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STUDY OF THE EFFECT OF STORAGE ON THE NUTRITIONAL QUALITY OF PAPRIKA SPICE

Supported on: 07/11/2024 **In front of the Jury composed by:**

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THÈSE

Présentée par

BOUZIANE-AIT BESSAI Keltoum

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Thème

ETUDE DE L'EFFET DU STOCKAGE SUR LA QUALITÉ NUTRITIONNELLE DE LA POUDRE DE PIMENT ROUGE

Soutenue le : 07/11/2024 **Devant le Jury composé de :**

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Dedication

I dedicate this work:

To my dear parents

To my dear husband

To my loving children; Amine, Ghiles and Adam

To my father-in-law and

the soul of my mother-in-law

Keltoum

PUBLICATIONS AND PRESENTATIONS RELATED TO THE THESIS

Scientific publications

- **Published papers:**

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- **Communications:**

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List of abbreviations

AAS: Atomic Absorbance Spectrophotometry

ANOVA: Analysis Of Variance

ATCC: American Type Culture Collection

BBD : Box-Behnken Design

CFU: Colony Forming Unit

C.V: Coefficient of Variation

DF: Degree of freedom

DIZ: Diameter of the Inhibition Zone

DPPH: 2,2-diphenyl -1-picrylhydrazyl reagent

DMSO: Di -Méthyl- Sulf- Oxyde

GAE: Gallic Acid Equivalents

HDPE: High-Density Polyethylene

IC50: Half-maximal inhibitory concentration

JMP: John's Macintosh Project, pronounced “jump”

LDPE: Low-Density Polyethylene

MHA: Mueller Hinton Agar

MHB: Mueller Hinton Broth

MIC: Minimum inhibitory Concentration

MLC: Minimum Lethal Concentration

MBC: Minimum Bactericidal Concentration

UAE: Ultrasound assisted extraction

pH :potential of Hydrogen

PPO : Polyphenol Oxydase

RSM: Response surface methodology

RMSE: Root Mean Square Error

R2 : Coefficient of determination

TF: total flavonoids

TPC: total phenolic content

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General introduction

General introduction

Plants have always been the primary source of food and medicine for humans (Pelt 2002). Plant extracts have received high interest as antimicrobials, flavor enhancers, preservation agents, and nutraceutical ingredients in the food industry (Marty-Dufaut 2015).

Worldwide, consumption of Capsicum fruit, probably one of the world's leading spices or food additives, is increasing all the time. Its importance stems from the different uses to which it is put. In many countries, it has played an important role in human culture since prehistoric times (P. W. Bosland, Votava, and Votava 2012). It is the source of an important condiment of high commercial and medicinal value, with antioxidant, antibacterial, anti-cancer and many other properties.

The red pepper (*Capsicum annuum L.*) (Zhang, Lu, and Dárcy 2002) is an important agricultural crop, not only because of its economic importance, but also because of its richness in antioxidants, mainly due to the fact that it is an excellent source of ascorbic acid, natural colourings and other antioxidant compounds. Exploring antimicrobial properties along with the antioxidant activity of the plant are an important aspect as there is a growing need to replace existing synthetic food additives with those of natural origin (P. W. Bosland, Votava, and Votava 2012).

In recent years, it has been recognized that many spices not only have properties that make food more pleasant and tasty, but also have important preservative and antioxidant properties. The antioxidant properties of many spices are well known, while their prooxidant properties are less so (Embuscado 2015).

Spices or food additives consumption , are steadily increasing. They widely used in food products to improve their sensory properties (Molnár, Bata-Vidács, et al. 2018). Among condiments, paprika is the most important one consumed throughout the world and used for flavoring (L.-Z. Deng et al. 2018).

Paprika displays potent biological activities such as antioxidant, anti-cancers, stimulation of the immune system, and prevention of cardiovascular diseases and delays the aging process (Batiha et al. 2020; Salehi et al. 2018). Sometimes it referred as chilli pepper, a characteristic red seasoning powder obtained from the drying and grinding of certain varieties of red peppers such as *Capsicum annuum* L. Its cultivars have been identified as potential solanaceous crop (Cetó et al. 2018) with important economically and nutritionally interests because of their richness in natural colors and antioxidants. In addition to their sensory features, they are important ingredients of a balanced diet thanks to the presence of diverse bioactive compounds with potential functional properties (Mokhtar et al. 2016).

Fruit pepper is transformed into paprika powder after being dried. Indeed, drying is used to remove moisture from fresh fruits and vegetables in order to preserve them against microorganisms and enzymes degradation which may occur during the growing season such as improper agrotechnical treatments or storage (Shirkole et al. 2021). Fruit pepper and other vegetables are prone to microbial spoilage because of their succulent nature. Postharvest handling and storage practices have a major impact on the quality and shelf-life of peppers and quality of paprika powder.

In this thesis we will focus our research on paprika, which is traditionally prepared in the Kabylie region of Algeria; women prepare this spice from locally grown chilli peppers.

The main objectives of this study are:

1) Physicochemical characterisation of traditionally-obtained paprika powder and the use of RSM in order to determine the conditions that would maximize the yields of total phenolics extracted by UAE.

2) Evaluate, the effect of three methods conservation: refrigeration (4 °C) and freezing (-18 °C) processes on the phenolic contents and biological activities (antioxidant and antibacterial) of the paprika powder stored. The obtained results were compared to those resulted from stored samples at room temperature.

Thesis contains two parts, namely: the first part presents a literature study on the subject, while the second deals with the experimental.

The literature study is further structured into two chapters. **Chapter I** present general botany, processing and preservation technology of paprika **Chapter II** present phytochemistry and biological role of paprika.

The experimental part is divided into two parts. In **first,** the research performed for characterization of paprika powder for its humidity, pH, color parameters and mineral composition with the use of Atomic Absorbance Spectrophotometry (AAS), the optimization of extraction of polyphenols from paprika is presented in this chapter in order to establish an extraction protocol that minimizes extraction time, minimizes error due to conventional extraction and standardizes extraction conditions to facilitate tracking of the material during storage. Description the ultrasound assisted extraction (UAE) of polyphenols from paprika, considering the influence of various parameters on the process. UAE of phenolic compounds from paprika powder was optimized using response surface methodology (RSM). A Box-Behnken Design (BBD), which is one of the most efficient designs of experiment methods, was adopted for the experiment planning. In **Second part,** quantification of total phenolic compounds and flavonoids in paprika powder over the entire storage period, as well as changes in antioxidant and antibacterial activity

The general conclusion and perspectives that this work suggests were presented in the last chapter of this thesis.

Literature review

Chapter I:

General botany, processing and preservation technology of paprika

CHAPTER I – General botany, phytochemistry and biological role of paprika

I.1. General botany

I.1.1. Brief history of spices

Since antiquity spices and herbs have been used throughout the world for flavoring and preserving foods, but also for medicinal and cosmetic purposes (Embuscado 2015).For contemporary nutritionists, the word "aromatics" refers to any substance added to a food or drink to modify its taste or aroma. At the time, the word "épice", which appeared in France around 1150 and is derived from the Latin species, " espèce ", did not yet exist (Pelt 2002) . But from the outset, gold and spices have been associated; throughout history, they have been considered the most precious of goods, they have been one of the driving forces behind commercial expansion (Delaveau 1987); as early as the 15th century, spices were supposed to come from heaven itself, symbolizing wealth, happiness and prestige, material comfort and social markers of taste and elegance. Their high price, due to their rarity and their distant, mysterious origins, reserved them for the elite of medieval society. They were indeed not a commodity like any other, since they were both condiment and medicine, dye and perfume, the race for spices led to the reconnaissance of the African coast, the establishment of the Indian route and the discovery of the Americas (Marty-Dufaut 2015; Balard 2023; Durel 2006).

Each spice has its own originality, its own merits, its own history, its own present. Modern research shows the complexity of flavors, the subtlety of physiological actions and the value of judicious use (Delmas and Reynard 2014).

In Europe during the 16th century. It wasn't until sailors, motivated by research, ventured to a new continent where they found not the spices they were looking for, but other plants that were to revolutionize the cuisine of Europe and the rest of the world. the American Solana, the pimiento/pepper (*Capsicum*), also came to spice up and color a cuisine that must have been dull until then (Katz 1992).

I.1.2. Pimento chili; pepper; paprika

First of all, what are chillies and peppers? Botanically speaking, chillies and peppers are the same plant, *Capsicum* (Moscone et al. 2007). There are several species of this genus, but one of them, *Capsicum annuum* , has many varieties, some of which, known as "peppers", are not hot (Monnier 1994).

I.1.2. 1. Pimiento: Etymology and origin

In 1493, when Columbus's crew returned to Spain, one of his companions they had brought back "a pepper hotter than that of the Caucasus", the same plant they had brought back from the Caucasus, the same plant they had discovered in Mexico under the name of "aji", or chili (P. W. Bosland, Votava, and Votava 2012; Andrews 1995; Bouilly 2016) . The term "pepper" was derived from this, and the name "piment", which appeared at the end of the 17th century, probably comes from the Spanish "pimienta". In Hungary, the term "paprika", which is of Serbo-Croatian origin, it stems from the Greek “peperi” and in the Latin “piper”, both referring to pepper (Klátyik et al. 2018), is only attested in the 18th century (Katz 1992; Monnier 1994; Katz 2019).

spread by the Portuguese to Africa and Asia. In Europe, the chilli pepper was quickly cultivated in gardens, and used as an ornamental and medicinal plant as well as in cooking (Bouilly 2016). It was considered a spice but not yet a vegetable, and was often ground into powder like pepper, or used as a condiment to enhance the flavour of pickles, and was even preserved in sugar (Katz 1992; Halikowski Smith 2015).

