°C for 5 days [35]**.**

7.Sensory Analysis (Annex 8)

7.1.Objective

The aim of this sensory analysis is to evaluate and determine which beverages exhibit the most

favorable organoleptic qualities—including color, smell, taste, flavor, and overall acceptability—based on the assessments of a trained tasting panel.

7.2.Subjects (Tasters, Examiners, Judges)

A panel of 40 participants was formed to conduct the sensory evaluation of the date pit beverage. The panel consisted primarily of individuals of varying ages and genders. The sensory analysis involved a specialized team and a coffe substitu. The person responsible for overseeing the sensory analysis process—including panel recruitment, selection, and testing coordination—is referred to as the facilitator.

The facilitator should possess expertise in the following areas:

Sensory analysis: to identify and implement the most suitable testing methods;

Food technology: to prepare standard reference products and interpret sensory data accurately;

Communication: to effectively manage and coordinate with the panel and team;

Statistics: strong training is essential to analyze responses and draw meaningful conclusions;

Basic computer skills: for data processing and reporting.[31]

7.3.Preparation of coffee substitu from date pit powder:

Substitute Brewing Procedure:

Fill the base of the Moka pot up to the safety valve with **mineral water**. [36]

Fill the filter basket with **ground coffee**, without pressing it; simply level the surface.

Assemble the Moka pot and **heat on medium-low heat**.

As soon as **coffee begins to flow**, reduce the heat slightly to ensure a **slow extraction**. [36]

Remove from heat **when the flow starts to slow** to avoid **over-extraction**.

Serve **immediately at 60–65 °C** [37].

Sensory analysis was conducted in a controlled environment (clean, well-ventilated, odor-free, and properly lit rooms), following. All sessions were carried out between 10:30 AM and 1:00 PM, which is considered an optimal time for sensory sensitivity.

7.4.Steps followed during evaluation:

Workstations were thoroughly cleaned and prepared to ensure a neutral and hygienic environment .

Anonymous coding of samples using neutral letters (A, B, C, etc.) was implemented to eliminate cognitive bias regarding origin or presentation

Each workstation was equipped with:

A glass of mineral water for palate cleansing between samples [38].

Tissues to maintain cleanliness.

A standardized sensory evaluation form for recording panelist feedback.

Before starting, each panelist received a detailed explanation of:

The objective of the sensory evaluation, emphasizing the hedonic method and sensory profiling.

The specific tasting procedure as described in the evaluation sheet.

The importance of strictly following instructions to ensure the reliability and validity of results.

7.5.Sample Coding and Presentation to Panelists

Samples were coded anonymously, with no indication of their identity or origin, to ensure objectivity and prevent bias during evaluation.

7.6.Sample Details

1.Sample (more roasted)

Raw material: Date pits roasted at **200 °C for 25–30 minutes.**

Treatment: Ground just before use.

2.Sample (less roasted)

Raw material: Date pits roasted at **200 °C for 10–15 minutes** .

Treatment: Ground just before use.

7.7.Use of a Structured Evaluation Grid

Each sample was evaluated using a standardized grid including sensory characteristics such as taste, aroma, color, and texture. A numeric scale was used for scoring.

6.8.Analysis of Sensory Data

After tasting, data were analyzed to identify **statistically significant differences** between samples and to determine **which sensory characteristics were preferred**.

6.9.Presentation of Results

Results were compiled and presented using tables, graphs, and charts, providing a clear and comparative view of the scores given to each sample.

8.Statistical Analysis

The statistical analysis of our study was performed using SPSS version 29 software further to test Anova, test T and Chi-square, accepting $\alpha = 0.05\%$. Results were presented as means ± standard deviations. Results are significant when $P < 0.05$, highly significant when $P < 0.01$, and very highly significant when $P < 0.001$.

Chapter III Results and discussions

III Results and discussions

1.Physicochemical Analyses

1.1.pH:

pH is a key indicator of food quality and microbiological safety, as it directly influences microbial growth and the shelf life of the product **.** pH control is crucial according to the Codex Alimentarius**.** The results obtained that there is a difference **significant** between the pH of coffee Robusta and more roasted date pits powder and less roasted date pits powder. The pH value noted in the pits powder is close to the pH recorded by the authors [39] **6,12 ± 0,09 f**or the Gharss variety. And is close to [40] **5.91 ± 0.03** for another variety.

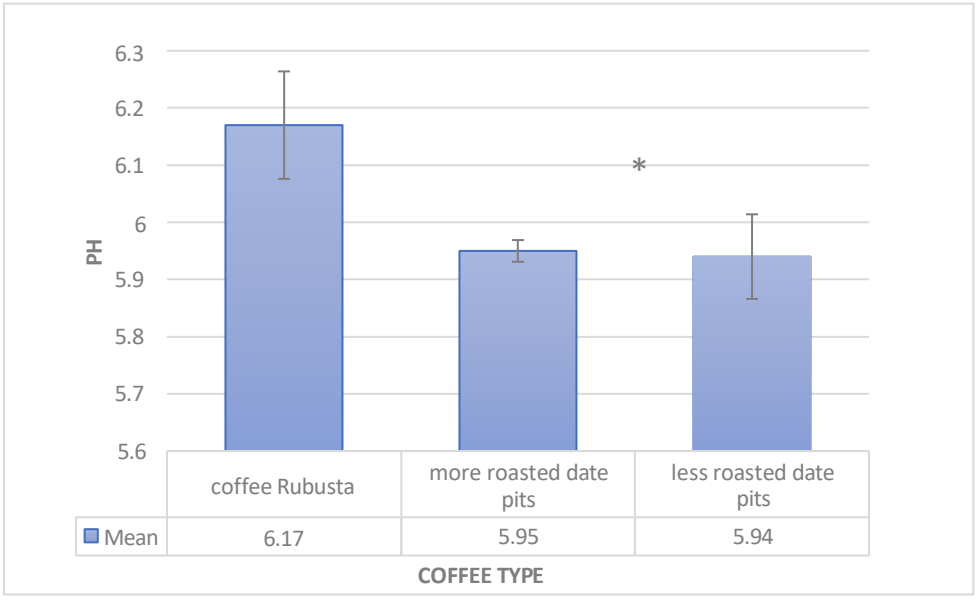


Figure13 : Comparison between the pH of Robusta coffee and more roasted date pits powder and less roasted date pits powder

1.2.Moisture Content

Moisture content is a key quality indicator, primarily used to determine the level of humidity present in date pits.Elle permet d'évaluer la résistance du produit aux altérations susceptibles de survenir au cours de sa conservation . The results obtained that there is a difference highly significant between the Moisture Content of coffee Robusta and more roasted date pits powder and less roasted date pits powder which are **4.00 ± 0.100, 1.68 ± 0, 525,2.92 ± 0.584** respectively. Robusta coffee showed the highest moisture content, while the most heavily roasted date pit powder had the lowest. The moisture contents measured for the date pit powder, lightly and highly roasted, were **1.68 ± 0.52%** and **2.92 ± 0.58%**, respectively. These values are significantly lower than those reported by [39], who observed a moisture content of **6.37%**, and by [41], whose study on various date varieties reported values ranging from **7% to 19%**. The fluctuation in the moisture content of the samples can be attributed to the effect of the heat treatment, which promotes water loss through evaporation, thereby causing a decrease in the product's moisture level.

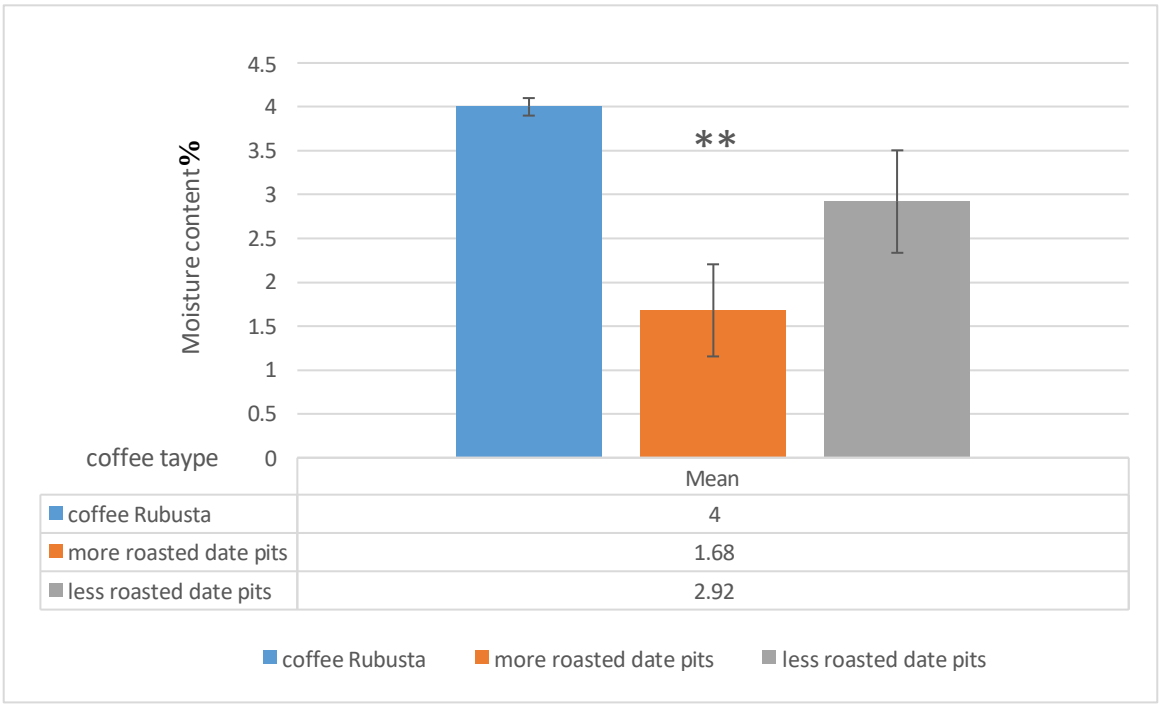


Figure14: Comparison between the Moisture Content of Robusta coffee and More roasted date pits powder and Less roasted date pits powder

1.3.Dry Matter Content

The results obtained that there is a difference highly significant between the of dry matter content coffee Robusta and more roasted date pits powder and less roasted date pits powder which are 96.00 ± 0.100, 98.31 ± 0, 525, 2.97.07± 0.584 **%** respectively. It appears that the three samples are particularly low in moisture, which explains their solid consistency. The low moisture content combined with a high dry matter content gives the pit its hard consistency The dry matter content of the seed powder is consistent with the values reported by [15].

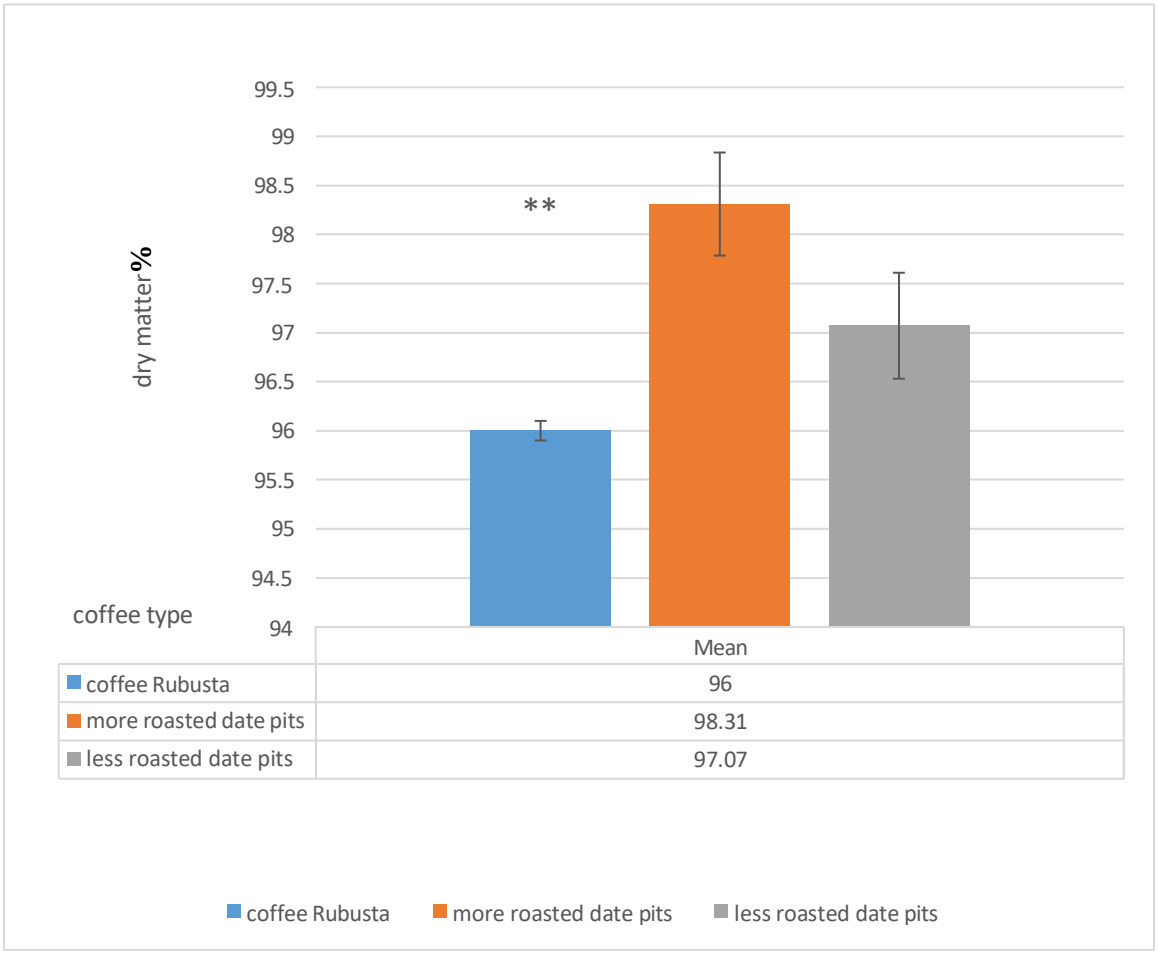


Figure15 : Comparison between the Dry Matter Content of Robusta coffee and more roasted date pits powder and less roasted date pits powder

1.4.Ash Content

The ash content serves as an indicator of the overall mineral load in the analyzed samples. The results obtained that there is a difference very highly significant between the Ash Content of coffee Robusta and more roasted date pits powder and less roasted date pits powder which are 4.33 ± 0.152, 1.06 ± 0, 020, 0.90± 0. 100 respectively. Several studies, including those by [14], [8] have reported that the ash content of date seed powder varies from 0.5% to 2% across different varieties. The ash content of the seed powder is consistent with the results reported by [14], [8]. The organic matter content of our samples is very high, with percentages of **95.67%, 98.94%**, and **99.9%** for coffee Robusta and more roasted date pits powder and less roasted date pits powder, respectively, reflecting their richness in organic matter. According to [15], dates contain various minerals (**Na, K, Mg, Cu, Fe, Ca, Zn)** whose concentrations vary depending on the variety, a variation that could be explained by the specific characteristics of the palm grove soils.

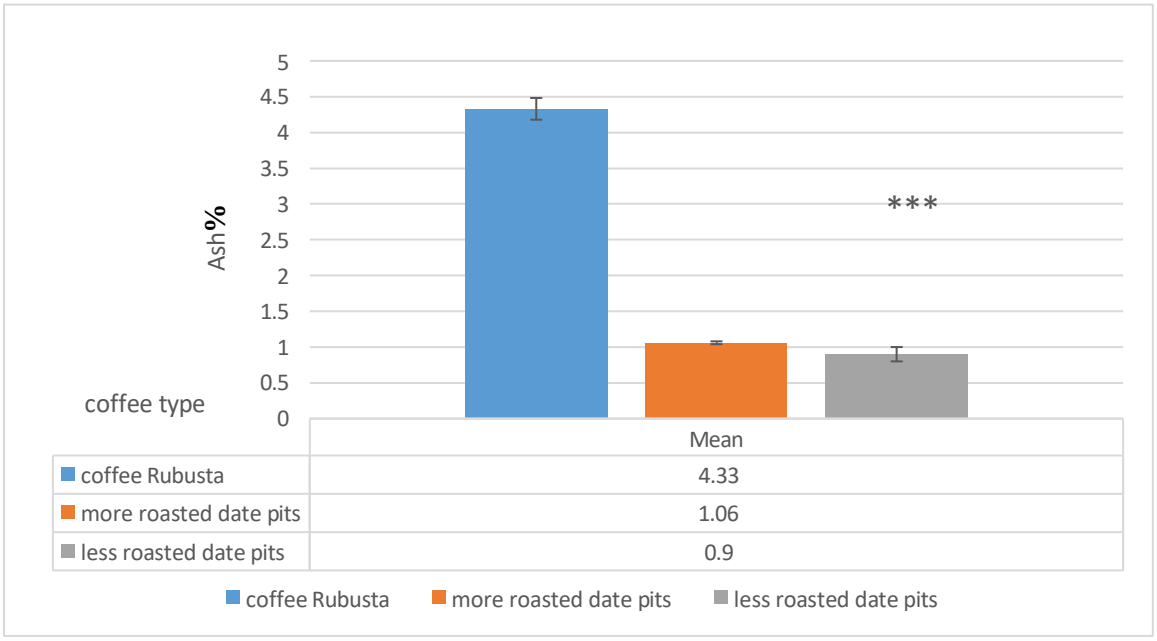


Figure16 : Comparison between the Ash Content of Robusta coffee and More roasted date pits powder and Less roasted date pits powder

1.5.Titratable Acidity

The titratable acidity of date seeds is a key parameter for assessing the physicochemical quality of extracts or powders derived from them, as it reflects the amount of free acids present in the food matrix [10].