I.1.2.2. Origin and classification

All peppers are American (Grubben and El Tahir 2004). There are three main areas of origin (Menichini et al. 2009). In Mexico we found **Capsicum annuum L.** known as "chilli", has solitary white flowers with a delicately curved stalk. This is the most widespread species in the world. It can have very large fruits of various shapes, usually hanging, and also, **Capsicum frutescens L.** called "chiltepin". This is the form closest to the wild. It has numerous greenish

flowers and, unlike the others, a clearly bent peduncle and small, conical, upright fruits. It is a bushy plant that can live for 5 to 6 years. In Andes **Capsicum baccatum L.** pendulum variety (Willd.) Known as "aji", it is grown between 500 and 1,500 metres. It bears large white flowers with golden chevrons. **Capsicum pubescens R.P. "Rocotto"** in Peru grows in Bolivia at altitudes of between 1,50 and 3,000 metres. Very hairy, it is resistant to the cold. It has purple flowers and black, rough seeds. In Andean, Amazon and Caribbean slopes **Capsicum chinense Jacq.** called "ChiIli habanero" in Mexico. It looks small, highly pleated Chinese lanterns. It bears white flowers.

Even today, the classification remains controversial. The majority of authors group peppers into five main species like Pino, Fuentes, and Barrios (2011) say that the Capsicum genus includes 25 wild and 5 domesticated species **(*C. annuum L., C. frutescens L., C. chinense Jacq, C. baccatum Jacq, and C. pubescens L*)** that include more than 200 varieties, which vary widely in size, shape, flavour and sensory heat. These fruits range from non-hot to very hot, with the most popular classification of chillies being: hot peppers and sweet peppers (Zhang, Lu, and Dárcy 2002; DeWitt and Bosland 2009). Sometimes combined into two species, *Capsicum annuum* and *Capsicum frutescens*. The current trend is to consider only one species, *Capsicum annuum* how is the most widely cultivated in temperate and subtropical countries, and the others becoming subspecies and varieties (Greenleaf 1986; Nwachukwu, Mbagwu, and Onyeji 2007). figure1 shows a demonstration of the different capsicum species (Olatunji and Afolayan 2018) the varieties and cultivars of *Capsicum annuum* are classified according to their morphological characteristics and their fruit shapes, the fruit is a berry and may be green, yellow, or red when ripe (Oecd 2006; Zhigila et al. 2014). The species encompass a wide variety of shapes and sizes of peppers, both mild and hot, ranging from bell peppers to chili peppers (Zhang, Lu, and Dárcy 2002).

Figure 1 : Images of the five cultivated *Capsicum* species (a) *Capsicum annuum* (b) *Capsicum frutescens* (c) *Capsicum chinense* (d) *Capsicum baccatum* (e) *Capsicum pubescens* (f) Ground pepper (Olatunji and Afolayan 2018).

I.2. Processing

I.2.1. Production of pepper in words and in Algeria

Worldwide, consumption of Capsicum fruit, probably one of the world's leading spices or food additives, is increasing all the time. spices or food additives, is steadily increasing (Kouassi Kouassi and Koffi-Nevry 2012). In fact, over the last ten years, this culture has consolidated in many countries, growing at an average rate of 7 % year worldwide (Issa 2019).

Global production of Capsicum peppers mainly comprises: the mild to highly pungent chili peppers; bell and sweet peppers utilized as vegetables; and paprika, primarily traded as a spice. The world production of fresh and dry chilies and green bell peppers in 2014 was 32.32 and 3.82 million metric tons (MMT), respectively. China was the leading producer of green types with 16.12 MMT, followed by Mexico, Turkey, Indonesia, and Spain. (Po, Siddiq, and Shahzad 2018).

The 2014 global production of *C. annuum* in the world was approximated to be 409,000 tons, which is anticipated to rise to about 454,000 tons in 2015. Vietnam was on top of the list in

2014 with a yield of 155,000 tons which is about 35% share of global production (Shoaib et al. 2021). Major countries that grow chili are China, India, Pakistan, Korea, Indonesia, Turkey, and Sri Lanka in Asia; Ghana, Nigeria, Tunisia, and Egypt in Africa; Mexico and the United States in North and Central America; Spain, Yugoslavia, Bulgaria, Romania, Italy, and Hungary in Europe; and Peru, Argentina, and Brazil in South America. The global trade in chili accounts for 16% of the world's total spice trade (Figure 2) (Olatunji and Afolayan 2018).

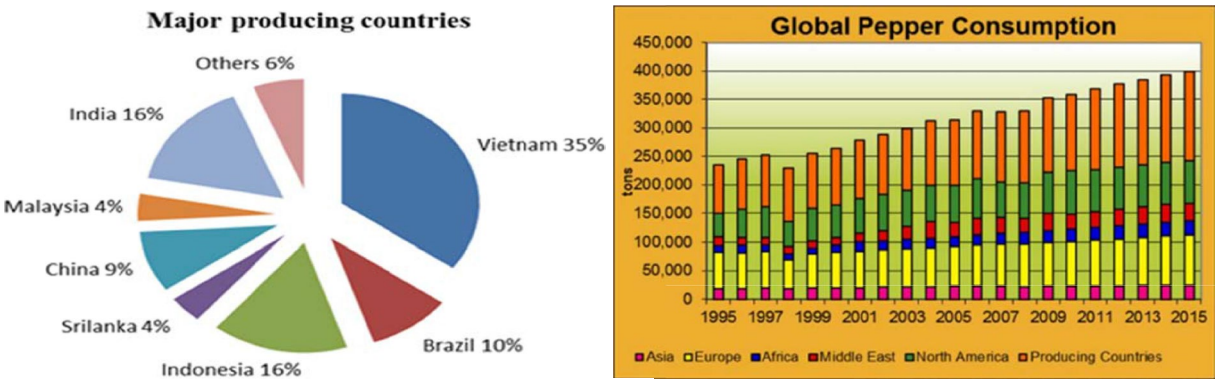

Figure 2: A: Percentage share of major *Capsicum annuum*-producing countries, B: Global pepper consumption (Olatunji and Afolayan 2018).

The global chili consumption is approximately 400,000 tons and has been rising progressively. China and India are the major consuming countries and are reported to consume about 49,000 and 59,000 mt, respectively, in 2014 (Le et al. 2019). Consumption rate for Asia and the Middle East is 3%–4%. Globally, the rate of consumption is 2%–3% per annum. Despite the fact that China is a producer, it is noteworthy that their domestic consumption demand has risen above their production capacity in the last 10 years; therefore, they should be seen as a net consuming country. Sometimes, India has to import pepper to meet up with their local needs. Nevertheless, with conducive weather, India can meet local demand and still have excess to export (Shoaib et al. 2021). the annual world production of paprika and paprika oleoresins was approximately 60,000 tons and 1400 tons, respectively. In recent years, the increasing attention on functional

foods has been also contributed to increase global trade of paprika (Topuz, Feng, and Kushad 2009).

Pepper is an unavoidable component of the Algerian population's diet. It is consumed in various forms including fresh consumption, dry powder and pasta (Bedjaoui et al. 2022). It is grown across the whole country in a different of settings varying from small-scale subsistence farming to large-scale commercial enterprises. In Algeria, 2 400.1 hectares are annually cultivated with chili peppers and total production is estimated at 174 234.1 tons (Mimouni 2023).

I.2.2. Processing of pepper to obtain paprika: **Technology steps of paprika**

Spice paprika is a condiment that consists of dried and ground paprika or chili, The industrialised production of the spice paprika started towards the end of the seventeenth century and grew to become highly developed by the mid-eighteenth century Both "hot" and "sweet" varieties of spice paprika, with practically closely similar cultivation and processing technologies, and similarly strict food safety requirements (Klátyik et al. 2018).

Traditionally, spices and herbs are dehydrated, but they can also be found fresh or frozen. Dehydrating plants is a very old preservation process (Arvy, Gallouin, and Roques 2015). The whole samples are laid out thinly on racks and then dried and are placed:

- In the sun, the temperature is below 35°C;
- A.way from the sun, if the pigments or aromas do not keep well; in driers, the most common drying method today. In all cases, they are turned over regularly until completely dry.

I.2.2. 1. Drying

The freshly picked raw spice paprika contains 16–18% dry matter. High quality ground paprika as raw material needs at least 4 weeks of after-ripening to decrease the rate of water content and increase the rate of dry matter and stable carotenoids (Klátyik et al. 2018) . From the middle of July to mid-September, solar energy can be used for pre-drying in a hygienic equipment, like grids under a shaded and ventilated plastic tunnel this is the method used in Hungary.

Since the middle of the 19th century, plants were dried in a specially equipped room. Air was circulated through a space between the top of the walls and the roof, and was increased by opening the doors in fine weather. The plants were kept in semi-darkness, which protected the color pigments and aromas. In good weather conditions, drying took eight days (Arvy, Gallouin, and Roques 2015).

Other, drying techniques faster and better controlled processes are now used; these include:

- Individual" dryers, in which a stream of hot, pulsating air circulates. The drying process, with an adapted temperature, takes place in 48 hours the quality of the plants is better than during natural drying;

- Prune dryers, in which carts of around ten trays can be loaded. The temperature is then the same for all the samples;

- Continuous ovens with independent settings. Dehydration, appropriate to the sample, is programmed according to a pre-established temperature curve, allowing control of the color of the plant, its organoleptic quality and the speed of dehydration.

Preparing the dried material for grinding, additional (max. 50°C) drying is needed until the dry matter content decrease to 6–8% or less. After gentle grinding, the final result is high quality paprika with excellent color content, outstanding aroma compounds and bioactive components.

I.2. 2.2. Decontamination steps

After drying, spices and are still frequently contaminated with micro-organisms (mould, yeast, bacteria, etc.), It is therefore necessary to decontaminate them after dehydration. to enhance food safety of spice paprika, a decontamination step needs to be carried out to secure the microbial purity of the product and to avoid contamination of food seasoned with it (Klátyik et al. 2018; Schweiggert, Carle, and Schieber 2007). There are various treatments available:

A. Irradiation treatment

Microbial decontamination is most often carried out by ionisation (or irradiation) e.g., gamma irradiation (Farkas 2006) in France allows an average dose of 11 kGy for the irradiation of

spices and dry aromatic substances, while the United States uses a maximum dose of 30 kGy. Scientists at the FAO (Food and Agriculture Organisation of the United Nations) have admitted that doses in excess of 10 kGy do not change the composition of foods, but greatly reduce the microbiological risk to the consumer (Arvy, Gallouin, and Roques 2015; Waje et al. 2008). In order to:

- Reduce the risk of food-borne illness by destroying pathogenic organisms;
- Reduce spoilage of foodstuffs by delaying or stopping decomposition processes and destroying the organisms responsible for these processes;
- Reduce the loss of foodstuffs due to premature ripening, germination or growth,
- Eliminating organisms harmful to plants or plant products from foodstuffs.

B. Steam treatment

Steaming is a decontamination technique of spices of proven and high utility. Due to steam treatment (saturated dry steam, 108–125°C for 20–120 s) the mesophilic aerobic total bacterial were reduced, while yeasts, coliforms, E. coli and Enterobacteriaceae could not be detected. The concentration of the main bioactive compounds, as capsanthin esters, total carotenoids, tocopherols, vitamin C and the color value did not change, however, the total tocopherol content decreased by 6%. The volatile aroma compounds was decreased.

high-temperature steaming is effective against contaminating microorganisms, it can decrease the volatile oil content, cause colour degradation and may increase the moisture content of dried paprika product, which then reduces shelf-life (Demirci and Ngadi 2012). Furthermore, steaming is not suitable for spore inactivation. These results confirm that steaming provides a good possibility to reduce the microbial load, without drastically changing the content of bioactive compounds.

C. Microwave and enhanced microwave heating

Microwave heating is alternative method advocated for effective reduction of the level of mesophilic bacteria, 30 s in dry and wet treatment was found to allow the highest reduction of the bacterial level in chilli among different spices studied (Shankarrao Shirkole et al. 2021). However, this method adversely affected the color of the paprika processed, giving it a darker, brownish appearance. To avoid the detrimental effect of the treatment, a modified microwave treatment (including re-wetting of the sample, intensive mixing during the entire treatment and post-drying to the initial moisture level), thus, enhanced microwave treatment allows a reduction of microbial contamination (principally for moulds and coliforms) without a decrease in the levels of bioactive compounds. The temperature did not significantly affect chemical composition, but had a significant effect on sample color. all samples became browner and darker, and as color changes did not correlate with the observed levels in carotenoids and the ASTA value, it has been concluded that color changes due to the treatment are likely to be related to plant carbohydrates and proteins (Klátyik et al. 2018).

D. Other treatments

Oregano essential oil was attempted as a natural anti-microbial agent to reduce microbial count in paprika (Casas et al. 2016). Although it was not found to be of adequate activity by itself to allow sufficient inactivation of microbial spores in paprika, when used in combination with high pressure carbon dioxide, microbial inactivation largely increased (by 99.5%).

In a number of food products, high hydrostatic pressures increase shelf-life and maintain nutritional and organoleptic properties better, the effect of high hydrostatic pressures and pasteurization (in a water bath at 70°C for 10 min) was examined on the levels of given bioactive components and on the texture of spice paprika (Po, Siddiq, and Shahzad 2018). Pasteurization treatment at high hydrostatic pressure (500 MPa) had less influence on the bioactive component content and on the texture, than at low pressure.

Chemical treatment with ethylene oxide is also a worldwide available decontamination technology, but the potential use is limited by its toxicity. Due to its carcinogenic potential to humans, the use of ethylene oxide is forbidden to be used in food processing in the EU (Al-Mamun et al. 2016).

I.2.3. Preparing paprika on an artisanal scale in Algeria

Figure 2 shows the main stages in the traditional paprika powder,

I.2.3.1. Harvest

When they reach their final size, chili fruits turn red. The ripe fruits are harvested by hand from the end of August to the end of September. It is carried out in several stages as the fruit ripens and reaches maximum color intensity (Abdelmajid 2016). This high natural pigment content and low water content. Partial dehydration of the fruit on the plant is recommended as the color stability in paprika is better when harvested late.

I.2.3.2. Post ripening

The harvested pods are stored in a well-ventilated place in heaps to undergo post-ripening for a period of 3 days. Post-ripening of the fruit contributes to the development of other intrinsic properties that determine the quality of the paprika (Díaz-Pérez, Muy-Rangel, and Mascorro 2006). During post-ripening, as well as the drying out of the fruit (loss of water), the sugar content is reduced, but always in such a way that the residues guarantee the characteristic flavor of the fruit. At the same time, in addition to the increase in red pigments, the total pigment content pigment content increases by 30 to 50% (Zaki et al. 2018).

The chilli pepper intended for drying is cut into pieces of a suitable diameter in order to avoid any kind of colour change when it comes into contact with hot air, which would subsequently affect the colour of the finished paprika product and the stalk is removed in the bigining

I.2.3.3. Drying

Paprika is dried using the traditional method of open air drying due to its low cost. The capsicum fruits are spread out on the ground, left in the sun for exposed to the sun for 5-7 days. At the end of the drying process the water content of the paprika pods falls from around 80%
to less than 10%. The shell obtained by drying in the sun is soft to the touch and slightly elastic (Kevrešan et al. 2013). The drying process can take up to 15 days depending on hours of sunshine and weather conditions. weather conditions.

Around 25-35 kg of paprika powder can be produced from 100 kg of fresh fruit (Mínguez-Mosquera, Pérez-Gálvez, and Garrido-Fernández 2000). The paprika obtained by drying in the sun has bright red characteristics, unlike those obtained others obtained in hot air dryers (Mokhtar et al. 2015).

I.2.3.4. Milling

At the customer's request, or the dried chopped and dried are ground in small-scale processing units to obtain particles of less than 0.5 mm (Hakmaoui, Ouatmane, and Fernández-Trujillo 2011). In other paprika-producing countries, the stalk is removed and the rest is ground. the stalk is removed at the start of grinding and the addition of the addition of seeds is controlled, since their percentage of seeds in the fruit varies from 10 to 40%. The dried pods are ground in finely-adjusted horizontal millstones to obtain paprika powder. The importance of grinding in millstones lies in the fact that the paprika heats up as it passes through the millstones heats up, and this heating, combined with the grinding process causes the oil contained in the seeds to be extracted and dissolve the fat-soluble pigments in the pulp, which then particles of paprika, giving them a uniform color. Give them a uniform color (Klieber and Bagnato 1999).

I.2.3.5. Storage and preservation

After grinding, the paprika powder is spread out overnight in a cool room where it could absorb the appropriate amount of water. The current technique used to package paprika is based on this traditional processing method traditional processing method used by farmers. At the end of the

milling process, the product must pass through a sieve with an opening of ≤ 0.5 mm and be packaged using methods and materials that meet food hygiene requirements. Depending on the customer's request, either this powder is delivered as it is after grinding or oil is added to it (Ladrón de Guevara et al. 2002). of the seeds This oil addition operation reduces its oxidation and this by reducing the surface area exposed to air. However, previous studies have shown that quantities of oil between 2% and 6% are sufficient to guarantee better stabilisation of paprika colours. Complete removal of the seeds would reduce paprika's antioxidant capacity by eliminating the tocopherols in the seeds, leaving only those in the shell (Addala et al. 2015). leaving only those in the shell (Klieber and Bagnato 1999) and ascorbic acid, lecithin and cephalin. The latter compounds exert synergistic effects with other antioxidants in terms of their ability to sequester metals. They also maintain colour stability (Park and Kim 2007). The addition of seeds to red pepper flesh prior to grinding and storage in a nitrogen atmosphere also slowed the rate of oxidation of carotenoids in paprika (Topuz, Feng, and Kushad 2009). The final product obtained can vary in colour from yellow to red, depending on the customer's requirements (Pérez-Gálvez, Hornero-Méndez, and Mínguez-Mosquera 2009).