The results obtained that there is a difference highly significant between the titratable Acidity of coffee Robusta and more roasted date pits powder and Less roasted date pits powder which are **0.41± 0.025, 0.32 ± 0, 020, 0.36± 0. 010%** respectively. Robusta coffee showed the highest Titratable Acidity, while the most heavily roasted date pit powder had the lowest. The Titratable Acidity of the seed powder is consistent with the values **0.39 ± 0.02** g of citric acid equivalent per 100 g of dry weight.reported by [8].

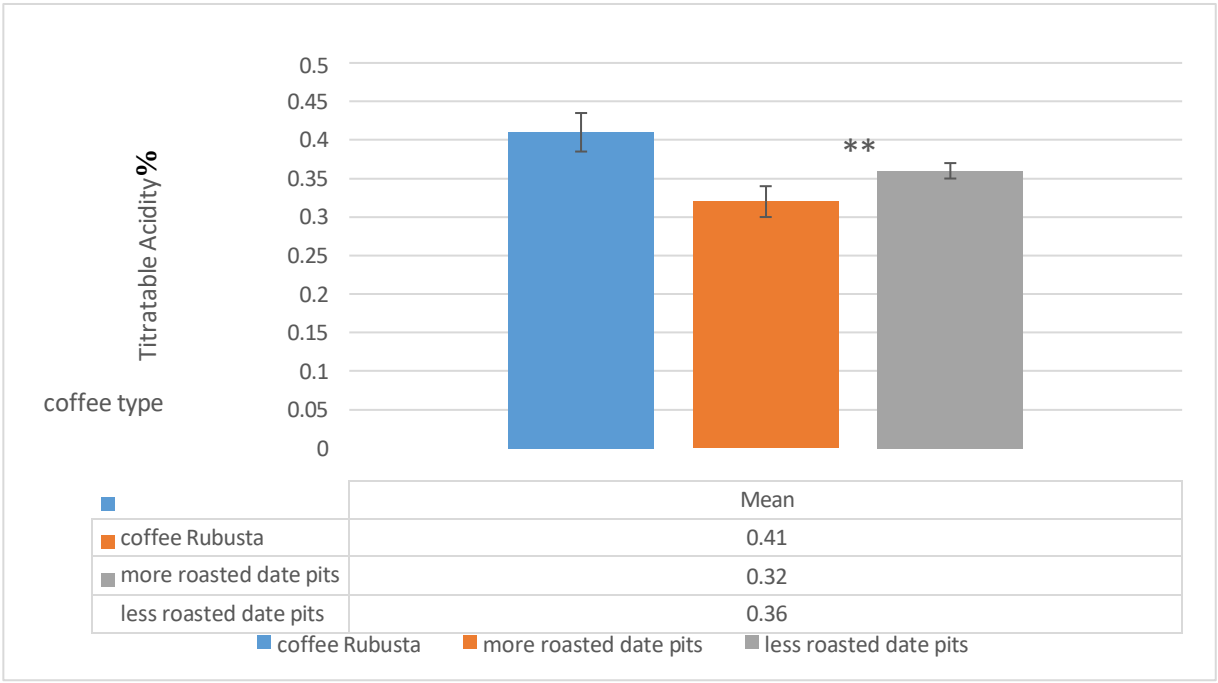


Figure17 : Comparison between the Titratable Acidity of Robusta coffee and More roasted date pits powder and Less roasted date pits powder

1.6 Fiber:

In food manufacturing, fiber also affects texture, water-holding capacity, and product stability, making control of its quantity essential for consumer acceptance and product function **.** dietary fiber as an important indicator in labeling and nutritional claims The results obtained that there is a difference **very highly significant** between the fibers of Robusta coffee and more roasted date pits and less roasted date pits.

Cellulose: The cellulose value noted in the pits powder more roasted and less roasted are **10.97 ± 0.000** and **11.42 ± 0.000** respectively; We have noticed that the less roasted date pits powder has more cellulose than more than more roasted date pits powder. Both date pit samples show higher cellulose content than coffee robusta valued at 10.17 ± 0.011; the date pit samples cellulose content noted is substantial difference with those in [42]

14,78 ± 0,60 for the Ghars variety; and These values are substantially not compatible with those in [42]**23.9±0.1** for Deglet Nour variety.

Hemicellulose: The hemicellulose value noted in the pits powder more roasted and less roasted are **13.63 ± 0.000 and 14.06 ± 0.011** respectively; We have noticed that the less roasted date pits powder has more hemicellulose than more than more roasted date pits powder. Both date pit samples show higher hemicellulose content than coffee robusta valued at 12.52± 0.000; the date pit samples hemicellulose content noted is the date pit samples hemicellulose content noted is substantial not according with those in 34.29±0.24.

Lignin: The Lignin in the pits powder more roasted and less roasted are 19.14 ± 0.005 and 19.96 ± 0.005 respectively. We have noticed that the less roasted date pits powder has more lignin than more than more roasted date pits powder. Both date pit samples show higher lignin content than coffee robusta valued at 18.68 ± 0.005; the date pit samples lignin content noted is the date pit samples lignin content noted is slightly difference with those in [44] 21.6±0.1 and [43] 21.20 ± 0.06.

Pectin: The Pectin value noted in the pits powder more roasted and less roasted are **3.81 ± 0.005,** and **4.24 ± 0.005** respectively. We have noticed that the less roasted date pits powder has more pectin than more roasted date pits powder. Both date pit samples show higher pectin content than coffee robusta valued at **3.33 ± 0.005**.

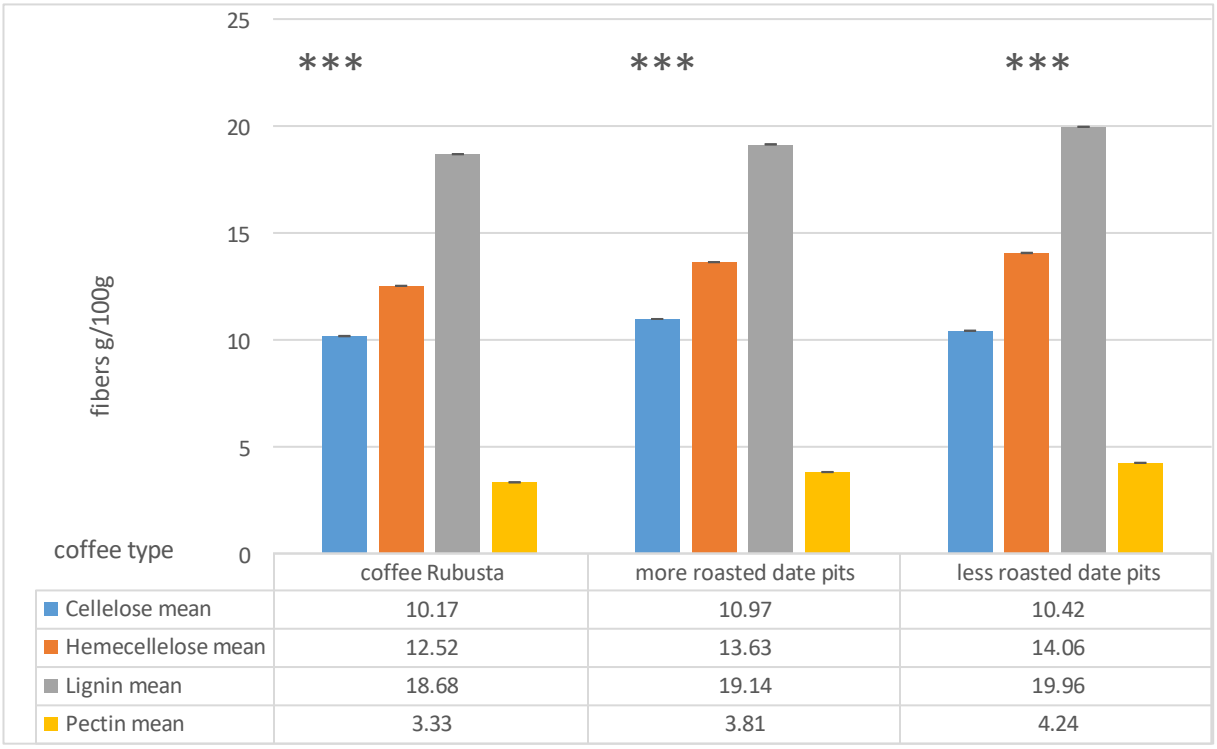


Figure18: Comparison between the fibers of Robusta coffee and more roasted date pits powder and less roasted date pits powder

2.Biochemical Analysis

2.1.Lipids

The results obtained that there is a difference very highly significant between all the acids :

Saturated fatty acid: 4.30± 0.15 for coffee Robusta and 3.14 ± 0, 017 of more roasted date pits powder and 3.86± 0. 15 of Less roasted date pits powder.

Palmitic acid: **2.73 ±0.005** of coffee Robusta and **1.74 ± 0, 011** of more roasted date pits powder and 2.13± 0.037of Less roasted date pits powder.

Steric acid: **1.50± 0.020** for coffee Robusta and **1.39 ± 0, 005** of more roasted date pits powder and **1.70± 0. 020** of Less roasted date pits powder.

Mono unsaturated fatty acid: 2.20 ± 0.025 for coffee Robusta and **2.88 ± 0, 005**of more roasted date pits powder and **3.42± 0. 000** of Less roasted date pits powder.

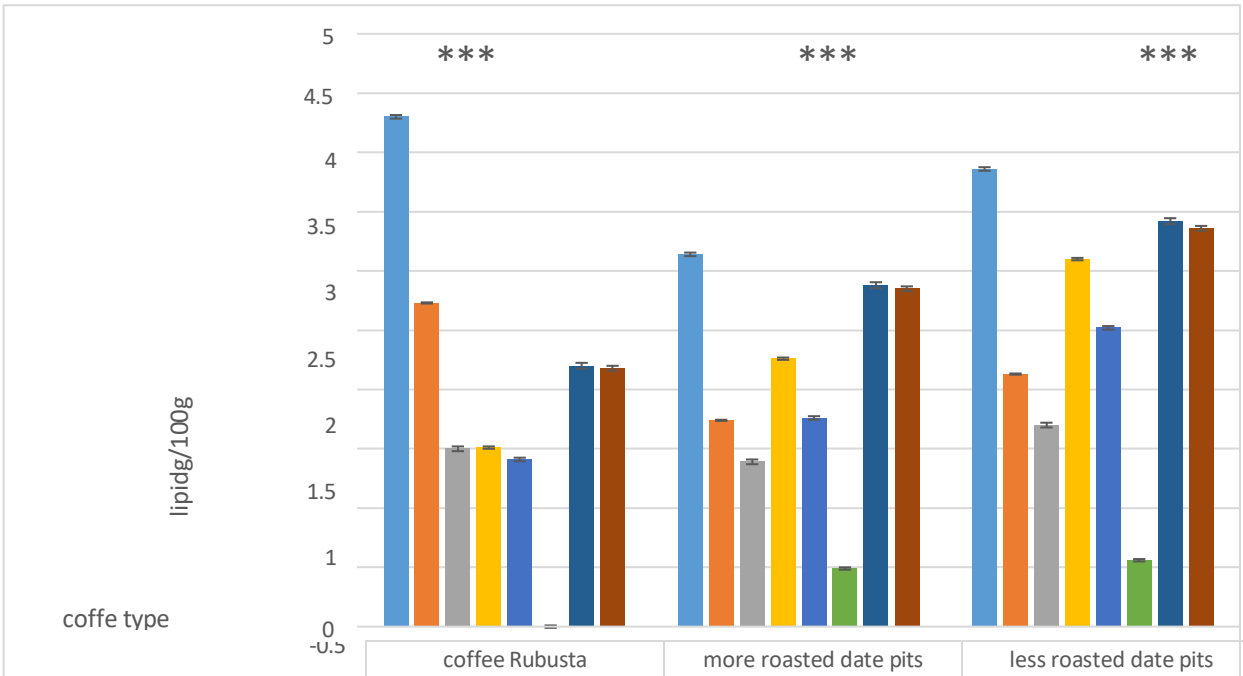
Oleic acid: 2.18± 0.020 for coffee Robusta and **2.85 ± 0,000** of more roasted date pits powder and **3.36± 0. 010** of Less roasted date pits powder.

Poly unsaturated fatty acid: **1.513± 0.011** for coffee Robusta and **2.26 ± 0,005** of more roasted date pits powder and **3.100± 0. 010** of Less roasted date pits powder.

Linoleic acid: **1.49± 0.005** for coffee Robusta and **1.76 ± 0, 015** of more roasted date pits powder and **2.52± 0. 015** of Less roasted date pits powder.

Linolenic acid: 0.49± 0.010 of more roasted date pits powder and **0.56 ± 0.005** of Less roasted date pits powder.

There are traces of lauric acid, myristic acid, and palmitoleic acid in all samples, and linolenic acid in Robusta coffee. Based on the results obtained, we observe that the sample Less roasted date pits powder contains a high percentage of unsaturated fatty acids, particularly oleic acid, linoleic acid, and linolenic acid. These fatty acids are known for their cardiovascular benefits. On the other hand, Robusta coffee contains a higher proportion of saturated fatty acids, particularly palmitic acid, which may make it less beneficial from a nutritional standpoint. It appears that high-degree roasting slightly reduces the fatty acid content, especially the unsaturated ones, possibly due to oxidation or thermal degradation. Under high-temperature roasting,The lipids in date seeds may undergo thermal degradation (pyrolysis), leading to a reduction in their measurable content [45] Additionally, some volatile oils may evaporate or oxidize, further contributing to the loss of fat content [46]**.** reduction in their measurable content [44] Additionally, some volatile oils may evaporate or oxidize, further contributing to the loss of fat content [45]**.**



	coffee Rubusta	more roasted date pits	less roasted date pits
Saturated fatty acid	4.3	3.14	3.86
Palmitic acid	2.73	1.74	2.13
Steric acid	1.5	1.39	1.7
Poly unsaturated fatty acid	1.51	2.26	3.1
Linoleic acid	1.41	1.76	2.52
Linolenic acid	0	0.49	0.56
Mono unsaturated fatty acid	2.2	2.88	3.42
Oleic acid	2.18	2.85	3.36

Figure19 : Comparison between the Lipid of Robusta coffee and More roasted date pits powder and Less roasted date pits powder.

2.2.Proteins

Protein is one of the critical nutrients for human nutritional systems [47]. The results obtained that there is a difference very highly significant between the protein of Robusta coffee and more roasted date pits and less roasted date pits.

The protein value noted in the pits powder more roasted and less roasted are **10.87 ± 0.005** and **9.79 ± 0.005** respectively. We have noticed that the less roasted date pits powder and coffee

robusta have more protein than more roasted date pits powder. the date pit samples porotein content noted is the date pit samples protein content noted is substantial difference with those in [47] **6.17 ± 0.28** for the saidy variety and [46] **6,51 ± 0,11** for el gharss variety and for deglet nour 8,59 ± 0,68. The protein content increases in more heavily roasted date seeds primarily due to moisture loss during roasting, which concentrates the dry components, including proteins.

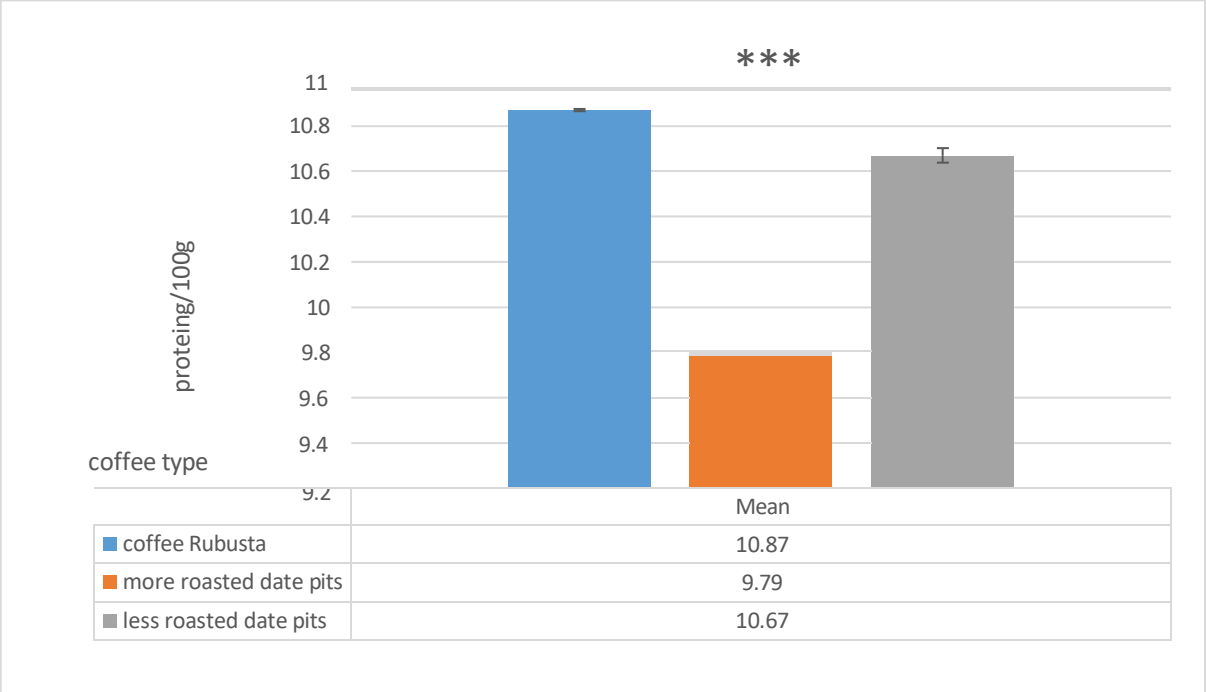


Figure20 : Comparison between the protein of Robusta coffee and more roasted date pits powder and less roasted date pits powder

2.3.Total sugars

The results obtained that there is a **significant** difference between the total sugars of Robusta coffee and more roasted date pits and less roasted date pits.

The total sugars value noted in the pits powder more roasted and less roasted are **0.003 ± 0.0042** and **0.016±0.003** respectively. It can be observed that less roasted date pits powder and coffee robusta valued at **0.015 ± 0.0072** have comparable sugar contents, with less roasted date pits powder exhibiting the highest value, whereas more roasted date pits powder recorded the lowest. We note that the previous results are not compatible from the results mentioned by

[46] for other varieties. Because heat stimulates sugar decomposition via caramelization and Maillard reactions [22], lightly roasted seeds contain more sugars than heavily roasted ones, which become darker in color and acquire richer flavors.

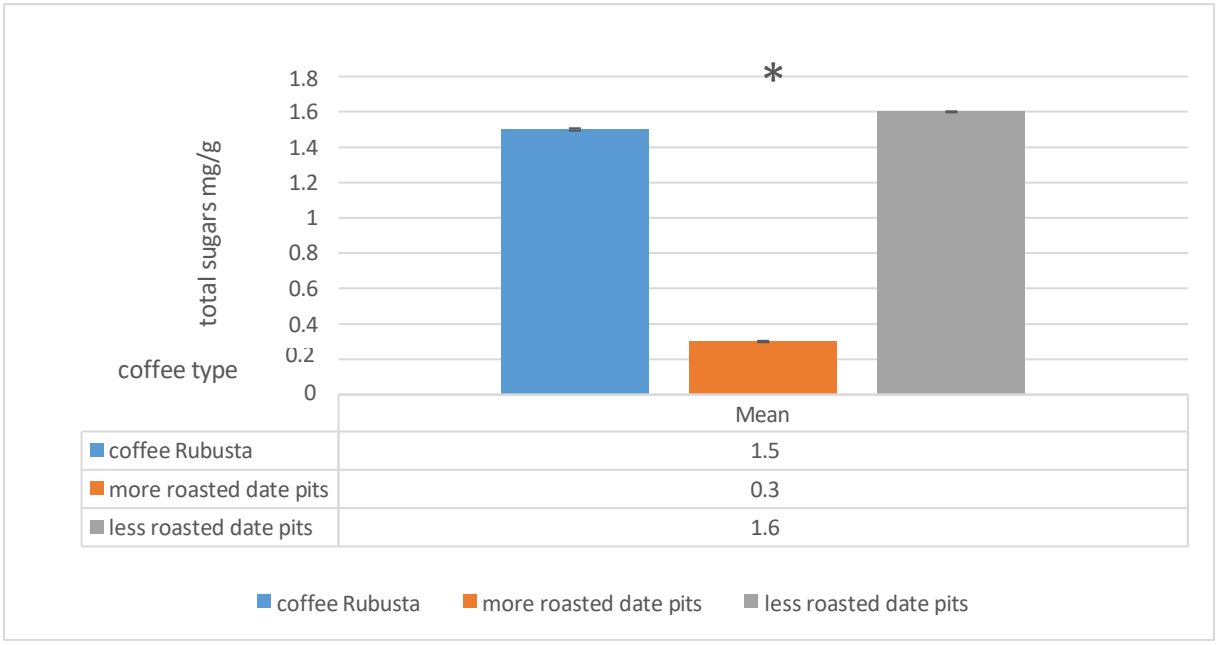


Figure21: Comparison between the total sugars of Robusta coffee and more roasted date pits powder and less roasted date pits powder

2.3.1.Reducing sugar analysis

Glucose: The results obtained that there is a difference **very highly significant** between the glucose of coffee Robusta and more roasted date pits powder and less roasted date pits powder which are **3.04 ± 0.020**, **40.67 ± 0.020** and **0.00 ± 0.000** respectively.

Fructose: The results obtained that there is a difference **very highly significant** between the fructose of coffee Robusta and more roasted date pits powder and less roasted date pits powder which are **3.95± 0.010**, **15.61 ± 0.010** and **9.12 ± 0.020** respectively.

Sucrose: The results obtained that there is a difference **very highly significant** between the fructose of coffee Robusta and more roasted date pits powder and less roasted date pits powder which are 15.15 **± 0.010**, **5.90 ± 0.010** and **2.003 ± 0.005** respectively.

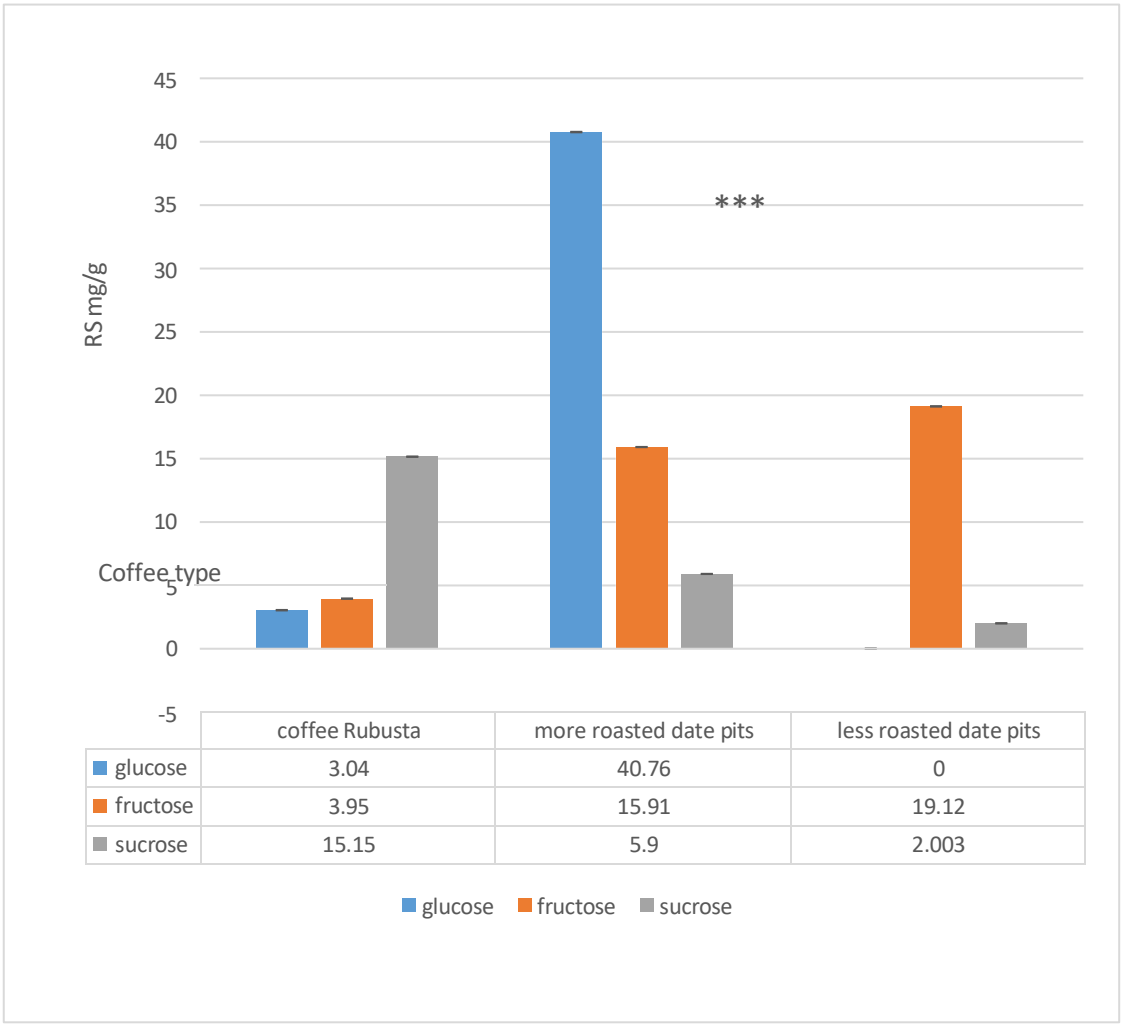


Figure 22: Comparison between the reducing sugars of Robusta coffee and more roasted date pits powder and less roasted date pits powder (Rati)

2.5.Total polyphenols

The results obtained that there is a difference **highly significant** between the TPC of Robusta coffee and more roasted date pits and less roasted date pits, **The total polyphenols** value noted in the pits powder more roasted and less roasted are 60.53 ± 15.88and 77.75 ± 6.13 mg GAE/g respectively. **We have noticed that the less roasted date pits** powder has more than more roasted date pits powder valued. Both date pit samples show lower Total polyphenols content than coffee robusta valued at **111.15 ± 1.15 mg GAE/g**.

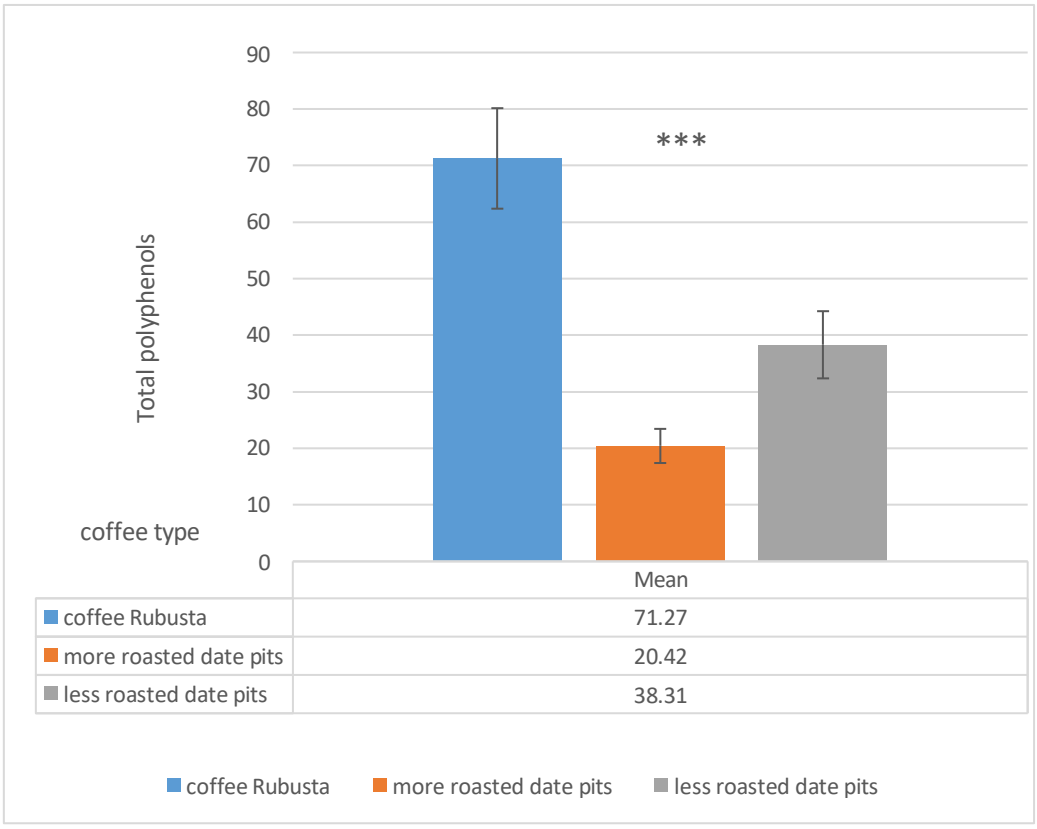


Figure23 : Comparison between the total polyphenols of Robusta coffee and more roasted date pits powder and less roasted date pits powder

- Polyphenol analysis by **HPLC** revealed that the sample was rich in gallic acid, and many other types as shown in the figure:

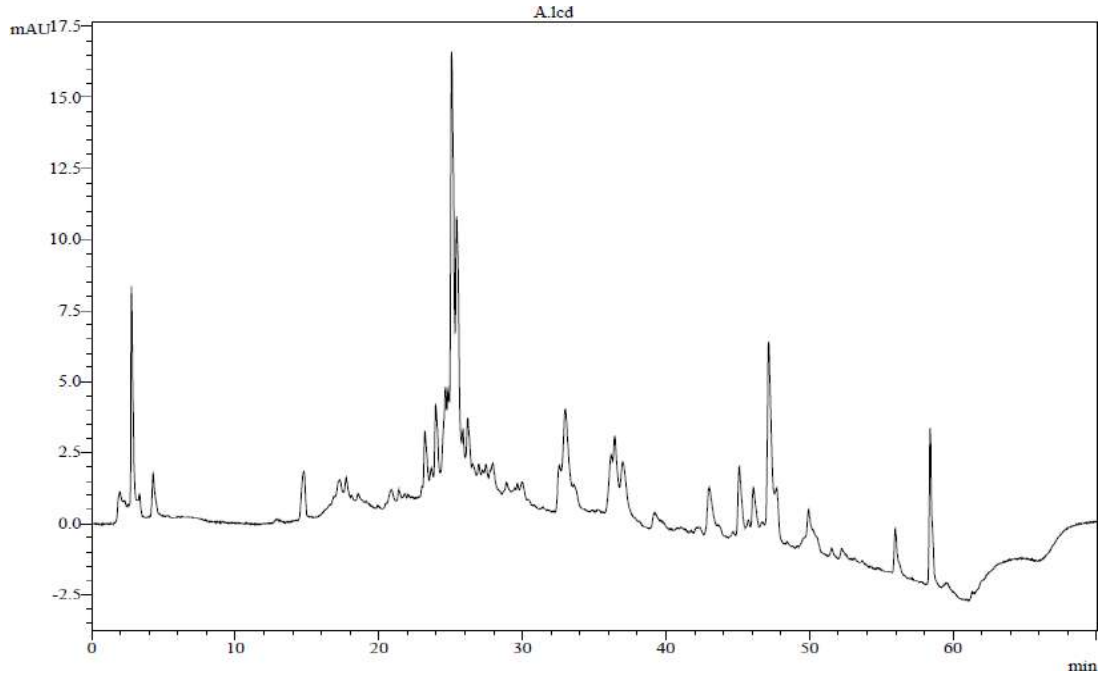


Figure 24: Results obtained for polyphenols of the less roasted date pits powder by the HPLC

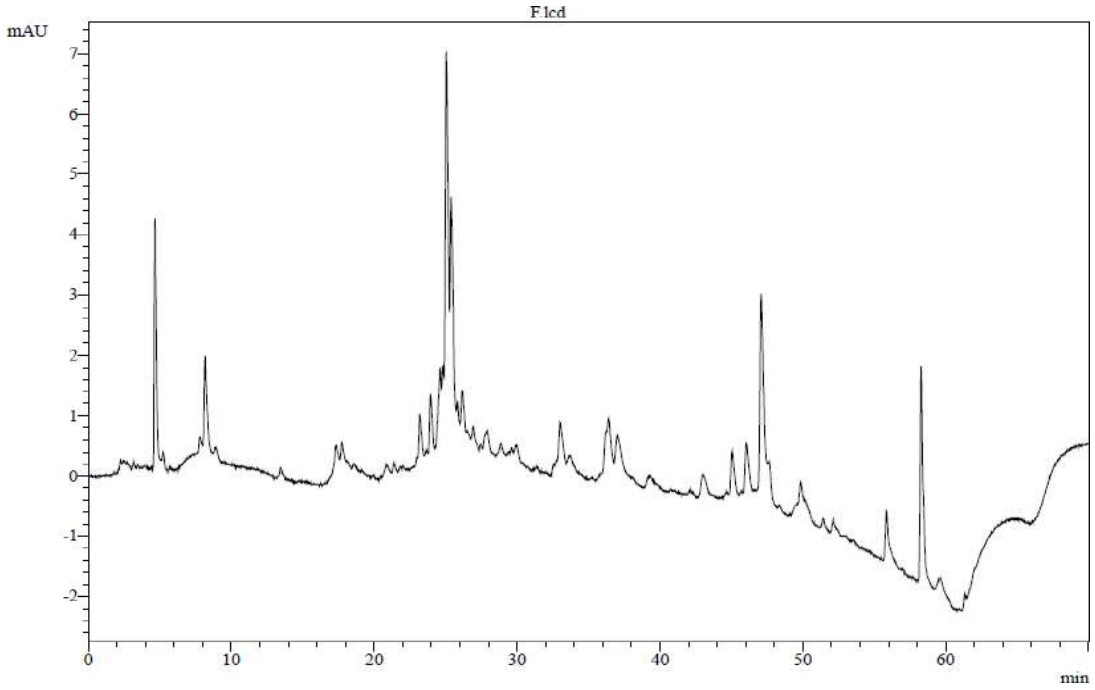


Figure 25: Results obtained for polyphenols of the More roasted date pits powder by the HPLC