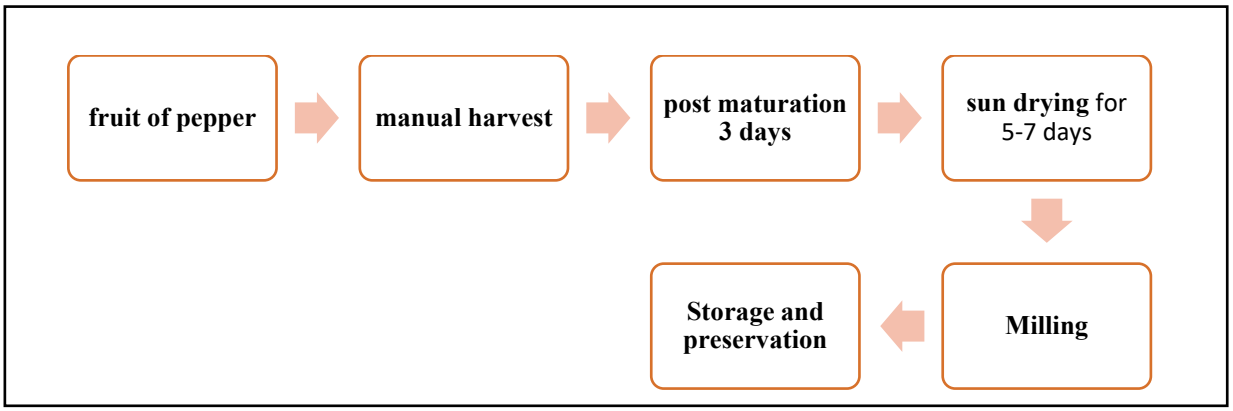

Figure 3: Traditional diagram for preparing of paprika powder (Zaki et al. 2018)

I.3. preservation technology

you can't talk about conservation without talking about water; Water plays many roles in food quality and stability. Water interacts with food ingredients in different ways. As a solvent for

reactants, a plastify for the reaction medium and a reactant, water is involved in the reactions that take place during food processing and preservation (Le Meste et al. 2001).

It is well known that the development of microorganisms responsible for food spoilage depends on the thermodynamic properties of water (water activity, osmotic pressure). The mechanisms that control the speed of chemical reactions or physical transformations in concentrated or even solid media are complex, and prediction models are rare and sometimes controversial. The concept of glass transition, which is increasingly used in food technology, means knowing the physical state as a function of temperature and hydration conditions, in order to control food processing and preservation operations (Colas et al. 2010; Schuck et al. 2008).

What guides the conservation of our product over a more or less long period is its composition protein, fat, carbohydrate and water content. The pH, the organic acids present and also whether the product contains natural anti-microbial agents. There are many ways in which food can deteriorate, including chemical and microbiological contamination, and contamination by insects, rodents and birds (Doyon 1990).

I.3.1. The main food preservation methods

Food safety associate's innovation and sustainability by being the primary theme of the food production and preservation industry. During food production, transport, storage, and final consumption, the food properties may be affected in several ways. To ensure safety and stability, without nutritional and sensory losses, suitable, effective and economic food preservation methods should be selected (Tavman et al. 2019).

The preservation process will restrain the development of microbial such as bacteria and fungi (Seetaramaiah et al. 2011) and oxydation ; aims of food preservations include:

- Maintaining food taste, texture, flavor, quality and nutritional value,
- Reduce the wastage of excess food,
- Maintain a products accessibility for a longer time, even in places it is not being produced,

- Preserve the food materials during transportation,
- Ease the handling of food materials (Devi et al. 2015)

There are several conventional and emerging preservation techniques such as pasteurization, sterilization, cooling, freezing, ohmic heating, microwave and radio frequency, which are thermal preservation technologies.

I.3.1.1. Thermal or heating processing

Is one of the methods used to preserve fruits. Thermal processing is the technology that is applied to prolong the shelf life and increase the longevity of fruits and vegetables (Rosa 2006) This is because the process is productively decreasing the growth of microbial population and pulverizes the natural enzymes (Barrett and Lloyd 2012) Thermal processing is the process of either heating unsterile foods in containers such as canning, or heating food products, prior to packaging or before they are put under sterile states such as aseptic handling.6Besides that, this method also used in the process of milk pasteurization.

I.3.1.2. Drying

Drying is another food preservation technique that has long been utilized. Food products can be dried using various techniques, such as drying under the sun (natural drying) or by using simulated heat under controlled temperature, which using the specially developed chambers called dehydrators or dryers (Devi et al. 2015). While this technique is commonly used for meat and fish, it can also be applied to fruits and vegetables. As a result, dried fruit and vegetable products will be lighter in weight, hence, the delivery cost of dried products can be reduced (Rosa 2006). The moisture content of the food is reduced to 10-15%, thus, the present microorganisms can be inhibited and become inactive (James and Kuipers 2003). The moisture content can be evaporated by using either sun drying or under controlled temperature.4 However, further dehydration is not recommended because regularly, the product will regularly be too brittle. Meanwhile, to guarantee that the fruits will not be damaged during the drying

process, they should be kept in a free-moisture environment. On the other hand, many studies have claimed that drying will degrade the quality of the products (James and Kuipers 2003)

Drying process can be divided into two types, natural drying and artificial drying. Natural drying is drying in the open surrounding and using economical procedure. It does not need any energy consumption, it only uses of sunlight and wind. Fruits will be dried in thin layers and turned routinely under sunlight. In the meantime, there are few methods for artificial drying, which are heated with fuel, with bush dryer, air dryers with artificial ventilation and others. Heating with fuel is the technique usually used in wet climates or when there are an abundant of fruits need to be processed. The bush dryer is the fire in an oven that is produced using oil drums to heat the surrounding air. The heated air will ascend through the thin layer of fruits that is placed on the racks and should be shaken or blended at general interims with regular monitoring. The last method is using the air dryer with an artificial ventilation technique. A motor-controlled ventilator can be utilized to blow warm air from the motor or warming the air using a burner through the products (James and Kuipers 2003).

I.3.1.3.Ohmic heating

Ohmic heating, also known as Joule heating, electrical resistance heating, and direct electrical resistance heating, is a process of heating the food by passing electric current, it has immense potential for achieving rapid and uniform heating in foods, providing microbiologically safe and high quality foods (Varghese et al. 2014). In ohmic heating the energy is dissipated directly into the food. Electrical conductivity is a key parameter in the design of an effective ohmic heater. Beyond heating, applied electric field under ohmic heating causes electroporation of cell membranes, which increase extraction rates, and reduce gelatinization temperature and enthalpy. Ohmic heating results in faster heating of food along with maintenance of color and nutritional value of food. Water absorption index, water solubility index, thermal properties, and pasting properties are altered with the application of ohmic heating (Kaur and Singh 2016). Ohmic heating results in pre-gelatinized starches, which reduce energy requirement during

processing. But its higher initial cost, lack of its applications in foods containing fats and oils, and less awareness limit its use (Knirsch et al. 2010).

I.3.1.4. Microwave and radio frequency

This paper brings to perspective issues related to research initiatives for the application of microwave (MW) and radiofrequency (RF) applications in foods. Both MW (300 MHz and 300 GHz) and RF waves (3 kHz — 300 MHz) are part of the electromagnetic spectrum that result in heating of dielectric materials by induced molecular vibration as a result of dipole rotation or ionic polarization (Ramaswamy and Tang 2008). They have been credited with volumetric heat generation resulting in rapid heating of foodstuffs. Due to their lower frequency levels, RF waves have a larger penetration depth than MW and hence could find better application in larger size foods (Datta and Davidson 2000). Besides the popular domestic use of MW ovens, commercialized applications of MW/RF heating include blanching, tempering, pasteurization, sterilization, drying, rapid extraction, enhanced reaction kinetics, selective heating, disinfestations, etc. This paper reviews the current status and research needs for in-packaged sterilization technologies for commercial applications (Brodie, Jacob, and Farrell 2016). Technological challenges include process equipment design, microbial destruction and enzyme inactivation kinetics, temperature and process monitoring, and achieving of temperature uniformity. Other issues also relate to the use of packaging material in in-package sterilization applications, package/container concerns in domestic MW ovens, receptor technology for creating dry-oven conditions, modeling and time-temperature process integrators. There is also the issue of non-thermal and enhanced thermal effects of microwave heating on destruction kinetics (Willert-Porada 2006).

Ultrasou*nd Ultrasound treatment involves use of high intensity and frequency sound waves which are passed into food materials. The efficient technology is chosen due to its simplicity in the equipment usage and being low cost as compared to other advanced instruments. The versatility of ultrasound is shown in its application in different fields ranging from medicine,

healthcare to food industry (Dai and Mumper 2010). Figure 2 illustrates a representation of different types of sonicators used for powdered and liquid foods. The process deals with ultrasonic radiation passing through the target solution. This action causes a disturbance in the solid particles in the solution leading to particles breaking and diffusing into the solvent (Cares et al. 2010). It should be noted that the intensity of the technique should be kept constant. This is because as intensity increases, intramolecular forces break the particle–particle bonding resulting in solvent penetrating between the molecules, a phenomenon termed as cavitation (Fu et al. 2020; Khan et al. 2020). Further enhancement of ultrasound extraction is dependent on factors like improved penetration, cell disruption, better swelling capacity and enhanced capillary effect (Huang et al 2020; Xu et al 2007). Table 3 shows the

I.3.1.5. Freezing

Freezing is one of the easiest and most economical way to preserve foods. Most products contain enzymes that can destroy the nutrients in the food. In this light, this enzyme will change the appearance and texture of the product during storage. Freezing will stop the development of microbial however, will not kill them. Freezing is being categorized as a form of preservation because it can reduce the water level activities which will inhibit the microbial activity and decrease the chemical reaction rates. As a result, thermal exposure in frozen foods is low. At the same time during freezing and defrosting process the tissue structure of the food will damage and it is occurring due to the rate and temperature applied. This will lead to the deterioration of texture, colour and the taste (Ingham 2008). Freezing food loss more nutrients during storage due to the oxidation process occurred (Barrett and Lloyd 2012). Even though it is the easiest method of preservation, but the process occurred is complex due to the involvement of heat transfer and also the changes in chemical and physical series that affect the food products‟ quality (Gambuteanu, Borda, and Alexe, n.d.) Research done by Kim et. al.(Kim et al. 2015) on the use of different types of freezing techniques, followed by using different

types of thawing for pork quality in ready to eat meals. The results show that of time and rates of freezing is the factors that affect the quality of meat.