2.5 flavonoids

The results obtained that there is a difference **very highly significant** between the total sugars of Robusta coffee and more roasted date pits and less roasted date pits . **The flavonoids** value noted in the pits powder more roasted and less roasted are **20.42±3.02** and **38.31±5.94** respectively. Both date pit samples show lower lignin content than coffee robusta valued at **71.27±8.88;** The less roasted date kernel powder showed the highest flavonoid content **(38.31 ± 5.94 mg quercetin equivalent/g**) compared to the more roasted one **(20.42 ± 3.02)**, the latter value being remarkably close to the flavonoid content of the (Algarni; 2020) sample **(5.15 ± 0.82)**, and both date samples differing from the highest value found **(47.30 ± 0.63).**

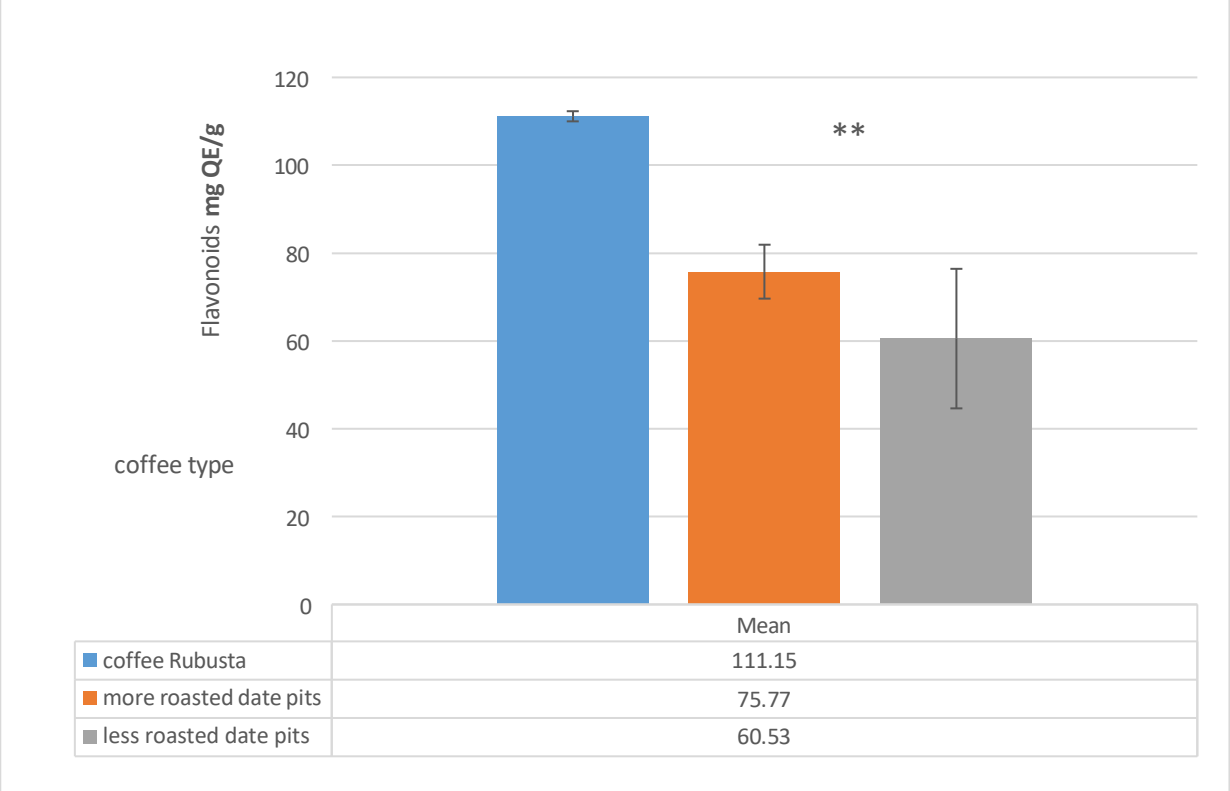


Figure26: Comparison between the flavonoids of Robusta coffee and more roasted date pits powder and less roasted date pits powder

3.Microbiological analysis

3.1 Enumeration of yeasts and molds

The results obtained that there is a difference highly significant between groups for yeast

and mold of coffee Robusta and more roasted date pits powder and Less roasted date pits powder which are **118.18± 0.010, 104.54 ± 0, 010, 123.09± 0. 010** respectively. The less roasted date pit shows the highest microbial contamination. The more roasted date pit has the lowest yeast and mold content. Robusta coffee shows an intermediate level of contamination between the two types of date pits. These results demonstrate that roasting temperature has a significant impact on reducing microbial load. The more heavily roasted date pit exhibits the lowest presence of yeasts and molds, likely due to the heat's destructive effect on microorganisms. In contrast, the less roasted pit shows higher contamination, suggesting that light roasting is less effective at disinfecting the product. Robusta coffee, although roasted, retains a higher microbial load than the heavily roasted date pit. This may be attributed to differences in matrix composition or post-roasting handling. This indicates that for food or health applications, the roasting level of date pits could play a critical role in ensuring the microbiological safety of the product.

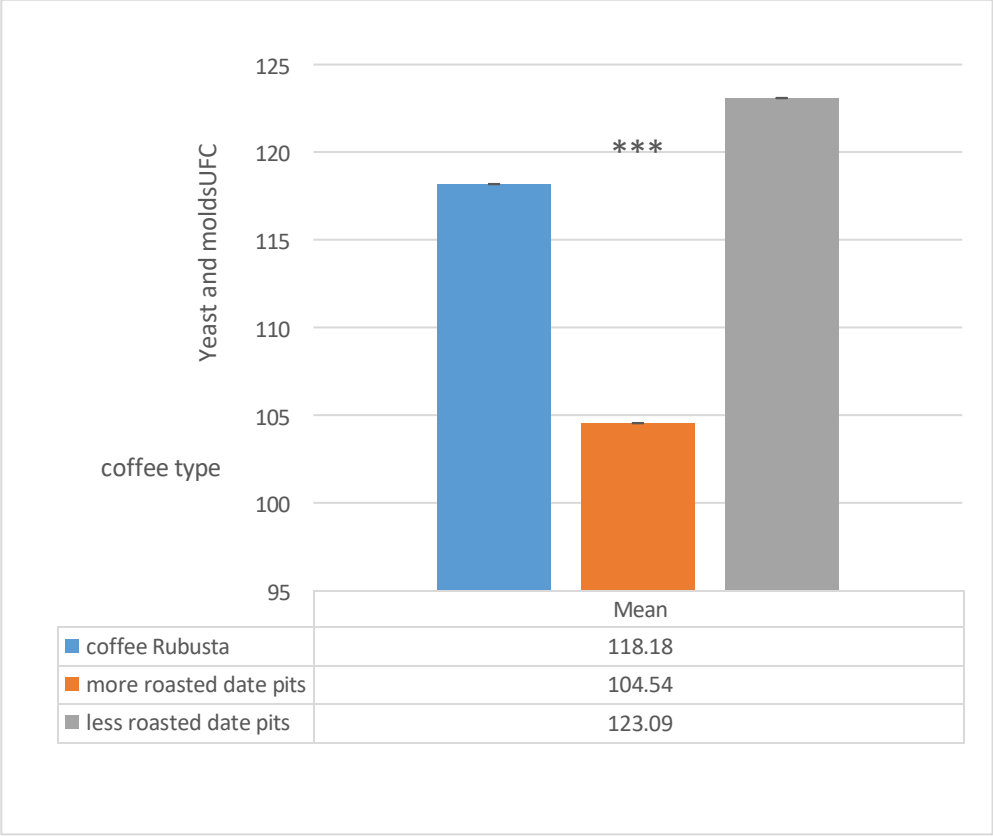


Figure27: Comparison between the **yeasts and molds** of Robusta coffee and more roasted date pits powder and less roasted date pits powder

4.Sensory analysis

- **SEX**:52.5 male**%** / 47.5**%** female
- **AGE:**19-40:.75**%** /41.62:25**%**

4.1.the color

The results indicate that **85%** of participants perceived the color of the unsweetened and sweetened less roasted samples as light brown, while **15%** identified it as brown.

Regarding the more roasted sweetened and unsweetened samples, **72.5%** of participants identified the color as dark brown, **20%** as brown, and **7.5%** as very dark brown.

The p values were very low (0.018 for more roasted samples), and p-values were greater than 0.05 (0.156), indicating no significant correlation between gender and color perception. There was no statistically significant difference between males and females for any of the sample types. the p values were very low (0.018 for less roasted samples), and p-values were greater than 0.05 (0.894), indicating no significant correlation between gender and color perception. The results also show no significant differences according to age across all sample types.

For the unsweetened and sweetened lightly roasted samples, the p-value was **0.609,** indicating that color perception did not differ significantly between the age groups **(19–40 years vs. 41– 62years).**

Similarly, for the unsweetened and sweetened more heavily roasted samples, a p-value of **0.157** confirms the absence of a significant difference in color perception between these age groups. The results indicate that roasting significantly influences the perception of the product's color, while sugar content, gender, and age appear to have no notable effect.

A higher degree of roasting leads most participants to perceive a darker color (dark brown to very dark brown), which aligns with the physicochemical changes that occur when the product is exposed to high temperatures.

The lack of variation based on gender or age suggests that color perception is a relatively universal sensory characteristic in this context.

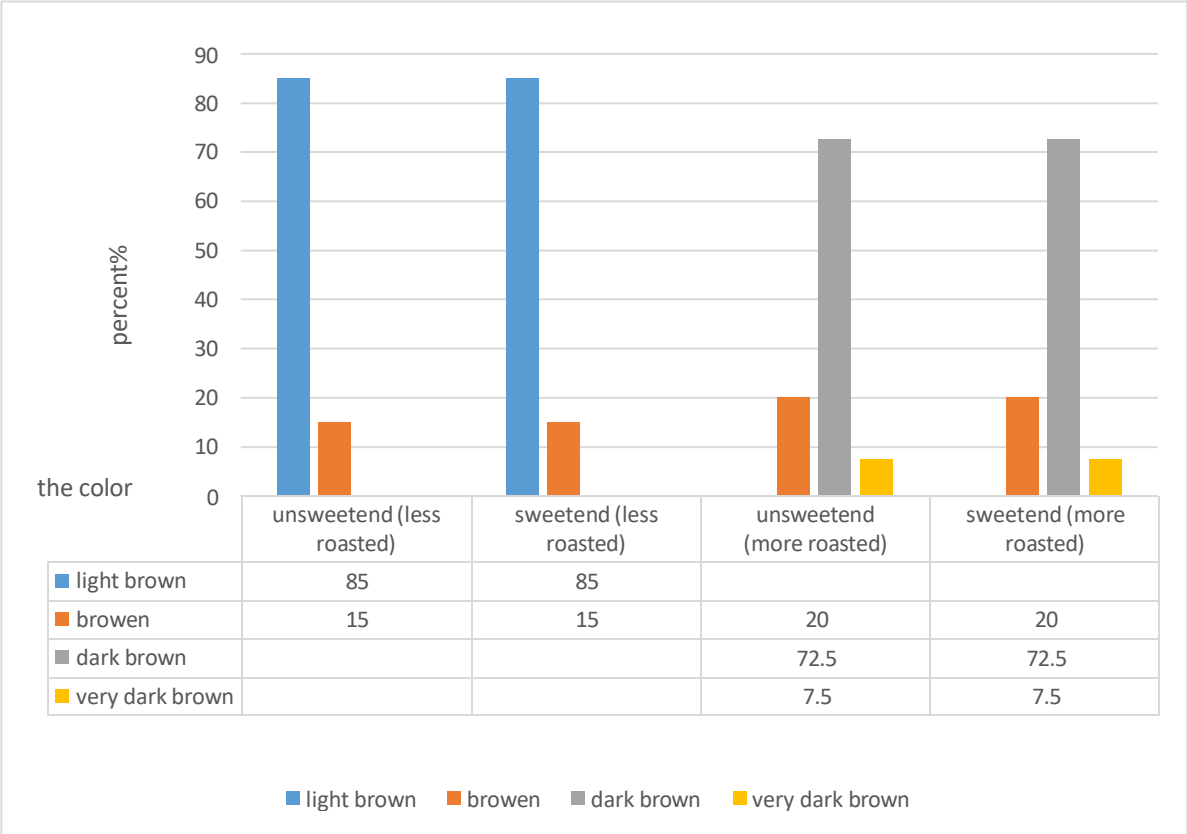


Figure28: Effect of Roasting and sweeting on the color distribution of date pits powder

4.2.Odor

For the unsweetened and lightly roasted samples, **47.5%** of participants described the odor as "strong" and **52.5%** as moderate".

The sweetened samples with the same roasting level received identical response proportions. In contrast, the more heavily roasted samples, both sweetened and unsweetened, were predominantly perceived as having a "very strong" odor **(62.5%)**, followed by a "strong" odor **(35%)**.

There is significant differences between genders in the evaluation of odor across all samples **($p > 0.05$).** although minor variations were observed between the two age groups (**19–40 and 41–62 years),** there is no statistically significant differences **($p > 0.05$)**.

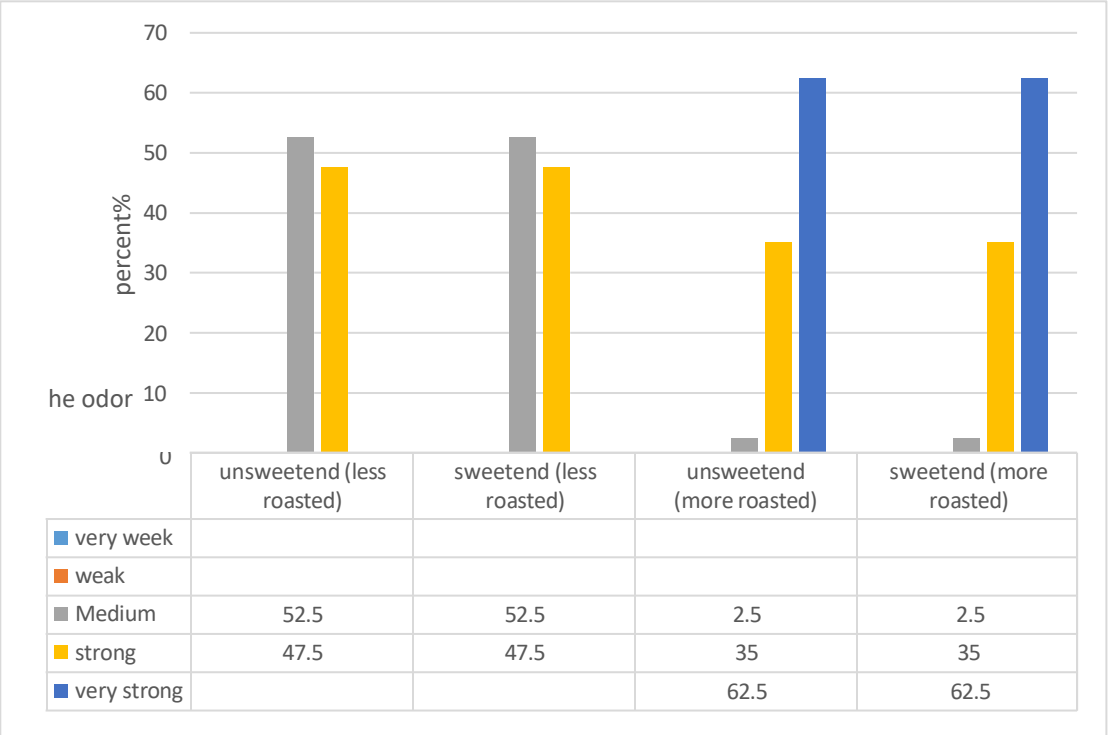


Figure29: Effect of Roasting and sweeting on the odor distribution of date pits powder

4.3. Sweet taste

The unsweetened, lightly roasted sample was predominantly rated as having a moderately sweet taste by **97.5%** of participants.

The sweetened, lightly roasted sample was perceived as strongly sweet by **82.5%** of participants. The unsweetened, heavily roasted sample was considered to have a slightly sweet taste by **82.5%** of participants.

The sweetened, heavily roasted sample received mostly "moderately sweet" ratings **(72.5%),** while **25%** of participants found it slightly sweet.

Ther is no significant association between gender or age and the perception of sweetness, with one exception: a moderate association with gender was observed for the sweetened, lightly roasted sample **($\chi^2 = 7.677$, $p = 0.053$)**, approaching the threshold of statistical significance.