I.3.2. Refrigeration technologies used for food preservation

Refrigeration has become an essential part of the food chain. It is used in all stages of the value chain, from farm, food processing, to distribution, retail and final consumption at home. The food industry employs both chilling and freezing processes where the food is cooled from ambient to temperatures above 0°C in chilling and between -18°C and -35°C in freezing to slow the physical, microbiological and chemical activities that may cause deterioration in foods. The use of refrigeration for food production and its preservation is undoubtedly the most extensive technique (Popa, Miteluț, and Popa 2019).

Cooling and freezing of products have been extensively applied for preservation of leafy vegetables, spices and milk products to maintain the sensorial attributes and nutrition qualities. Extensively used freezing techniques involve air blast, cryogenic, direct contact and immersion freezing, while advanced techniques involve high pressure freezing, ultrasound assisted freezing, electromagnetic disturbance freezing and dehydration freezing (Cheng et al. 2017; Barbosa de Lima et al. 2020). Cooling and freezing process mainly relies on the process of heat transfer. During cooling, there is a transfer of heat energy from the food and packaged container to the surrounding environment leading to an agreement of cooling. Thus, thermal conductivity and thermal diffusivity greatly affect the cooling or freezing rate. During the recent years, the storage technique has gained significant interest with the start of ready-to-eat foods catering to the needs of the consumer. The foods with their appropriate packaging material and cool temperature will always inhibit entry of microorganisms as well as maintain food safety. Although cooling and freezing are effective in their own terms, cooling time, uneven speed of ice crystal formation, storage expenses and specialized environments are concerning issues. In order to understand and overcome these challenges, technological tools like three-dimensional mathematical models and computational fluid dynamics models were evaluated to understand

the heat transfer and fluid flow patterns with various food formulations thus showing an approach to minimize the issue (Barbosa de Lima et al. 2020;Stebel et al. 2020). Table 1 shows a description of the various advanced freezing techniques applied to different foods.

Table 1. Advanced freezing techniques widely applied for different foods (Sridhar et al. 2021)

Advanced freezing techniques	Technology involved	Application in foods		References
		Sample	**Conclusions**	
High-pressure freezing	Involves freezing water at high pressure below 0 °C so that it forms small ice crystals instantly once the pressure is released Process takes place with the absence of heat	Comparison of sugar-rich dairy-based food foams (ice creams) and a non-aerated liquid system Maximum pressure applied: 360 MPa at −25 °C	Volume fraction of the air after treatment—78% Crystal size reduction—40 µm to 34 µm Overall improvements in sensorial properties	(Volkert et al. 2012)
	Crystallization occurs instantly once high pressure is released Preservation of original properties and quality improvements noticed	Kombu seaweed *(Laminaria ochroleuca)* Process conditions: 5 °C, 400–600 MPa, 5 min followed by refrigeration at 5 °C or freezing at -24 °C	Comparison of salted and unsalted seaweed Detection of 103 volatile compounds found. Major compounds detected were aldehydes, alcohols, ketones, alkanes, alkenes, and acids Freezing lowered levels of hydrocarbons, alkanes and thiazoles Salting increased levels of acids, alcohols, pyranones, lactones and thiazoles	(López-Pérez et al. 2020).
Ultrasound-assisted freezing	Involves passing of sound waves in between the food. Can be of low frequency	Cantaloupe melon juice *(Microcystis aeruginosa)*	Testing for probiotic substrate *Lactobacillus casei* Study done for a period of 42 days at 4 °C	(Zendebood i et al. 2020)

	(< 100 kHz) or high frequency (20–100 kHz) No destruction of food Intensity, frequency of ultrasound, position of samples, cooling medium temperature key parameters for the process Can be used to treat both solid and liquid samples		Reduced caloric value observed	
		Grape juice Amplitude of 50% and 70% with treatment times of 0, 2.5 and 5 min Temperature maintenance: 50–80 °C	Comparison of ultrasound and pasteurization treatment was done Total phenolic content (TPC) was same for both the treatments at 10 min with amplitude of 70% pH decreased and total soluble solids increased with amplitude and treatment time Results indicated usefulness of juice sonication to enhance inactivation of pathogens	(Margean et al. 2020).
		Pomegranate juice	Results showed ultraviolet 5.1 W/cm^2 dosage, 3.5 L/min flow rate and 50 °C microbes were below the detection limits Lower temperatures could reduce the microbial activity preserving the bioactive compounds	(Khan et al. 2022; Alabdali et al. 2020).
Radioactive freezing	Not predominantly used in freezing Radio waves generate a turning force in the water molecule, and an ice cluster is created due to dielectric and dipolar properties of water	Onion, potato, ginger, carrot Dosage: 0.05–0.15 kGy	Inhibition of sprouting Shelf-life enhancement	(Prakash 2016).
		Cereals, fruits Dosage: 0.15–0.5 kGy	Phytosanitation Sterilization purposes Mycotoxin decontamination observed most effect with advantages in nutrient qualities	(R. Ravindran and Jaiswal 2019; Khaneghah, Moosavi, and Oliveira 2020)

Dehydration freezing or osmodehydrofreezing	Involves osmotic dehydration and freezing techniques Food is first dehydrated (water removal) and immediately frozen Shelf life extension observed due to accelerated freezing process Low energy consumption, low cost of packaging	Mango *(Unripe vs Ripe "Kent" mangoes)* Treatment: 50 °C in 60 brix sugar solution with 2 g calcium lactate/100 g with pectin methyl esterase	Unripe mangoes showed two- to fivefold soluble solid gain as compared to ripe Unripe samples had lowest water loss with reduction in lightness. Ripe samples were stable Pectin methyl esterase improved rigidity in mangoes	(Sulistyawati et al. 2018).
		Pineapple with sucrose syrup Treatment: 2 h at 40 °C	Changes in pH, total acidity, soluble solids, and water observed Dry matter content increase during multiple stage osmodehydrofreezingStudy conducted showed multistage osmodehydrofreezing gave better performance than single stage osmodehydrofreezing	(Fernández, Lovera, and Ramallo 2020).

Chapter II

phytochemistry and biological role of paprika

Chapter II- Phytochemistry and biological role of paprika

II. Micronutrients in paprika

The universal consumption of paprika, known for their high nutritional content (which includes a good range of vitamins, minerals, phytochemicals, and dietary fiber), may play a role in decreasing human micronutrient deficiencies (Molnár, Kónya, et al. 2018).

Micronutrients, which include minerals, vitamins, antioxidants, phytochemicals, and trace elements, are indispensable and are needed in smaller amounts and this is why they are known as "micro" nutrients (Loizzo et al. 2017). Paprika have been recognized to be good sources of minerals, provitamin A, vitamins C and E, carotenoids, and phenolic compounds, metabolites with renowned antioxidant properties which influence human health positively (Molnár, Kónya, et al. 2018).

Paprika is a rich source of capsaicinoids, carotenoids (with some of them having provitamin A activity), flavonoids, tocopherols (vitamin E), and ascorbic acid (vitamin C). The consumption of Paprika is rising, and this may represent an essential source of vitamins for the world populace. Several antioxidants, vitamins C, E, and provitamin A, are sufficiently available in high concentrations in paprika. Paprika apart from carotenoids are also good sources of xanthophylls and may contain high amounts of vitamins B, (riboflavin) B, (thiamine), B3 (niacin), and P (citrin), and are richer sources of vitamins A and C than the regularly recommended food sources (Molnár et al. 2018) in Table 2 shows the micronutrient composition of paprika.

II. 1. Major vitamins in Paprika

II.1.1. Vitamin A

vitamin A is not found in paprika, high levels of the provitamins A including α-carotene, β-carotene, and β-cryptoxanthin, which in the human liver can all be transformed into vitamin A, do occur. The most abundant form of provitamin A is α-carotene, which cleaves to form two

molecules of retinol, which is the physiologically active form of vitamin A (Wahyuni et al. 2013).

II.1.2. Vitamin C

Vitamin C also referred to as ascorbic acid, a water-soluble vitamin is an important antioxidant and a cofactor for enzymes that partake in metabolism of human.

Paprika provide high contents of vitamin C. pepper fruit can contain six times as much vitamin C as an orange. In paprika, ascorbic acid level attributed to intensity of light and high glucose levels which is the ascorbic acid precursor. the amount of vitamin C in paprika depend on maturation of pepper fruit as well as variations in environmental growing conditions and genetic background (Molnár et al. 2018).

Since the activity of water in a sample of paprika depends on the manufacturing procedure used, it is clear that the drying system, including its preprocessing steps, will have an influence on pigment stability. Pigment degradation in paprika coincides with the destruction of vitamins C and E and continues with degradation of carotenoids, due to their oxidation in air. At the same time, oxidation is influenced by external factors which may be a physical (temperature, humidity, light, etc.,) or chemical nature (presence of metallic ions, enzymes, peroxides etc.,) (Ramesh et al. 2001).

Paprika differed in their response, in terms of stability, to drying conditions. Natural drying was favorable for the highest carotenoid retention, but not for the retention of vitamin C. It was surprising that drying at 50 °C for 24 h resulted in a paprika with vitamin C content 1.5–3 times higher than that found in paprikas produced with other drying methods (Daood et al. 2014).

II.1.3. Vitamin E (Tocopherols)

The major component of vitamin E was α-tocopherol (Gnayfeed et al. 2001). There are several forms of tocopherols which includes the α-, β-, γ-, and δ-forms, and their occurrence is based on the number and position of methyl groups on the aromatic ring. The different tocopherol compound contributes relatively to the total vitamin E activity. They also reported that α-

tocopherol has the utmost vitamin E activity. Paprika spice is the best sources of natural vitamin E, her α-tocopherol level is similar to those of spinach and asparagus and fourfold higher than that of tomatoes. The recommended daily intake of vitamin E is 15 mg/day of α-tocopherol of both women and men (Loizzo et al. 2017). At present, its only known function is as an antioxidant(Birlouez et al. 2002).

II.2. Minerals contents in paprika

Paprika is rich in the following minerals, which are : potassium>phosphorus>magnesium (Ryu et al. 2021). The content of metals in paprika samples is essential for food safety and quality; in addition, it provides elemental fingerprints about the region where the pepper was produced. It has been demonstrated by principal component analysis (PCA) that the samples containing the highest amounts of K, Mg, Mo and As were among hot paprika samples, while the lowest amounts of Mg, B and As and the highest amounts of Cu were mostly found in sweet paprika (Poór et al. 2018). The metal content in paprika can be influenced by different factors, such as the concentrations of these elements in the soil, fertilizing practices and processing conditions(Palacios-Morillo et al. 2014). For this reason, the characterization and determination of the elemental content in paprika are important in plant biology and food science.

Table 2: composition of paprika in micronutrients (vitamins and minerals) in g per 100 g of paprika (Muduli et al. 2012)

Paprika: vitamines pour 100 g

Water-soluble vitamins		% RDA[1]
Vitamine B1 - thiamine	0,33 mg	30 %
Vitamine B2 - riboflavine	1,23 mg	87,86 %
Vitamine B3 - niacine, ex- vitamine PP	10,1 mg	63,13 %
Vitamine B5 - acide pantothénique	2,51 mg	41,83 %
Vitamine B6 - pyridoxine	2,14 mg	179,29 %
Vitamine B9 - folates, acide folique	49 µg	24,5 %
Vitamine B12 - cobalamines	0 µg	0 %
Vitamine C - acide ascorbique	0,9 mg	1,13 %
Fat-soluble vitamins		% RDA[1]
Vitamine A - activité vitaminique A[2]	0 µg	0 %
soit	0 UI	-
Rétinol	0 µg	-
Bêta-carotène	-	-
Vitamine D - ergocalciférol, cholécalciférol	0 µg	0 %
soit	0 UI	-
Vitamine E - tocophérols, tocotriénols	29,1 mg	242,5 %
Vitamine K	80,3 µg	107,07 %
Vitamine K1 - phylloquinone, phytoménadione	80,3 µg	-
Vitamine K2 - ménaquinone	-	-

1: Recommended Daily Allowances (RDA) are defined by European Union regulation 1169/2011. As with the other data presented on this page, the percentage of recommended daily allowance (% RDA) corresponds to a 100-gram portion of the food "Paprika".

2: Vitamin A activity corresponds to the sum of retinol and precursors in retinol equivalent. The coefficient used for beta-carotene is 1/12.

Paprika: principaux minéraux pour 100 g		
Minérals		% RDA[1]
Calcium - Ca	229 mg	28,63 %
Magnésium - Mg	178 mg	47,47 %
Phosphore - P	314 mg	44,86 %
Potassium - K	2280 mg	114 %
Sodium - Na	68 mg	-
Trace elements		% RDA[1]
Chlore - Cl	-	-
Cuivre - Cu	0,71 mg	71 %
Fer - Fe	21,1 mg	150,71 %
Iode - I	-	-
Manganèse - Mn	1,59 mg	79,5 %
Sélénium - Se	-	-
Zinc - Zn	4,33 mg	43,3 %

II.3. Phytomicronutrients

Phytomicronutrients belong to families of molecules with very diverse structures, such as terpenoids (including terpenes, triterpenes, phytosterols, saponins and carotenoids), polyphenols (including flavonoids), organosulphur or nitrogen compounds, iridoids and others (co-enzyme QlO, betacyanins, chJorophyJles) (IFN et al. 2012). All these substances are derived from the secondary metabolism of plants. Phenolic compounds are known as metabolites that accumulate in plants in response to stress. In some cases, they are known as phytoalexins (El Gharras 2009).

The quantities of these molecules vary depending on the variety and the physiological stage of the plant, the climate (light, temperature), cultivation practices (fertilisation, irrigation), post-harvest conservation and storage conditions, and culinary practices (fresh, cooked, dried, etc.) (Rasouli, Farzaei, and Khodarahmi 2017).

These substances are not considered to be essential for humans, and their recommended dietary intakes have not yet been established. However, numerous biological activities have been reported in the literature, in nutritional doses and more frequently in pharmacological doses (Williamson 2017).

II.3.1. Phytomicronutrients in paprika

II.3.1.1. Terpenoid compounds (Carotenoids)

Carotenoids are biosynthesized from isopentenyl diphosphate, with the chain gradually lengthening. The precursors, such as phytoene, are colorless compounds, but with the introduction of double bonds, colored compounds appear (carotenoids often have more than 10) (Mackinney 2012).

Lycopene, which is a metabolic intermediate, has a linear form. Cyclisation of its extremities transforms it into B-carotene, which is the precursor of vitamin A. The oxidation of B-carotene in turn produces xanthophylls. When the hydroxyl group of xanthophylls is conjugated with fatty acids, esters are formed, such as capsanthin, lutein and B-cryptoxanthin (IFN et al. 2012). Carotenoids are present in the chromoplasts, organised in crystalloids, and in the chloroplasts, they are linked to proteins. Carotenoids are lipophilic pigments that protect against UV rays. Nutritionists classify carotenoids as provilamin A (including B-carotene, a-carotene, 9-cis-B-carotene and B-cryptoxanthin) and non-provitamin A (including lycopene, lutein and zeaxanthin). Carotenoids range in colour from yellow to red. B-cryptoxanthin and B-carotene are orange carotenoids. The former is present in many fruits and vegetables, including apricots and carrots. Lycopene and capsanthin are red in colour. Lycopene is more specific to tomatoes. Capsanthin is found in peppers or paprika. It should be noted that the extraction of carotenoids from different plants is the source of food colourings (Sajilata, Singhal, and Kamat 2008).

Paprika powder is a good source of different carotenoids, which be used as natural food colorants (Rodríguez-Burruezo, González-Mas, and Nuez 2010). The main carotenoids

identified in paprika powder are beta-carotene, capsorubin, capsanthin, lutein, zeaxanthin, and violaxanthin (Ponder, Kulik, and Hallmann 2021). It can be used in food as spices, colorants, and antioxidant agents. carotenoid content are shown in tables 1 and 2 (Ponder, Kulik, and Hallmann 2021)

Drying methods, storage temperatures, and packaging materials of paprika affected color values, the loss of red color was closely related to the reduction of moisture content during storage (Park and Kim 2007; Addala et al. 2015) also according to (Addala et al. 2015) paprika stored at high temperature - humidity storage degraded significantly rapidly compared to paprika stored in room conditions. Refrigerated and freezer stored samples showed minimal extractable and visual color +loss. ,

Table 3 : The content of total carotenoids and individual identified carotenes in hot and sw eet paprika (in mg 100 g DW) (Ponder, Kulik, and Hallmann 2021)

Product	**Total Carotenoids**	**beta-Carotene**	**cis-beta-Carotene**	**alpha-Carotene**	**Capsorubin**
Sweet Paprika	52.4 5± 0.27	4.24 ± 0.09	1.54 ± 0.02	22.34 ± 0.25	2.30 ± 0.02
Hot Paprika	66.63 ± 1.11	5.23 ± 0.10	4.15 ± 0.0	29.87 ± 1.01	3.12 ± 0.23

Table 4 :The content of xanthophylles in hot and swet paprika (in mg 100 g DW) (Ponder, Kulik, and Hallmann 2021)

.**Product**	**Cryptoxanthin**	**Cryptoflavin**	**beta-Cryptoflavin**	**beta-Cryptoxanthin**	**Lutein**	**Zeaxanthin**	**cis-Zeaxanthin**
Sweet Paprika	14.88 ± 0.08	1.08 ± 0.03	0.30 ± 0.01	0.23 ± 0.00	3.12 ± 0.11	0.45 ± 0.01	1.98 ± 0.01
Hot Paprika	17.07 ± 0.23	0.92 ± 0.03	0.44 ± 0.01	0.28 ± 0.00	2.93 ± 0.06	0.54 ±0.00	2.10 ± 0.01

II.3.1.2. Phenolic compounds

Phenolic compounds are molecules containing a phenol group (aromatic nucleus with a hydroxyl group). Polyphenols therefore have several phenol nuclei and several hydroxyl

groups. The functions of phenolic compounds are numerous and very important for plants: defence (phyto-alexins), food deterrence against parasites (tannins), attraction of pollinators (flavonols), protection against UV radiation, participation in the color (anthocyanins, flavonols), aromas and fragrances of plants, structural role (lignin). Man has exploited the properties of phenolic compounds, for example in the food industry (coloring agents with anthocyanins). Phenolic compounds are classified into two main groups: non-flavonoids and flavonoids (IFN et al. 2012).