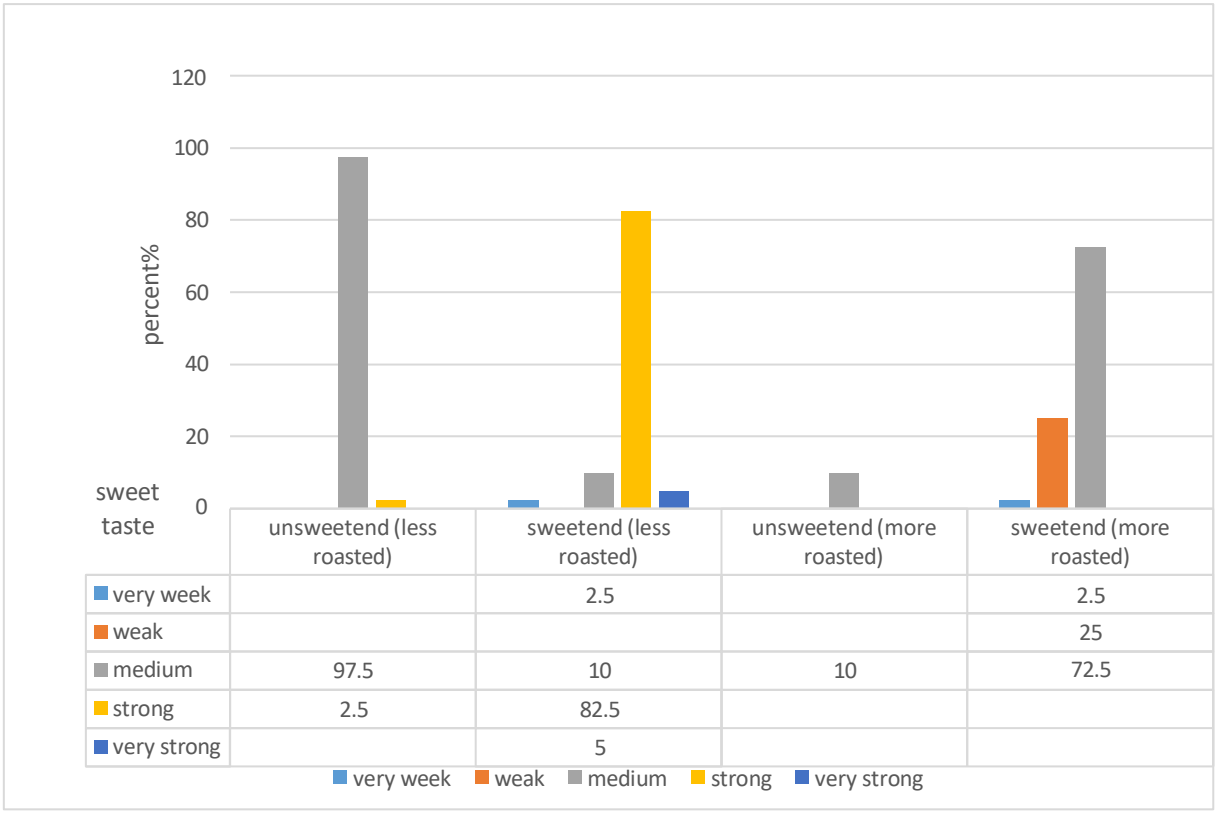


Figure30: Effect of Roasting and sweeting on the sweet taste distribution of date pits powder

4.4. Sour teste

Unsweetened, lightly roasted sample: **82.5%** of participants perceived the acidity as "strong", **12.5%** as "moderate", and **5%** as "very strong". Sweetened, lightly roasted sample: **65%** rated the acidity as "moderate", **32.5%** as "strong", and **2.5%** as "weak". Unsweetened, dark roasted sample: **85%** perceived the acidity as "weak" and **15%** as "very weak". Sweetened, dark roasted sample: **67.5%** rated the acidity as "veryweak" and **32.5%** as "weak". No statistically significant differences were found between male and female participants or between the two age groups **(19–40 years and 41–62 years)**, with all p-values exceeding **0.05**.

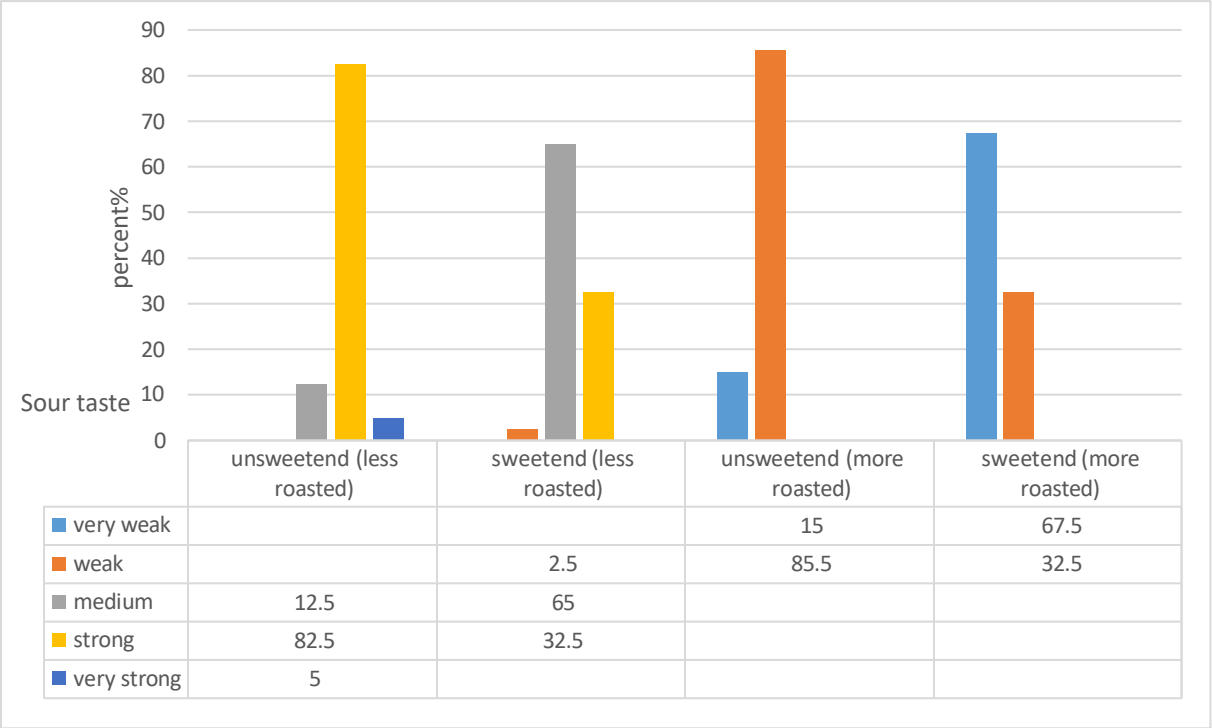


Figure31: Effect of Roasting and sweeting on the sour taste distribution of date pits powder

4.5. Bitter teste

The perceived intensity of acidity is mainly influenced by the degree of roasting and the presence of sugar. Light roasting without sugar leads to a stronger perception of acidity, while darker roasting and the addition of sugar significantly reduce it.

The lightly roasted samples were predominantly associated with low to moderate bitterness. In contrast, the more heavily roasted samples were perceived as bitter or even very bitter.

In some cases, particularly with unsweetened samples, a trend toward statistical significance was observed in relation to gender and age. However, these differences did not reach a statistically significant level**(p-value > 0.05).**

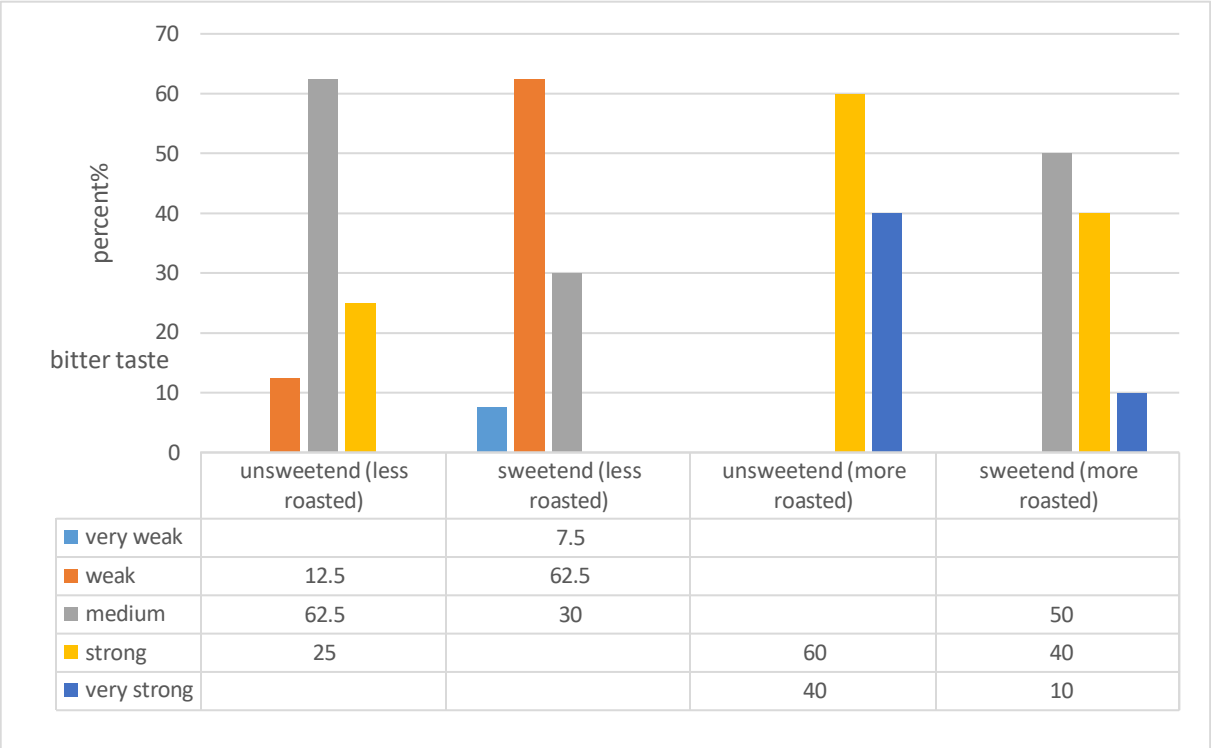


Figure32: Effect of Roasting and sweeting on the bitter taste distribution of date pits powder

4.6. Consistency

In the lightly roasted samples, whether sweetened or not, the majority of participants **(70%)** described the consistency as "light." Conversely, in the more heavily roasted samples, sweetened or unsweetened, **77.5%** of participants described the consistency as "heavy." No statistically significant differences were found between male and female participants regarding texture perception across all four samples **($p > 0.05$)**. There was a slight trend showing that younger participants **(aged 19–40)** were more likely to perceive the texture of lightly roasted samples as "light." However, this trend was not statistically significant **($p > 0.05$)**.

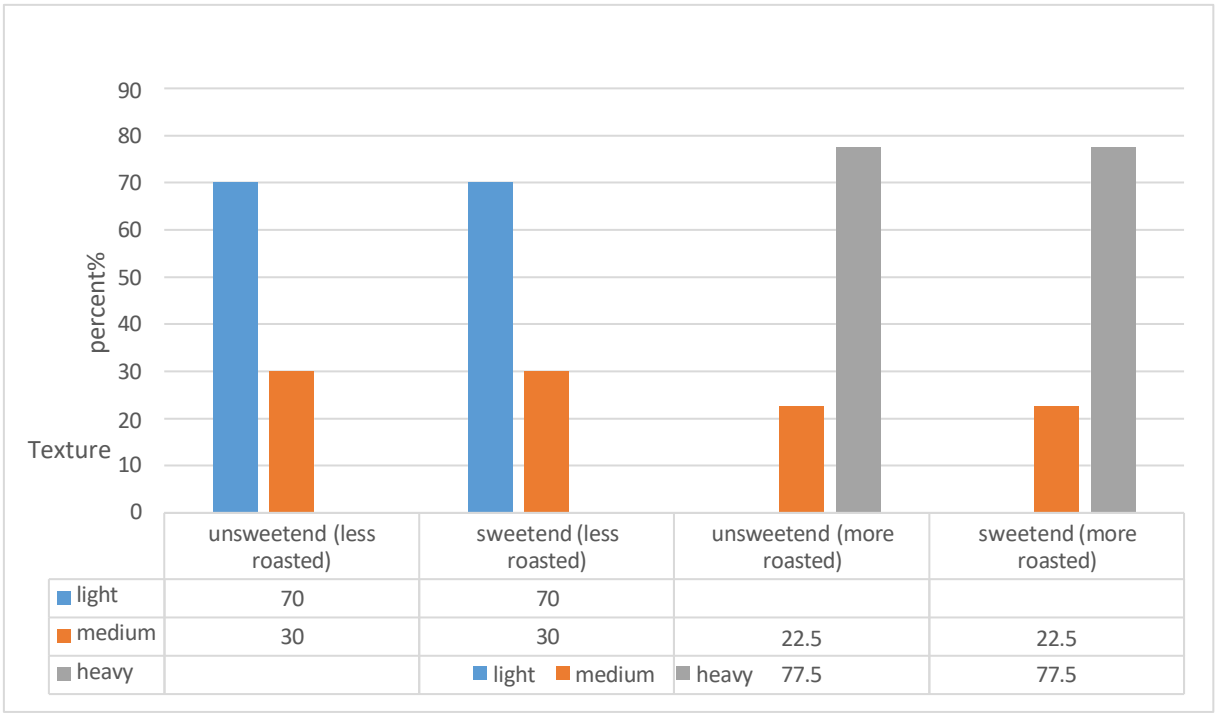


Figure33: Effect of Roasting and sweeting on the Consistency distribution of date pits powder

4.7. Aftertaste (the lingering taste of the coffee)

The analysis of aftertaste (the lingering taste of the coffee) revealed a predominance of "Strong" and "Medium" intensities for the lightly roasted samples, and "Very strong" for the more heavily roasted ones.

Specifically: For lightly roasted coffee, 60% of participants perceived a medium aftertaste, while 40% reported it as strong. For dark roasted coffee, 72.5% indicated a strong aftertaste, and 27.5% perceived it as very strong.

Chi-square tests showed no statistically significant differences in perceived aftertaste based on gender or age **(p-value > 0.05).**

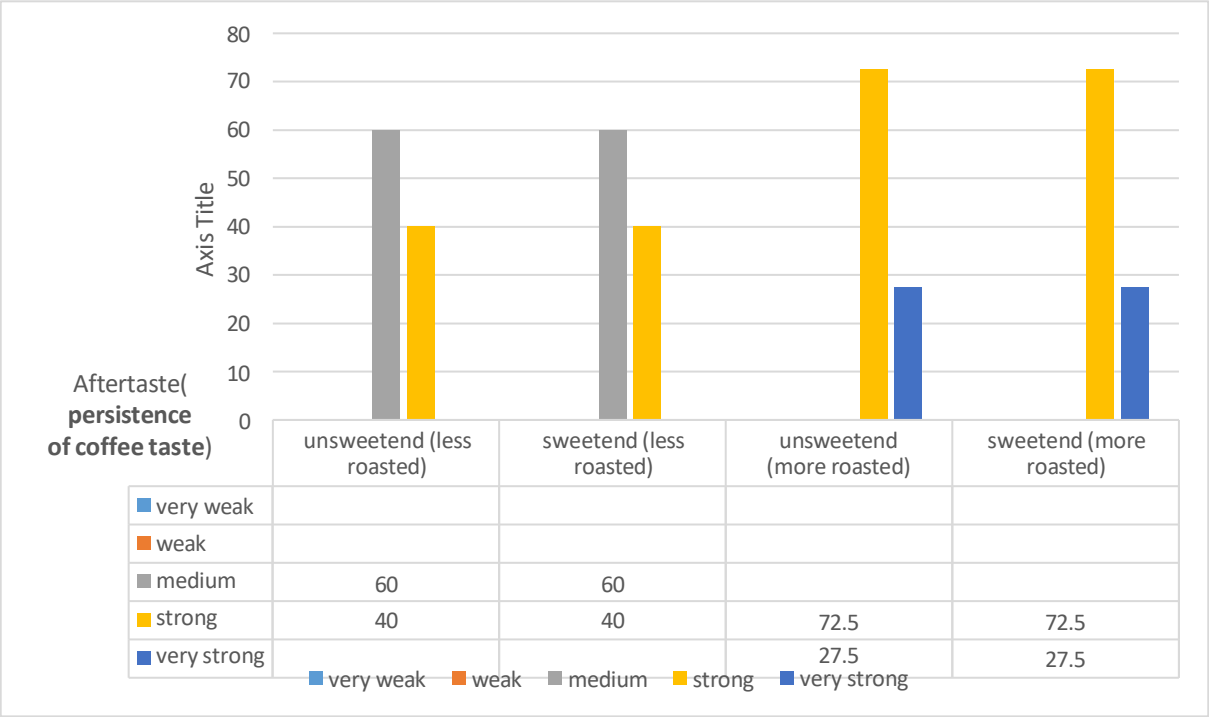


Figure34: Effect of Roasting and sweeting on the after taste distribution of date pits powder

4.8. Identified flavor or aroma

As for aroma perception, all participants (100%) identified it as "burnt" across all sample types. No variation was observed by gender or age, and no statistical analysis could be performed due to the uniformity of responses.