Non-flavonoid phenolic compounds are molecules with C6-C1 or C6-C3 units. The leaders are gallic acid and caffeic acid respectively. Caffeic acid can link up with other molecules to form, for example, caftaric acid, found in grapes, or chorogenic acid, found in a large number of fruits (apples, pears, etc.) (El Gharras 2009).

The second major group is made up of C6-C3-C6 **flavonoids**. Flavonoids differ according to the degree of oxidation of the heterocycle and may be more specific to certain fruits and vegetables, such as the flavones in peppers. In paprika the most important phenolic compounds are flavonoids and phenolic acids (Ponder, Kulik, and Hallmann 2021).

paprika powder was characterized by a higher content of capsaicin; According to a study on polyphenols, they have identified the different molecules and their content that are found in paprika they are represented in the table 3 and 4 .

Table 5: The content of total polyphenols, total phenolic acids, capsaicin, and individual identified phenolic acids in hot and sweet paprika (in mg 100 g DW) (Ponder, Kulik, and Hallmann 2021).

Product	Capsaicin	Total polyphenols	Total Phenolic Acids	Gallic	Chlorogenic	Caffeic	p-Coumaric	Ferulic
Sweet Paprika	242.67 ± 8.58	226.44 ± 6.51	198.29 ± 6.38	102.58 ± 7.24	66.98 ± 4.45	11.16 ± 1.31	14.59 ± 0.88	2.98 ± 0.23

Hot Paprika	731.82 ± 15.96	275.50 ± 5.45	243.49 ± 6.99	85.03 ± 4.59	58.44 ± 1.70	12.09 ± 0.03	73.99 ± 6.26	13.95 ± 0.37

Table 6: The content of flavonoids and individual identified flavonoids in in hot and sweet paprika (in mg 100 g DW) (Ponder, Kulik, and Hallmann 2021).

Product	*Total Flavonoids*	*Quercetin-3-o-Rutinoside*	*Myricetin*	*Quercetin*	*Quercetin-3-o-Glucoside*	*Kaempferol*
Sweet Paprika	28.14 ± 0.30	5.95 ± 0.63	4.11 ± 0.09	2.73 ± 0.26	13.49 ± 0.60	1.87 ± 0.01
Hot Paprika	32.00 ± 1.55	5.08 ± 0.22	3.68 ± 0.12	2.52 ± 0.12	19.05 ± 1.38	1.67 ± 0.01

II.3.2. Principle Phyto-micronutrient variation factor

Plants vary widely in their composition and phytonutrient content. This diversity depends on the specific characteristics of each species, as well as variations linked to a number of factors. These variations may be due to biological, physiological, genetic, agronomic or environmental factors, or to the way in which they are preserved and processed (Amiot-Carlin 2008).

Carotenoids, and vitamin C, seem to undergo oxidation in these processes, particularly in dried products. The lycopene content of dried tomatoes is reduced by 10% when manufactured at 110°C, but it is not stable in storage. However, there are marked differences in the stability of polyphenols, from the most stable to the most volatile, for example bydroxycinnamic acids, flavonoids and anthocyanins. The slower the drying process (e.g. in the sun), the more the polyphenols are destroyed, due to the combined action of UV rays and endogenous enzymes. If drying is carried out quickly, the consequences may be slight. For capsaicinoids in chilli and paprika, after drying in the sun, around 20-25% disappear, and this continues during storage, with levels falling from 244 mg to 176 mg/kg D.W. in 10 months (IFN et al. 2012).

II.4. Biological activities of polyphenols

Paprika spices *(Capsicium annuum)* are very popular seasoning for culinary and industrial utilization due to the change of sensory quality (taste, aroma, color) of foods and meals with their addition; their health promoting properties; and also, relevant antioxidant activity. Polyphenols are often responsible for the antioxidant capacity of plant products (Škrovánková et al. 2017), Antimicrobial and antioxidant properties of spices render them useful as preservative agents (Chatterjee et al. 2007).

II.4.1. Antioxidant activity

The antioxidants generally in spices and specifically in paprika powder are very effective because they possess excellent antioxidant activity. The spices have been used as antioxidants as whole or ground spice, extracts, encapsulated or as emulsions (Embuscado 2015; P. N. Ravindran 2017). Aside from their efficacy as antioxidants, spices and are classified as all natural, an attractive quality for consumers. spices may be used as a means to control lipid oxidation in foods , for exemple according to Boudalia et al. 2020 adding paprika to cold meats and cheese preparations such as bouhazza, a traditional Algerian cheese.

Antioxidants are substances that prevent oxidation of other compounds. One of the classic definitions of oxidation is combination of an element or compound with oxygen, hence.

The term oxidation. It comes from the French Word *oxider. The word oxide* was coined *by Guyton de Morveau and Antoine Lavoisier,* both French chemists, from oxygene and acide in 1787, Oxidation therefore means gain of oxygen while reduction is loss of oxygen (Embuscado 2015).

To prevent food degradation due to oxidation, employment of antioxidants has become a necessity for food products which are sensitive to this type of chemical change. Phenolic compounds, Tocopherols, ascorbic acid, carotenoids, amino acids, phospholipids, and sterols are the natural antioxidants found in foods, they delay or inhibit lipid oxidation at low concentration (Choe and Min 2009) .

An important area of research today is the control of 'redox' status through the consumption of foods with high antioxidant properties. Natural antioxidants in the diet increase resistance to oxidative stress and can have a substantial impact on human health. Numerous epidemiological studies have suggested that consumption of foods and beverages rich in polyphenols is associated with a reduced risk of cardiovascular disease, stroke and certain forms of cancer (Ghasemnezhad, Sherafati, and Payvast 2011).

Food phenolics render antioxidant activity mainly due to their role as reducing agents, hydrogen donors, and singlet oxygen quenchers. Some phenolics also have the ability to chelate metal ions which act as catalysts in oxidation reactions (Muskawar 2022). Flavonoids are natural polyhydroxylated aromatic compounds they have the ability to scavenge free radicals, including hydroxyl, peroxyl and superoxide radicals and can form complexes with catalytic metal ions rendering them inactive. It has also been found that flavonoids can inhibit lipoxygenase and cyclooxygenase enzymes, the enzymes responsible for development of oxidative rancidity (Choe and Min 2009).

Antioxidants provide protection against oxidation (Amorati, Foti & Valgimigli, 2013). The different factors which affect lipid oxidation include the presence of oxygen and transition metal ions, moisture, heat and light. To prevent, minimize or slow down the rate of lipid oxidation, oxygen and metal catalysts must be removed, or sequestered to render them unreactive (Frankel 2014). The Food prône to oxydation must be stored at low temperatures and/ or shielded from light. Most of the antioxidants isolated from spices and herbs act by reacting with free radicals created during the initiation stage of autoxidation. Others form complexes with metal ions (Waraho, McClements, and Decker 2011).

Paprika is known to contain polyphenolic compounds that have good antioxidant properties (Hong et al. 2020), the study found that paprika could be used as a natural food additive in yogurts.

II.4.2. Antimicrobial activity:

Polyphenols, which play multiple essential roles in plant physiology and have potential healthy properties on human organism, mainly as antioxidants, anti-allergic, anti-inflammatory, anticancer, antihypertensive, and antimicrobial agents (Daglia 2012).

Moreover, the antimicrobial activity of polyphenols occurring in vegetable foods and medicinal plants has been extensively investigated against a wide range of microorganisms. Among polyphenols, flavan-3-ols, flavonols, and tannins received most attention due to their wide spectrum and higher antimicrobial activity in comparison with other polyphenols, and to the fact that most of them are able to suppress a number of microbial virulence factors (such as inhibition of biofilm formation, reduction of host ligands adhesion, and neutralization of bacterial toxins) and show synergism with antibiotics (Othman, Sleiman, and Abdel-Massih 2019). The antimicrobial properties of certain classes of polyphenols have been proposed either to develop new food preservatives (Côté et al. 2010), due to the increasing consumer pressure on the food industry to avoid synthetic preservatives.

According (Nagy et al. 2015) the aromatic spices studied have levels of phenolic constituents that contribute as an antibacterial activity against all Gram (+) bacteria tested and especially against S. aureus, a gainst Gram (-) bacteria was also observed. According to these results, selected aromatic spice may be onsidered as a natural preservative against Food-borne pathogens. And they promote the consumption of spices in fresh or in dry form.