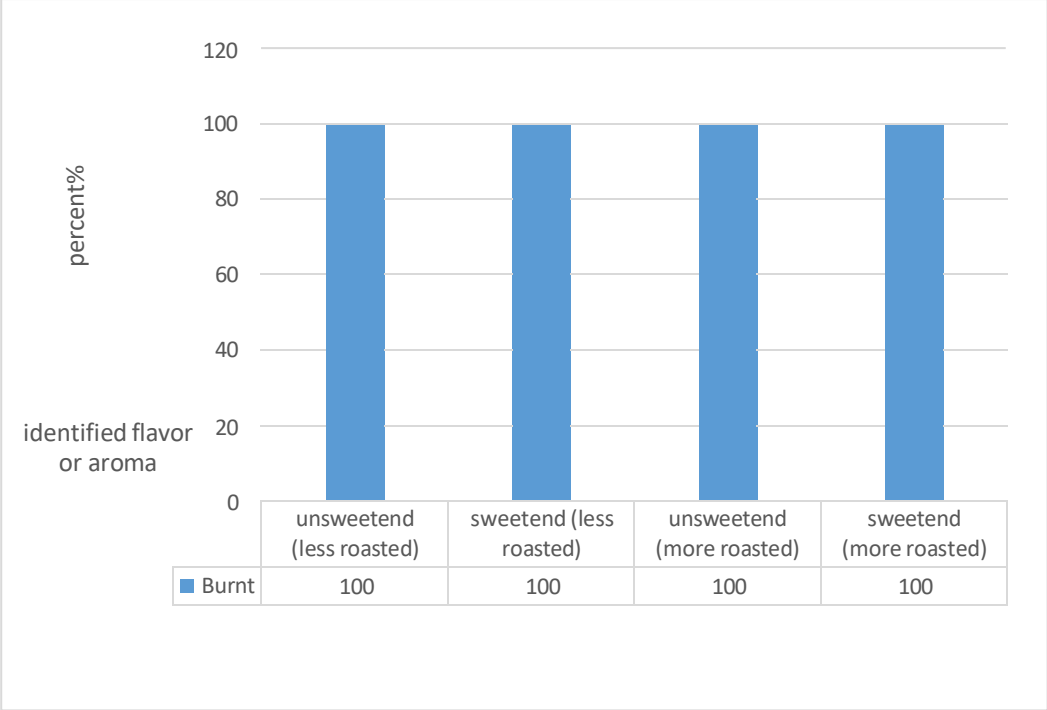


Figure35: Effect of Roasting and sweeting on the identified taste distribution of date pits powder

4.9.the preferance scale (hedonic taste)

Sample 1: Unsweetened, less roasted

72.5% of participants found it pleasant to varying degrees, while 5% rated it as slightly unpleasant. There is no statistically significant differences based on gender or age ($p > 0.05$), although a trend related to age was observed ($p = 0.093$ for the chi-square test and $p = 0.024$ for the likelihood ratio test).

Sample 2: Sweetened, less roasted

All participants (100%) rated the coffee as at least slightly pleasant. A statistically significant difference was found based on age ($p = 0.012$ for the chi-square test; $p = 0.004$ for the likelihood ratio test), suggesting that age influences preference for this type of coffee.

Sample 3: Unsweetened, more roasted

75% of participants found it very or extremely pleasant. No significant differences were found based on gender or age ($p > 0.05$).

Sample 4: Sweetened, more roasted

This sample received the highest ratings, with 100% of participants finding it pleasant, including 15% who rated it as extremely pleasant. Statistical tests revealed no significant effect of gender or age ($p > 0.05$).

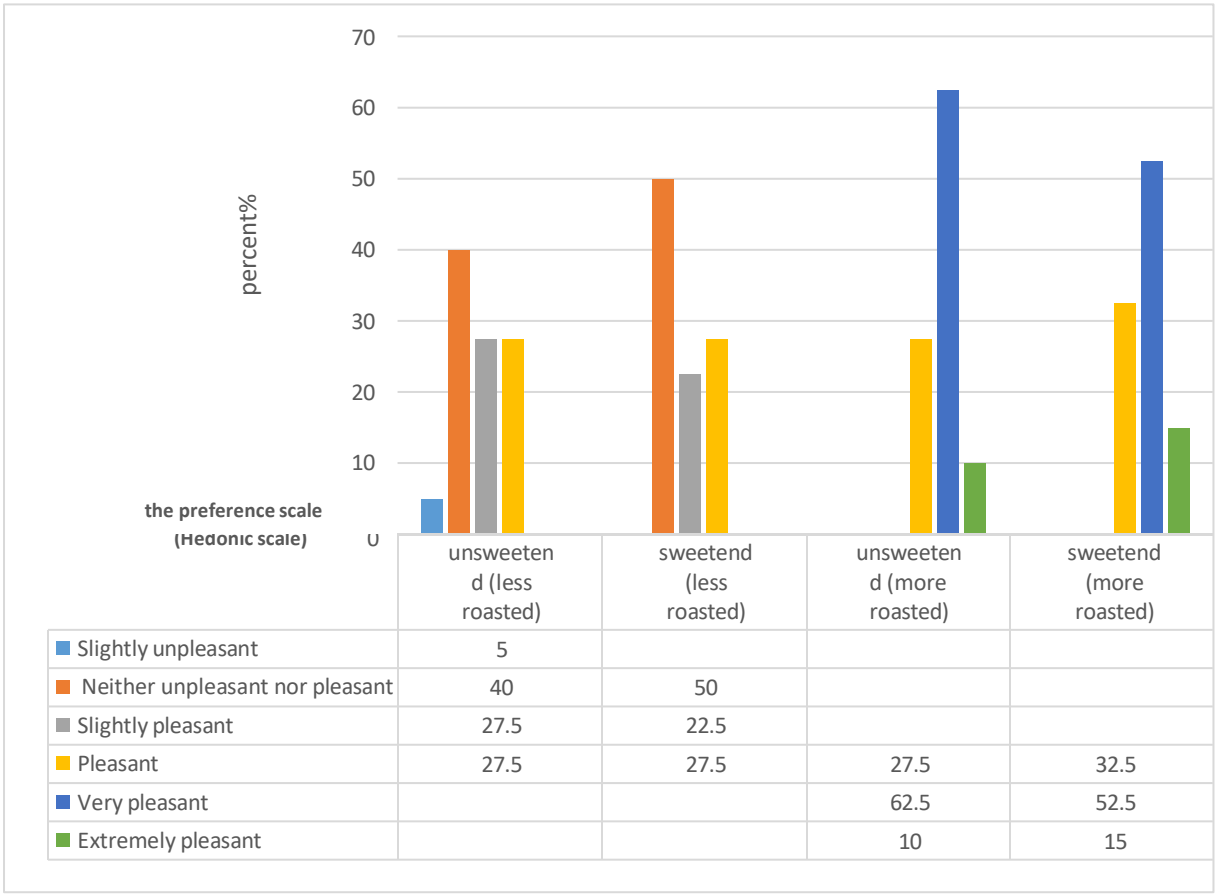


Figure36: Hedonic evaluation of date pit coffee substitutes with varying roasting and sweeting levels

Conclusion

The valorization of by-products from the agri-food industry represents a relevant approach to the sustainable management of agricultural resources. The use of date pits in the development of a coffee substitute beverage aligns with this strategy, offering an innovative and efficient way to repurpose residues from date processing.

In this context, our work centers on the valorization of plant-based waste, with a particular focus on date pits as a potential coffee substitute. This approach involves analyzing the key physicochemical properties of date pit powders and comparing them to those of traditional coffee. Furthermore, the study explores how different roasting processes influence these properties and assesses their impact on the overall quality of the beverage produced from roasted date pits.

The results of the analyses showed that the roasting degree significantly influences the physical, chemical, and microbiological properties of the date seed coffee substitute.

In terms of physical and chemical properties, the highly roasted sample exhibited higher values of pH, ash content, dry matter, and titratable acidity, while the lightly roasted sample had a higher moisture content.

Regarding biochemical properties, the lightly roasted sample contained higher levels of proteins, lipids, total sugars, fibers, polyphenols, and flavonoids.

From a microbiological standpoint, the lightly roasted sample showed greater contamination, particularly with yeasts and molds, indicating the role of roasting in reducing microbial load.

These findings highlight that the degree of roasting has a notable impact on the overall quality of the coffee substitute and emphasize the need to balance biochemical benefits with microbial safety. Sensory analysis revealed that the highly roasted date seed coffee was the closest in taste and aroma to traditional coffee, and it was the most preferred by the panelists. Although the lightly roasted date seed coffee is less similar in flavor and aroma to traditional coffee, it offers superior nutritional benefits due to its higher content of proteins, lipids, sugars, fibers, polyphenols, and flavonoids. These bioactive compounds contribute to better health-promoting properties, making the lightly roasted option a valuable functional beverage despite its sensory differences.

table of references

Wondemagegnehu, B., et al. (2019). The state of global coffee production: Annual statistics. International Coffee Organization.

Benitez, L. M., et al. (2019). Global coffee trade and its commercialization. International Coffee Organization.

Reis, A. G., et al. (2020). Environmental impacts of coffee production and waste management. Sustainable Agriculture Reviews, 32(4), 223-235.

Dos Santos, C. F., et al. (2021). Coffee industry by-products and environmental concerns. Environmental Sustainability, 22(6), 231-245.

Rogers, P. J. (2012). Caffeine and its effect on the central nervous system. Food Research Reviews, 17(2), 115-129.

Faupel, J. P., et al. (2013). Caffeine and health: The risks of daily consumption. Journal of Health and Pharmacology, 15(4), 210-218.

Boussena, S., & Khali, M. (2016). Date palm by-products: Applications and utilization. Journal of Agricultural Science, 4(3), 112-119.

Al-Farsi*, M. A., & Lee, C. Y. (2008). Nutritional and functional properties of dates: a review. *Critical reviews in food science and nutrition*, *48*(10), 877-887.

Alharbi K.L., Raman J., & Shin H.J. (2021). Date Fruit and Seed in Nutricosmetics. Cosmetics, 8(3), 59.1-12.

Al-Farsi M. A., & Lee C. Y. (2011). Usage of date (Phoenix dactylifera L.) seeds in human health and animal feed, 447–452.

Habib, S. H., & Ibrahim, H. (2009). Nutritional enhancement using date pit powder. Journal of Food Science and Nutrition, 22(2), 75-81.

Aldhaheri A., Alhadrami G., Aboalnaga N., Wasfi I., & Elridi M. (2004). Chemicalcomposition of date pits and reproductive hormonal status of rats fed date pits. Foodchemistry, 86, 93-9.*tiers in Physiology*, *13*, 874172.

Djerbi, M. (1994). Précis de phoeniciculture. *Ed. FAO, Rome*, *24*(4).

Besbes, S., Blecker, C., Deroanne, C., Drira, N. E., & Attia, H. (2004). Date seeds: chemical composition and characteristic profiles of the lipid fraction. *Food chemistry*, *84*(4), 577-584.

Lecheb, F. (2010). Extraction et caractérisation physico-chimique et biologique de la matière grasse du noyau des dattes: essai d'incorporation dans une crème cosmétique de soin.

Dobignard, A., & Chatelain, C. (2010). *Index synonymique de la flore d'Afrique du Nord: volumen 3: Dicotyledoneae: Balsaminaceae-Euphorbiaceae*. Éditions des conservatoire et jardin botaniques.

Espiard E. (2002). Introduction à la transformation industrielle des fruits. Ed. Tech et Doc-Lavoisier. p147-155.

Ghnimi, S., Umer, S., Karim, A., & Kamal-Eldin, A. (2017). Date fruit (Phoenix dactylifera L.): An underutilized food seeking industrial valorization. *NFS journal*, *6*, 1-10.

Bouabid, A., Lepreux, S., & Kolski, C. (2019). Design and evaluation of distributed user interfaces between tangible tabletops. *Universal Access in the Information Society*, *18*, 801-819.

Estanova P., 1990 : Note technique : valorisation de la datte. IRFA,CIRAD(France)301- 318p

Al-Farsi*, M. A., & Lee, C. Y. (2008). Nutritional and functional properties of dates: a review. *Critical reviews in food science and nutrition*, *48*(10), 877-887.

Zehdi-Azouzi, S., Cherif, E., Moussouni, S., Gros-Balthazard, M., Abbas Naqvi, S., Ludeña, B, & Aberlenc-Bertossi, F. (2015). Genetic structure of the date palm (Phoenix dactylifera) in the Old World reveals a strong differentiation between eastern and western populations. *Annals of botany*, *116*(1), 101-112.

Ghnimi, S., Almansoori, R., Jobe, B., Hassan, M., & Afaf, K. (2015). Quality evaluation of coffee-like beverage from date seeds (Phoenix dactylifera, L.). *J. Food Process. Technol*, *6*(12), 1-6.

Boudechiche, L., Arab, A., Tahar, A., & Ouzrout, R. (2009). Étude de la composition chimique des noyaux de dattes en vue d'une incorporation en alimentation animale. Livestock Research for Rural Development, 21(5).

Sartini, M., Bragazzi, N. L., Spagnolo, A. M., Schinca, E., Ottria, G., Dupont, C., & Cristina, M. L. (2019). Coffee consumption and risk of colorectal cancer: a systematic review and meta-analysis of prospective studies. *Nutrients*, *11*(3), 694.

Anchisi, C., Maccioni, A. M., Sinico, C., & Valenti, D. (2001). Stability studies of new cosmetic formulations with vegetable extracts as functional agents. *Il Farmaco*, *56*(5-7), 427-431.

Audigier, F. Un geste politique: la poignée de main Mitterrand-Kohl du 22 septembre 1984 à Douaumont. *Témoignage, mémoire et histoire*, 249.

Van Soest, P. V., Robertson, J. B., & Lewis, B. A. (1991). Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal of dairy science*, *74*(10), 3583-3597.

Stephen, A. M., & Phillips, G. O. (2016). *Food polysaccharides and their applications*. CRC press.

Besombes, C., Berka-Zougali, B., & Allaf, K. (2010). Instant controlled pressure drop extraction of lavandin essential oils: Fundamentals and experimental studies. *Journal of Chromatography A*, *1217*(44), 6807-6815.

Brezin, E., Zinn-Justin, J., & Le Guillou, J. C. (1976). Renormalization of the nonlinear σ model in 2+ ε dimensions. *Physical Review D*, *14*(10), 2615.

Ribéreau-Gayon, P. (1972). *Plant phenolics* (No. 3, p. 254pp).

Ouchemoukh, S., Hachoud, S., Boudraham, H., Mokrani, A., & Louaileche, H. (2012). Antioxidant activities of some dried fruits consumed in Algeria. *LWT-Food Science and Technology*, *49*(2), 329-332.

GUIRAUD, J. (2003). Méthode d'analyse en microbiologie alimentaire. *Microbiologie alimentaire. Edition: Dunod, Paris. 651p*.

International Organization for Standardization. (2008). *ISO 6668:2008 – Coffee — Preparation of samples for sensory analysis*.

International Organization for Standardization. (2008). *ISO 6668:2008 – Recommended serving temperature for sensory analysis*. *(Incluse dans la même norme ISO 6668:2008)*

Stone, H., & Sidel, J. L. (2004). *Sensory evaluation practices* (3rd ed.). Academic Press.

Achour, H. Y., & Khali, M. (2014). Composition physicochimique des miels algériens. Détermination des éléments traces et des éléments potentiellement toxiques. *Afrique Science: Revue Internationale Des Sciences Et Technologie*, *10*(2).

Belguedj, N., Mizab, O., & Mesnoua, M. (2023). Morphometric and physicochemical characterization of fruit of seven date palm cultivars cultivated in the in southwest of Al- geria. *Journal Algérien des Régions Arides*, *15*(1), 64-71.

Mebirouk-Boudechiche, L., Cherif, M., Boudechiche, L., & Sammar, F. (2014). Teneurs en composés primaires et secondaires des feuilles d'arbustes fourragers de la région humide d'Algérie. *Revue Méd. Vét*, *165*(11), 344-352.

Alavian, K. N., Beutner, G., Lazrove, E., Sacchetti, S., Park, H. A., Licznerski, P., ... & Jonas, E. A. (2014). An uncoupling channel within the c-subunit ring of the F1FO ATP synthase is the mitochondrial permeability transition pore. *Proceedings of the National Academy of Sciences*, *111*(29), 10580-10585.

Ammar, A., Brach, M., Trabelsi, K., Chtourou, H., Boukhris, O., Masmoudi, L., ... & ECLB-COVID19 Consortium. (2020). Effects of COVID-19 home confinement on eating behaviour and physical activity: results of the ECLB-COVID19 international online survey. *Nutrients*, *12*(6), 1583.

Nabili, A., Fattoum, A., Passas, R. A. P. H. A. Ë. L., & Elaloui, E. L. I. M. A. M. E. (2016). Extraction and characterization of cellulose from date palm seeds (Phoenix dactylifera L.). *Cellul. Chem. Technol*, *50*(9-10), 1015-1023.

O'Connor, R. E., Al Ali, A. S., Brady, W. J., Ghaemmaghami, C. A., Menon, V., Welsford, M., & Shuster, M. (2015). Part 9: acute coronar

Wilson, W. M., & Maughan, R. J. (1992). Evidence for a possible role of 5-hydroxytryptamine in the genesis of fatigue in man: administration of paroxetine, a 5-HT re-uptake inhibitor, reduces the capacity to perform prolonged exercise. *Experimental Physiology: Translation and Integration*, *77*(6), 921-924.

Mansour, A. T., Ashour, M., Abbas, E. M., Alsaqufi, A. S., Kelany, M. S., El-Sawy, M. A., & Sharawy, Z. Z. (2022). Growth performance, immune-related and antioxidant genes expression, and gut bacterial abundance of Pacific white leg shrimp, Litopenaeus vannamei, dietary supplemented with natural astaxanthin. *Frontiers in Physiology*, *13*, 874172.

Abstract

This study evaluated date pits as a healthy coffee substitute, focusing on how roasting intensity affects their properties. Less and more roasted date pit powders were compared to Robusta coffee. less roasting preserved more fiber, unsaturated fatty acids, and antioxidants, while more roasting reduced microbial counts. Protein content was similar to coffee, but polyphenols and flavonoids were lower. Sensory tests favored lightly roasted samples for their milder taste, while sweetening improved overall acceptance. Date pit powder, especially when lightly roasted, shows strong potential as a nutritious and safe coffee alternative.

Key words : Date pits, coffee substitute, less and more roasting, sensory evaluation, nutritional properties.

Résumé

Cette étude a évalué les noyaux de dattes comme substitut sain au café, en analysant l'effet de l'intensité de torréfaction. Deux niveaux de torréfaction (légère et forte) ont été comparés au café Robusta. La torréfaction légère a mieux préservé les fibres, les acides gras insaturés et les antioxydants, tandis que la torréfaction forte a réduit la charge microbienne. La teneur en protéines était similaire à celle du café, mais les polyphénols et flavonoïdes étaient plus faibles. Les tests sensoriels ont favorisé la torréfaction légère pour sa saveur douce, et le sucrage a amélioré l'acceptabilité. Les noyaux de dattes torréfiés légèrement présentent un fort potentiel comme alternative nutritive et sûre au café.

Les mots clé : Noyaux de dattes, substitut de café, torréfaction légère et forte, évaluation sensorielle, propriétés nutritionnelles.

الملخص

هدفت هذه الدراسة إلى تقييم إمكانية استخدام نوى التمر كبديل صحي للقهوة، من خلال دراسة تأثير درجة التحميص على خصائصها. تم مقارنة نوى التمر المحمصة تحميصًا خفيفًا وثقيلًا مع قهوة الروبوستا. أظهر التحميص الخفيف احتفاظًا أكبر بالألياف والأحماض الدهنية غير المشبعة والمضادات الأكسدة، بينما قلل التحميص الثقيل من الحمل الميكروبي. كانت نسبة البروتين قريبة من القهوة، لكن محتوى البوليفينولات والفلافونويدات كان أقل. فضّلت التقييمات الحسية النوى المحمصة بخفة لمذاقها المعتدل، كما زاد التحلية من تقبّلها. تظهر نوى التمر، خاصة عند تحميصها بخفة، إمكانات قوية كبديل مغذٍ وآمن للقهوة.

الكلمات المفتاحية: نوى التمر، بديل القهوة، التحميص الخفيف والثقيل، التقييم الحسي، الخصائص الغذائية.

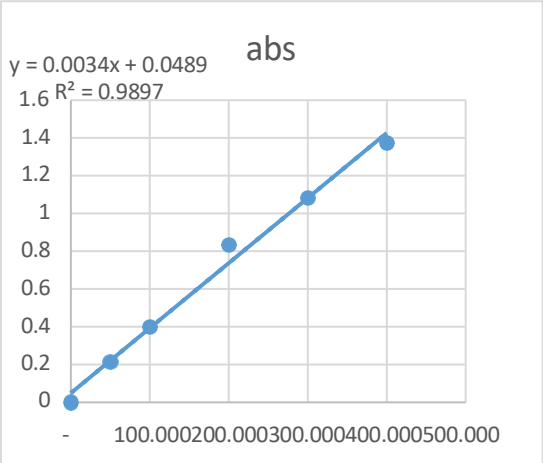
The annex

	Coffee Robusta	More roasted date pits	Less roasted date pits	*P*
pH	6.17 ± 0.094	5.95 ± 0.019	5.94 ± 0.074	> 0.05
Moisture Content	4.00 ±0.100	1.68 ± 0.525	2.92 ± 0.584	> 0.002
Dry Matter Content	96.00 ±0.100	98.31 ± 0.525	97.07 ± 0.584	> 0.002
Ash	4.33 ±0.152	1.06 ± 0.020	0.90 ± 0.100	> 0.001
Titratable Acidity	0.41±0.025	0.32 ± 0.020	0.36 ± 0.010	> 0.003
Cellulose (g/100g)	10.17 ± 0.011	10.97 ± 0.000	11.42 ± 0.000	> 0.001
Hemicellulose (g/100g)	12.52 ± 0.000	13.63 ± 0.000	14.06 ± 0.011	> 0.001
Lignin (g/100g)	18.68 ± 0.005	19.14 ± 0.005	19.96 ± 0.005	> 0.001
Pectin (g/100g)	3.33 ± 0.005	3.81 ± 0.005	4.24 ± 0.005	> 0.001
Saturated fatty acid**(g/100g)**	4.30±0.015	3.14 ± 0.017	3.86 ± 0.015	> 0.001
Palmitic acid (g/100g)	2.73±0.005	1.74 ± 0.011	2.13 ± 0.037	> 0.001
Steric acid**(g/100g)**	1.50±0.020	1.39± 0.005	1.70 ± 0.020	> 0.001
Mono unsaturated fatty acid**(g/100g)**	2.20±0.025	2.88 ± 0.005	3.42 ± 0.000	> 0.001
Oleic acid**(g/100g)**	2.18±0.020	2.85± 0.000	3.36 ± 0.010	> 0.001
Poly unsaturated fatty acid **(g/100g)**	1.51±0.011	2.26 ± 0.005	3.100 ± 0.010	> 0.001
Linoleic acid(g/100g)	1.41±0.005	1.76 ± 0.015	2.52± 0.015	> 0.001
Linolenic acid(g/100g)	Traces<0.1	0.49 ± 0.010	0.56± 0.005	> 0.001

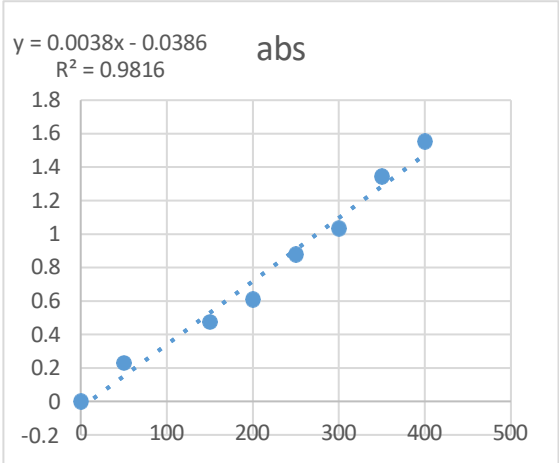
The annex

Protein(g/ 100g)	10.87 ± 0.005	9.79 ± 0.005	10.67 ± 0.032	> 0.001
Total sugars (mg / g sample)	0.015 ± 0.0072	0.003 ± 0.0042	0.016 ± 0.003	>0.05
TPC (µgGAE / g sample)	111.15 ± 1.15	60.53 ± 15.88	77.75 ± 6.13	>0.01
Flaco noids(mg QE/ g sample)	71.27±8.88	20.42±3.02	38.31±5.94	> 0.001

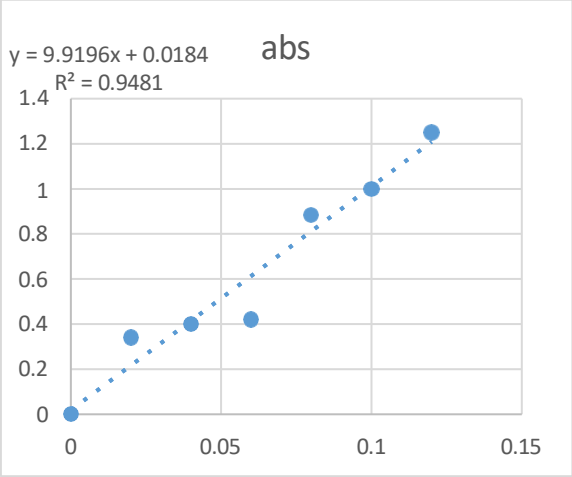
Annex 1: Results table



Annex2: calibration curve of flavonoides



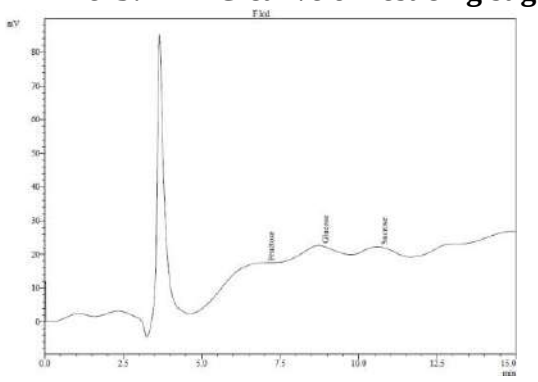
annex3 :calibration curve of polyphenoles



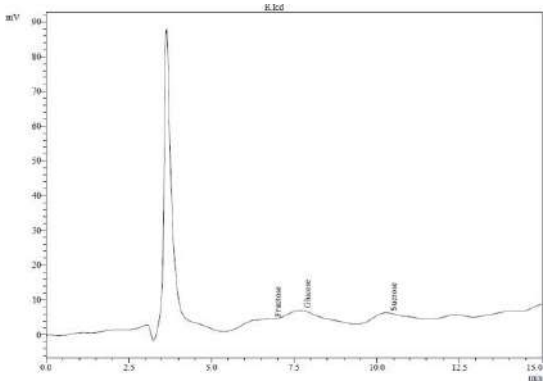
Annex4: calibration curve of total sugars

The annex

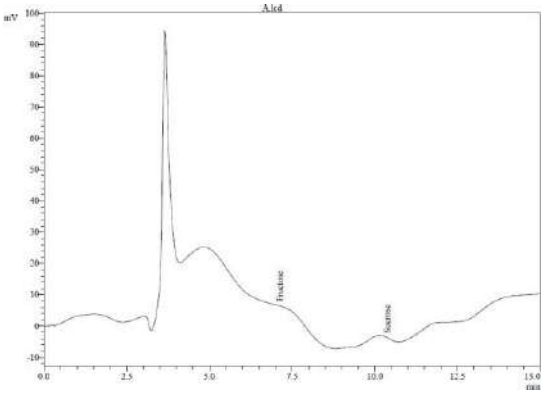
Annex5: HPLC curve of resucing sugars



More roasted curve



Robusta coffee curve



Less roasted curve

Annex6: lab equipment

HPLC

muffle furnace

The annex

oven

Pasteur oven

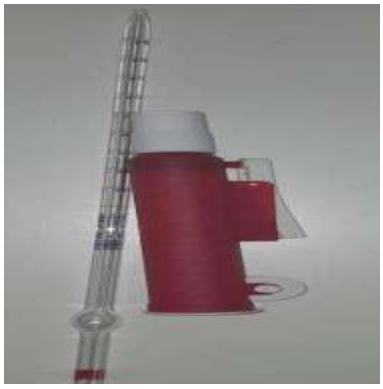
bain-marie

spectrophotometer

hot plate

microbiological pipette

The annex



Pipette

electric scale

Autoclave

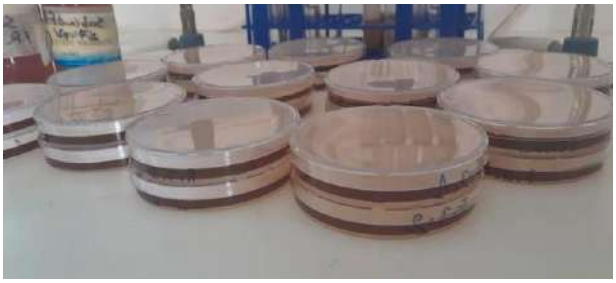
desiccator

Coffee grinding

the vats

The annex

the vats



inoculation of yeasts and molds in petri dishes

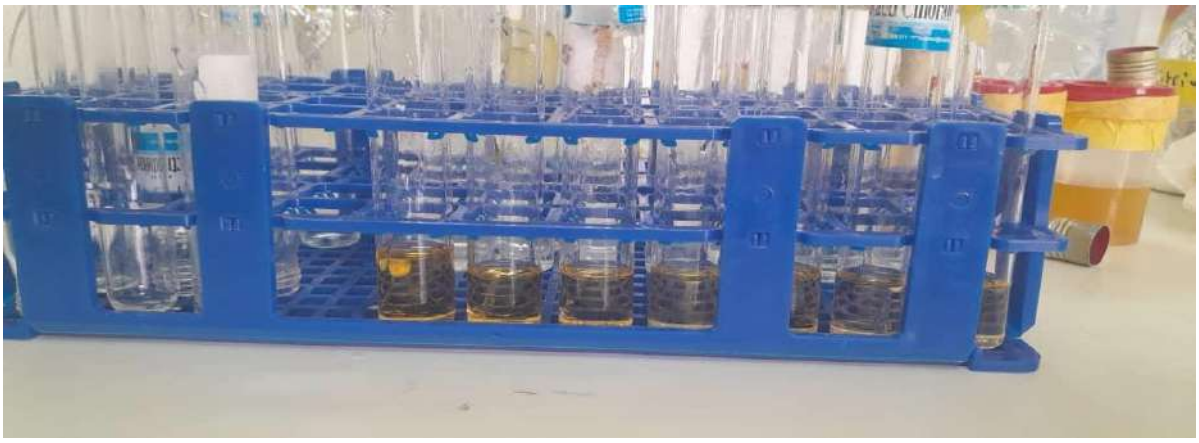
Sabouraud environment

Sabouraud environment preparation

pH meter

The annex

samples diluted in water



example of the range: total sugars

annex7: range results: fibers

	Ech 1 : R	Ech 2 : plus torréfié	Ech 3: moins torréfié
Cellulose	10.16	10.97	11.42
	10.18	10.97	11.42
	10.18	10.97	11.42
Hémicellulose	12.52	13.63	14.07
	12.52	13.63	14.07
	12.52	13.63	14.05
Lignine	18.68	19.15	19.96
	18.69	19.15	19.97
	18.69	19.14	19.96
Pectine	3.34	3.82	4.25
	3.33	3.81	4.24
	3.33	3.81	4.24

The annex

Lipids:

	Ech 1 : R	Ech 2 : plus torréfié	Ech 3: moins torréfié
Acides gras saturés	4.32	3.16	3.88
	4.29	3.13	3.87
	4.31	3.13	3.85
Acide palmitique	2.74	1.75	2.16
	2.74	1.73	2.15
	2.73	1.75	2.09
Acide stéarique	1.50	1.40	1.69
	1.49	1.39	1.70
	1.53	1.40	1.73
A laurique et A myristique	Traces (<0.1)	Traces (<0.1)	Traces (<0.1)
AG Monoinsaturés	2.23	2.89	3.42
	2.20	2.89	3.42
	2.18	2.88	3.42
Acide oléique	2.20	2.85	3.36
	2.18	2.85	3.37
	2.16	2.85	3.35
Acide palmitoléique	Traces (<0.1)	Traces (<0.1)	Traces (<0.1)
AG Polyinsaturés	1.52	2.27	3.11
	1.52	2.26	3.09
	1.50	2.27	3.10
A Linoléique	1.50	1.78	2.54
	1.50	1.76	2.51
	1.49	1.75	2.52
A Linolénique	Traces (<0.1)	0.48	0.56
	Traces (<0.1)	0.49	0.57
	Traces (<0.1)	0.50	0.57

Proteins:

	Ech 1 : R	Ech 2 : F	Ech 3: A
Protéines	10.87	9.79	10.71
	10.88	9.80	10.65
	10.88	9.79	10.66

Annex 8:

The annex

Questionnaire d`analyses sensorielles (plan expert)

Date :.....

Age :..... Sexe : féminin ☐ masculin ☐

Vous disposez de neuf échantillons de café identifiés par les codes A à D. Il vous est demandé de les déguster successivement et d'évaluer chaque échantillon en attribuant une note à chacune des caractéristiques sensorielles décrites, selon votre impression.

NB : veuillez rincer votre bouche à chaque dégustation d`un échantillon

La couleur :

A/ **1) marron clair 2) marron 3) marron foncé 4) marron très foncé 5) noir**

Échantillon	A	B	C	D
Note				

B/ **1) satisfaisant 2) non satisfaisant**

Échantillon	A	B	C	D
Note				

L`odeur :

A/ **1)Très forte 2) forte 3) moyenne 4faible 5) très faible**

Échantillon	A	B	C	D
Note				

B/ **1) satisfaisant 2) non satisfaisant**

Échantillon	A	B	C	D
Note				

Goût amère :

A/ **1)Très forte 2) forte 3) moyenne 4faible 5) très faible**

Échantillon	A	B	C	D
Note				

B/ **1) satisfaisant 2) non satisfaisant**

Échantillon	A	B	C	D
Note				

Goût sucré :

A/ **1)Très forte 2) forte 3) moyenne 4faible 5) très faible**

Échantillon	A	B	C	D

The annex

Note				

B/ **1) satisfaisant** **2) non satisfaisant**

Échantillon	A	B	C	D
Note				

Goût acide :

A/ **1)Très forte 2) forte 3) moyenne 4faible 5) très faible**

Échantillon	A	B	C	D
Note				

B/ **1) satisfaisant** **2) non satisfaisant**

Échantillon	A	B	C	D
Note				

Goût astringent :

A/ **1) Très forte 2) forte 3) moyenne 4faible 5) très faible**

Échantillon	A	B	C	D
Note				

B/ **1) satisfaisant** **2) non satisfaisant**

Échantillon	A	B	C	D
Note				

Rémanence (goût persistent de café) :

A/ **1)Très forte 2) forte 3) moyenne 4faible 5) très faible**

Échantillon	A	B	C	D
Note				

B/ **1) satisfaisant** **2) non satisfaisant**

Échantillon	A	B	C	D
Note				

Arôme ou gout identifié :

A/ **1) Fade 2) Grillé 3) Epicé 4) Brulé 5) Fruité**

Échantillon	A	B	C	D
Note				

B/ **1) satisfaisant** **2) non satisfaisant**

The annex

Échantillon	A	B	C	D
Note				

Préférence globale :

Attribuez à chaque échantillon une note de préférence comprise entre 1 et 9, où 1 correspond à l'échantillon le moins apprécié et 9 à l'échantillon le plus apprécié, selon l'échelle suivante :

1 : Extrêmement désagréable
2 : Très désagréable
3 : Désagréable
4 : Assez désagréable
5 : Ni agréable ni désagréable
6 : Assez agréable
7 : Agréable
8 : Très agréable
9 : Extrêmement agréable

Échantillon	A	B	C	D
Note				

Merci pour votre coopération

1. Échantillon A (noyaux torréfiés)

- **A1** : Pur (sans sucre)
- **A2** : une cuillère de sucre

2. Échantillon F (noyaux rôtis)

- **F1** : Pur (sans sucre)
- **F2** : une cuillère de sucre

Protcols:

1/**Moisture Content Determination :**

An amount of 5g of the sample is weighed and placed in a drying oven at 105°C for 24 hours. After drying, the sample is transferred to a desiccator to cool down to room temperature and to achieve a constant weight.

2/Ash:

A total of 0.4 g of the sample is dried in an oven at 105°C. The dried sample is then divided equally into four crucibles, with 0.1 g placed in each.

The crucibles are placed in a muffle furnace at 600C for 3 hours until the sample becomes fully ashed. The resulting ash should appear white or gray in color (Barkhatov & Elissev, 1979)

3/ PH:

weighed 1 g of date seed powder, added 10 mL of distilled water, let the mixture sit for 30 minutes, then measured the pH using a calibrated pH meter by immersing the electrode in the extract,
ensuring to rinse and dry the probe between two successive readings.

4/Acidty titrable:

prepared an aqueous suspension by dissolving 10 g of date seed powder in 100 mL of distilled water, then titrated it with 0.1 N sodium hydroxide using phenolphthalein as an indicator until a light pink color appeared, and expressed the titratable acidity in grams of citric acid per 100 g of product.1.

5/Flavnoid:

Imixed 1 mL of date seed extract with 1 mL of 2% aluminum chloride solution, incubated the mixture for 10 minutes, then measured the absorbance at 410 nm using a UV-Vis spectrophotometer, and
calculated the flavonoid content based on a calibration curve prepared with rutin, expressing the results as milligrams of rutin equivalents per 100 grams of date seed powder.

6/Quantitative Sugar:

Materials and Reagents

Phenol

Sulfuric acid (95–97%)

5% (w/v) aqueous phenol solution

Glucose

Water bath

Spectrophotometer.

Preparation of the 5% Phenol Solution:

Dissolve 5 g of phenol in 100 mL of distilled water. Mix thoroughly until fully dissolved. The resulting solution should be colorless and transparent, and it remains stable at room temperature.

2. Procedure

1. In a Pyrex test tube, add:

1 mL of the sample solution to be

analyzed 1 mL of the 5% phenol

solution

After mixing, add 5 mL of concentrated sulfuric acid (H_2SO_4, 95–98%) to the mixture.

Incubate the tubes in the dark at room temperature for 30 minutes.

Measure the absorbance at 490 nm using a spectrophotometer.

7/Polyphénols:

Materials and Reagents

Folin–Ciocalteu reagent.

Sodium carbonate .

10% aqueous sodium carbonate solution.

Gallic acid .

Ultrapure water.

Samples to be analyzed (concentration between 1–10 g/L, with dilutions as needed).

Spectrophotometer .

Procedure

Dilute 0.5 mL of each sample in 10 mL of ultrapure water.

Add 0.5 mL of Folin–Ciocalteu reagent to each tube and let the mixture stand for 3 minutes.

Add 2 mL of saturated Na_2CO_3 solution and mix for 10 seconds.

Incubate the test tubes in the dark for 30 min , until a blue coloration develops.

Measure the absorbance at 750 nm using a spectrophotometer.

8/Detection and Enumeration of Yeasts and molds

Materials and Media Required

Stock solution (coffee extract)

Sterile diluent: physiological saline (0.85% NaCl) Sterile test tubes with 9 mL diluent

Sabouraud Dextrose Agar (SDA)

Sterile spreader

Micropipette + sterile tips

Incubator set at 25°C

Sterile Petri dishesallic acid is used as the reference standard at concentrations ranging from 0 to 0.2 g/L.

Preparation of the Stock Solution ("Solution Mère")

1. Weigh 10 grams of the coffee powder (produced from date pits).

2. Add 90 mL of sterile distilled water or Buffered Peptone Water (BPW) into a sterile flask or container.

3. Mix thoroughly to homogenize the solution (using a shaker or manual agitation).

4. Let the mixture sit for a few minutes to allow solid particles to settle, or filter using sterile gauze if needed.

5. The resulting liquid is used as the stock solution (dilution 10^0) for the serial dilution processdure

6. 1. Serial Dilution

Under sterile conditions, transfer 1 mL of the stock solution into a test tube containing 9 mL of sterile diluent.

Mix thoroughly to obtain the first 10^{-1} dilution.

Transfer 1 mL of this dilution into another test tube with 9 mL of diluent to obtain 10^{-2}.

Repeat the process sequentially to obtain the desired dilution series (e.g., up to 10^{-4} or 10^{-6}).

2. Plating and Incubation

From each selected dilution (e.g., 10^{-1}, 10^{-2}...), transfer 0.1 mL onto the surface of a sterile Sabouraud Agar plate.

2. Spread evenly using a sterile spreader.

3. Incubate the plates at 25°C for 5 days.

2- Lipid Determination

 Detailed protocol for lipid determination using the Soxhlet method;

Objective:

To extract and quantify the lipid content in date pits used as a coffee substitute.

Materials and Reagents:

Materials:

Soxhlet extractor

250 mL flask

Heating mantle

Filter paper or extraction cartridge

Beakers (100 mL and 250 mL)

Analytical balance (accuracy 0.0001 g)

Desiccator

Oven (105°C)

Mortar and pestle or grinder

Graduated pipette

Solvent:

n-Hexane (or petroleum ether, depending on availability)

Experimental Procedure:

Sample Preparation:

Dry the date pits at 105°C for 24 hours to remove all moisture.

Finely grind the dried kernels using a grinder or mortar and pestle.

Accurately weigh 5 to 10 g of date kernel powder (0.0001 g).

Setting up the Soxhlet extraction:

Place the powder in an extraction thimble (or folded filter paper).

Insert the thimble into the Soxhlet extraction chamber.

Fill the Soxhlet flask with 150 to 220 mL of n-hexane.

Assemble the extractor, condenser, and heating mantle.

Lipid extraction:

Heat gently to boil the solvent (n-hexane boiling point: 68-70°C).

Allow the extraction to proceed for 4 to 6 hours, with approximately 6 to 10 siphonings per hour. • Once the extraction is complete, allow to cool and then disassemble the apparatus.

Solvent evaporation and lipid weighing:

Transfer the lipid extract into a pre-weighed beaker.

Evaporate the solvent in a water bath (50-60°C) under a fume hood or on a rotary evaporator.

Dry the extracted lipids at 105°C for 1 hour to remove any solvent residue.

Cool in a desiccator and weigh immediately.

Calculation of lipid content (%)

 Transesterification of lipids into FAMEs (Fatty Acid Methyl Esters)

The objective of this step is to convert the extracted fatty acids in the form of triglycerides into methyl esters (FAMEs), which are volatile and therefore suitable for GC-MS analysis.

Preparation of the Methylation Solution

In a clean beaker, prepare an anhydrous methanol solution by adding a catalyst for each sample:

Methanol/HCl (5%): Add 0.5 mL of concentrated hydrochloric acid to 10 mL of methanol.

Methanol/NaOH (0.5 M): Dissolve 20 mg of NaOH in 10 mL of methanol.

Methylation Reaction

Weigh 100 mg of extracted lipids for each sample and place them in a sealed glass tube.

Add 4 mL of the methylation solution (HCl-methanol or NaOH-methanol).

Heat at 60°C for 1 hour in a water bath with gentle stirring.

Cool to room temperature.

FAME Extraction

Add 5 mL of heptane or chloroform to extract the FAMEs. • Add 1 mL of saturated NaCl solution to facilitate phase separation.

Centrifuge at 3000 rpm for 5 minutes.

Collect the organic phase (upper layer if heptane, lower layer if chloroform).

Dry with 0.5 g of anhydrous sodium sulfate to remove all traces of water.

Filter and transfer to a GC-MS vial.

GC-MS Analysis

Chromatographic Conditions

Column: DB-23 or DB-5MS (30 m × 0.25 mm × 0.25 µm)

Carrier gas: Helium, flow rate 1 mL/min

Temperature program:

50°C (1 min)

10°C/min → 180°C (5 min)

3°C/min → 230°C (10 min)

Injector: 250°C, 10:1 split mode

Ionization source: 230°C

Acquisition mode: SCAN (50-500 m/z) or SIM for specific quantification.

Identification and Quantification

Compare peak retention times with a standard mixture of FAMEs. • Identify the main fatty acids present:

Palmitic acid (C16:0)

Stearic acid (C18:0)

Oleic acid (C18:1)

Linoleic acid (C18:2)

Calculate the relative composition (%) of each fatty acid by integrating the areas under the peaks.

Expected observations

After methylation and FAME extraction:

Organic solution clear to slightly yellowish after filtration.

Aqueous phase cloudy due to phase separation.

After GC-MS analysis:

Chromatogram showing several distinct peaks corresponding to FAMEs.

Identification of fatty acids based on their retention times and mass spectra.

Kjeldahl Protein Determination Protocol

Principle

The Kjeldahl method involves:

Digestion: Conversion of nitrogen in organic matter into ammonium sulfate using sulfuric acid and a catalyst.

Distillation: Ammonia is released by alkalization and distilled into a boric acid solution.

Titration: The amount of ammonia (and thus nitrogen) is determined by titration with standard acid.

Materials & Reagents

Equipment

Kjeldahl digestion unit

Distillation unit (manual or automatic)

Burette

Analytical balance

Reagents

Concentrated H_2SO_4 (sulfuric acid)

Catalyst mixture (e.g., K_2SO_4 + $CuSO_4$ or selenium)

Sodium hydroxide solution (40% NaOH)

Boric acid solution (4% H_3BO_3)

Mixed indicator (bromocresol green + methyl red)

Standard HCl or H_2SO_4 (usually 0.1 N)

Procedure

Digestion

Weigh **0.5–1.0 g** of the finely ground dry sample into a Kjeldahl digestion flask.

Add **10–15 mL** of concentrated H_2SO_4 and a **catalyst tablet** or ~0.5 g of catalyst mixture.

Heat gently at first, then strongly until the solution becomes clear (about **2–3 hours**).

Cool the digested sample.

Distillation

Add **excess 40% NaOH** (carefully) to make the solution strongly alkaline.

Immediately distill the released ammonia into **25–50 mL of 4% boric acid** containing the mixed indicator.

Collect the distillate until all ammonia is recovered (about **150 mL** distillate).

Titration

Titrate the boric acid solution with **0.1 N HCl** until the endpoint (color changes from green to pink).

Perform a blank determination using the same procedure but without the sample.

Calculations

% Nitrogen

% Protein

Méthode de Van Soest (méthode des détergents)

C'est la méthode la plus utilisée pour fractionner les fibres végétales. Elle permet de doser :

NDF (Neutral Detergent Fiber – fibres au détergent neutre) :

Contient **cellulose + hémicellulose + lignine**

Permet de séparer les parois cellulaires (fibres totales) du contenu cellulaire soluble.

ADF (Acid Detergent Fiber – fibres au détergent acide) :

Contient **cellulose + lignine**

Hémicellulose = NDF – ADF

Lignine (ADL) :

Obtenue en traitant le résidu ADF avec **acide sulfurique concentré (72 % H_2SO_4)**.

Cellulose = ADF – ADL

Résumé des calculs :

Hémicellulose = NDF – ADF

Cellulose = ADF – ADL

Lignine = ADL

Matériel	Description / Usage

Système d'extraction à reflux	Type **Fibertec™**, ou système de Soxhlet modifié, pour les extractions NDF, ADF et ADL
Creusets filtrants en verre fritté (Gooche)	Pour la filtration et la pesée des résidus
Pompe à vide ou système de filtration sous vide	Pour accélérer la filtration des résidus
Bouteilles ou flacons de digestion (500–1000 mL)	Pour faire bouillir les échantillons dans les solutions détergentes
Agitateur magnétique avec plaque chauffante	Pour un chauffage et une agitation homogène
Hotte aspirante	Pour manipuler l'acide sulfurique concentré en toute sécurité
Balance analytique (± 0,1 mg)	Pour les pesées précises des échantillons et résidus
Bain-marie ou réacteur à température contrôlée	Pour le chauffage à température constante (environ 100 °C)
Thermomètre	Pour vérifier la température d'extraction
Pinces, spatules, béchers, entonnoirs, ciseaux	Accessoires de laboratoire généraux
Tamis (1 mm)	Pour homogénéiser la taille des particules de l'échantillon
Réactif	**Usage**
Solution de détergent neutre (NDS)	Pour extraire le contenu cellulaire ; contient **borate de sodium, EDTA, SDS, disulfure de sodium**
Solution de détergent acide (ADS)	Pour extraire l'hémicellulose ; contient **acide sulfurique (1 N) + bromure de cétalkyltriméthylammonium (CTAB)**
Acide sulfurique concentré (72 %)	Pour solubiliser la cellulose et isoler la lignine (étape ADL)
Acétone	Pour le rinçage des résidus après digestion
Eau distillée	Pour les dilutions, rinçages et extractions
Tampon pH neutre (optionnel)	Pour ajuster les solutions si nécessaire

Méthode de dosage de la pectine (méthode gravimétrique par précipitation alcoolique)

Principe :

Extraction de la pectine avec de l'eau chaude acide → précipitation à l'alcool → filtration → séchage → pesée.

Matériel nécessaire

Matériel	Description / Usage
Béchers (250–500 mL)	Pour les extractions
Agitateur magnétique + plaque chauffante	Pour maintenir l'ébullition douce lors de l'extraction
Thermomètre	Pour contrôler la température (~85–90 °C)

Centrifugeuse (ou filtration classique)	Séparation du liquide après extraction
Filtre à papier / creuset fritté	Pour récupérer le précipité de pectine
Balance analytique	Pour pesée précise du résidu sec
Étuve (105 °C)	Pour sécher le précipité de pectine
Pipettes, éprouvettes, spatules, pinces	Accessoires divers

Réactifs nécessaires

Réactif	Rôle
Acide oxalique ou acide citrique (0,1–0,2 N)	Agent d'extraction doux, à chaud
Alcool éthylique (95 % ou absolu)	Précipite la pectine en solution
Eau distillée	Pour préparer les solutions et rincer les résidus
Chlorure de calcium (optionnel)	Parfois utilisé pour stabiliser les pectines
Acide sulfurique (pour méthode colorimétrique alternative)	Si dosage par méthode colorimétrique au carbazole (quantification des acides uroniques)

Résumé des étapes (méthode gravimétrique)

Extraction :
Chauffer l'échantillon dans une solution d'acide oxalique/citrique à 85–90 °C pendant 1 h.
Filtration ou centrifugation du mélange pour récupérer le filtrat (extrait de pectine).
Précipitation :
Ajouter 2 à 3 volumes d'éthanol au filtrat → formation d'un précipité gélatineux.
Filtration et lavage du précipité à l'alcool.
Séchage à l'étuve (105 °C) jusqu'à masse constante.
Pesée du résidu sec = **pectine totale extraite (g/100 g échantillon sec)**.

Méthode alternative : Colorimétrie au carbazole

Permet de doser **les acides galacturoniques** (constituants de la pectine).
Plus sensible, utile pour faibles concentrations.
Spectrophotomètre requis (lecture à 530 nm).