Various studies have demonstrated the antibacterial potential of different species of *Capsicum* spp. Methanolic extracts of *C. annuum* and *C. frutescens* were found effective against food-borne pathogens *Staphylococcus aureus*, *Vibrio cholerae*, and Salmonella Typhimurium. Recently, an aqueous extract of yellow-colored *C. annuum* was found to demonstrate the highest antimicrobial activity against pathogen *P. aeruginosa*. In another study, phenolic compounds capsaicin, dihydrocapsaicin, and chrysoeriol isolated from the hexane and acetonitrile extracts of fruit, peel, and seed of *C. frutescens* demonstrated promising antimicrobial activity against three Gram-negative bacteria (*E. coli*, *P. aeruginosa,* and *K.*

pneumoniae), three Gram-positive bacteria (*Enterococcus faecalis*, *Bacillus subtilis,* and *S. aureus*), and yeast (*C. albicans*). Flavanoid chrysoeriol was found to possess potent antimicrobial potential as compared to the other two isolated compounds (Mohammad Salamatullah et al. 2022)**.**

II.4.3. Other activities

Paprika has received attention as a functional food and as a food additive, with various studies reporting that the phytochemicals in paprika exert antioxidant (Halah and Nayra 2011), anti-cancer (Šaponjac et al. 2014), anti-inflammatory (Chen and Kang 2013), anti-obesity (Maeda et al. 2013), and anti-arteriosclerotic activities (Tsui et al. 2018), 2018). However, to date, there has been no active study on the colors of paprika, especially with yogurt and green paprika juice combined, and no research has been reported on the manufacturing of paprika color-specific yogurts.

Compounds of paprika plus They show strong antioxidative effect, these carotenoids show preventive effect of obesity-related diseases. Dietary paprika carotenoids are absorbed in blood, and they are detected in erythrocytes. It contributes to upregulate endurance performance of athletes by reducing oxygen consumption (VO_2) and the heart rate (Maeda, Nishino, and Maoka 2021).

Components of pepper species effective in reducing the risk of various degenerative, mutagenic, and chronic diseases (Parisi, Alioto, and Tripodi 2020). It has also been used for alleviating toothaches and in the management of the respiratory disease (P. Bosland 1996).Menichini et al. 2009 reported the inhibitory effect of *C. annuum* var. Acuminatum on the enzyme acetylcholinesterase, which is a therapeutic method for the symptomatic management of Alzheimer's disease Menichini et al. 2009. In animal assays, peppers have shown hypocholesterolemic properties (Srinivasan 2005; Aizawa and Inakuma 2009). Capsaicin, the main representative of the pungent components, is a lipophilic alkaloid and because of its analgesic and anti-inflammatory activity has been used in clinical practice. An analysis on rats'

revealed peppers antioxidant capacity, which has defensive effects on the brain cells (Oboh and Rocha 2008).

There is ample historical and scientifically proven information regarding the health ben-efits of spice paprika, including favorable physiological effects, anti-oxidant and anti-inflammatory properties, nonetheless, even though it is consumed in small portions. Also, they preventing diseases such as obesity, heart diseases, and different cancers. An alkaloid, namely, capsaicin, is present in pepper which has antimicrobial, anti-inflammatory, and anticancer effects on the digestive system and is used in relieving pain and also to lose weight.

Experimental part

FIRST PART:

physico-chemical characterisation of paprika powder and optimisation of polyphenol extraction using MSR

Chapter I:

MATERIALS AND METHODS

MATERIALS AND METHODS

Pepper transformed to paprika is traditionally grown in Bejaia, Akbou region and surrounding areas on privately-owned plots. the harvest took place in august 2017.

Survey was carried out among housewives in the region to know the traditional process that they used to prepare the powder and the various stages involved.

I. Sample preparation

Paprika powder was prepared using a series of operations (Figure 4): cleaning, cutting, the pepper intended for drying is cut into a suitable diameter 2-3 cm, which would subsequently affect the colour of the paprika, drying in the sun during 5 to 7 days and grinding in traditional grinding wheel Akbou region Bejaia, average particle size of the obtained powders was between 150µm and 500µm ; Then obtained powder was divided into three batches of the same materials ; which will be stored at three different temperatures: refrigeration (4 °C), freezing (−18 °C) and ambient temperature (for comparison). In this study, the powder was analyzed after 1, 3 and 6 months of storage.

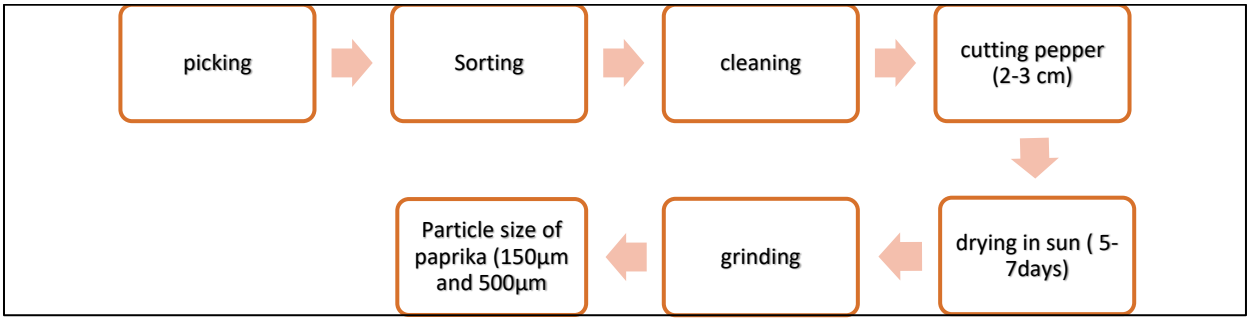

Figure 4: Traditional paprika powder production diagram in Algeria

II. Physicochemical analyses

II.1. Moisture content and dry matter

The thermal drying method was used to determine the moisture content of paprika powder as described by (B. Nagy and Simándi 2008).10 g of sample were placed in an oven at 105 ◦C until the stabilization of the weight. The moisture content was calculated by expressing the weight loss during drying as a fraction of the initial weight.

$$\text{Moisture content (\%)} = (W0 / Wi) \times 100 \quad \text{(Eq.1)}$$

Where W0 corresponds to the weight loss on drying (g) and Wi corresponds to the initial weight of the sample (g).

The determination of the dry matter was carried out by difference between the initial weight (100%) and the moisture content, the result was expressed as follows:

$$\text{Dry matter (\%)} = 100\% - \text{\% moisture} \quad \text{(Eq.2)}$$

II.2. Ash content

The ash content was measured by incinerating two grams of paprika powder, in a calibrated crucible, in the muffle furnace at 550 ◦C for 4 h as described by (Lee et al. 2017). The desiccator-cooled crucibles were weighed and the result was expressed as follows:

$$\text{Ash content (\%)} = [(P2 - P0) - (P0 / P1)] \times 100 \quad \text{(Eq.3)}$$

Where P0: mass of the crucible empty, P1: mass of the crucible empty + sample, P2: mass of ash crucibles + ashes.

II.3. Mesurement of pH

The pH was measured by pH meter using the sample solution made by pouring 90 mL of distilled water into 10 g of paprika sample, mixing it with homogenizer and filtering it with filter paper (Whatman No. 4) in a 100 mL volumetric flask. After calibrating the pH meter with pH 4, 7 and 9 buffer solutions, the electrodes were then immersed deeply into the prepared solution. The pH value was linked on the screen (Lee et al. 2017).

II.4. Color determination

Color is a very important factor visually for consumers who consume the product and plays a pivotal role in stimulating preference. The CIE laboratory coordinates (*L*, a*, b**) were read directly into a glass cuvette with a Mini Scan XETM Spectro-photo-colorimeter. In this coordinate system, the *L** value is a measure of brightness, ranging from 0 (black) to 100 (white), the *a** value ranges from -100 (greening) to +100 (redness) and the *b** value ranges

from -100 (blue) to +100 (yellowing) in another way *L* (lightness), *a* (redness), and *b* (yellowness). Paprika were measured 6 times respectively (Lee et al. 2017).

II.5. Determination of mineral element Concentrations

Atomic Absorbance Spectrophotometry (AAS) is the most widely used method for analysing mineral elements in foods. It is based on the nebulisation of the sample (previously incinerated and extracted in an acid medium) over a flame, which atomises it.

The incineration residues were recovered using the method of Zafar et al. 2010. The white ash was moistened with a few drops of demineralised water and dissolved in 5 mL of aqua regia solution (100 mL of HNO_3 and 300 mL of HCl were placed in a volumetric flask and the volume was adjusted to 1000 mL with demineralised water), then filtered through ash-free grade filter paper (Albet® No. 145) and made up to a final volume of 25 mL with demineralised water (Figure 4). The microelements (iron, copper, manganese, zinc, nickel and cadmium) were measured directly at the appropriate wavelength for each element. Macroelements (potassium, sodium, calcium and magnesium) were quantified after decimal dilution (v/v). To avoid possible interferences, 1 mL of sample solution was mixed with 2 mL of $LaCl_3$ solution (1.8%, w/v) and 7 mL of deionised water for Ca and Mg quantification, and a further 1 mL was added to 2 mL of $CsCl_2$ solution (0.2%, w/v) and 7 mL of deionised water for Na and K determination. Calibration curves were made using standard solutions (1 g/L) of each element (Antunes et al. 2014).

The measurement was carried out using a Solaar M6 series dual-beam atomic absorption spectrophotometer (Thermo Fisher Scientific, Waltham, USA) which contains:

- Sample aspiration and nebulisation system;
- Atomisation system: oxidising air/acetylene flame;
- Primary emission source: a turret that can hold six hollow cathode lamps for successive dosing; hollow cathode lamps (multiple 10 mA intensity lamp (Fe, Cu, Mn and Zn); 12 mA

intensity lamp for K and Na, 6 mA intensity lamp for Ca and Mg; 15 mA intensity lamp for Ni and 6 mA intensity lamp for Cd).

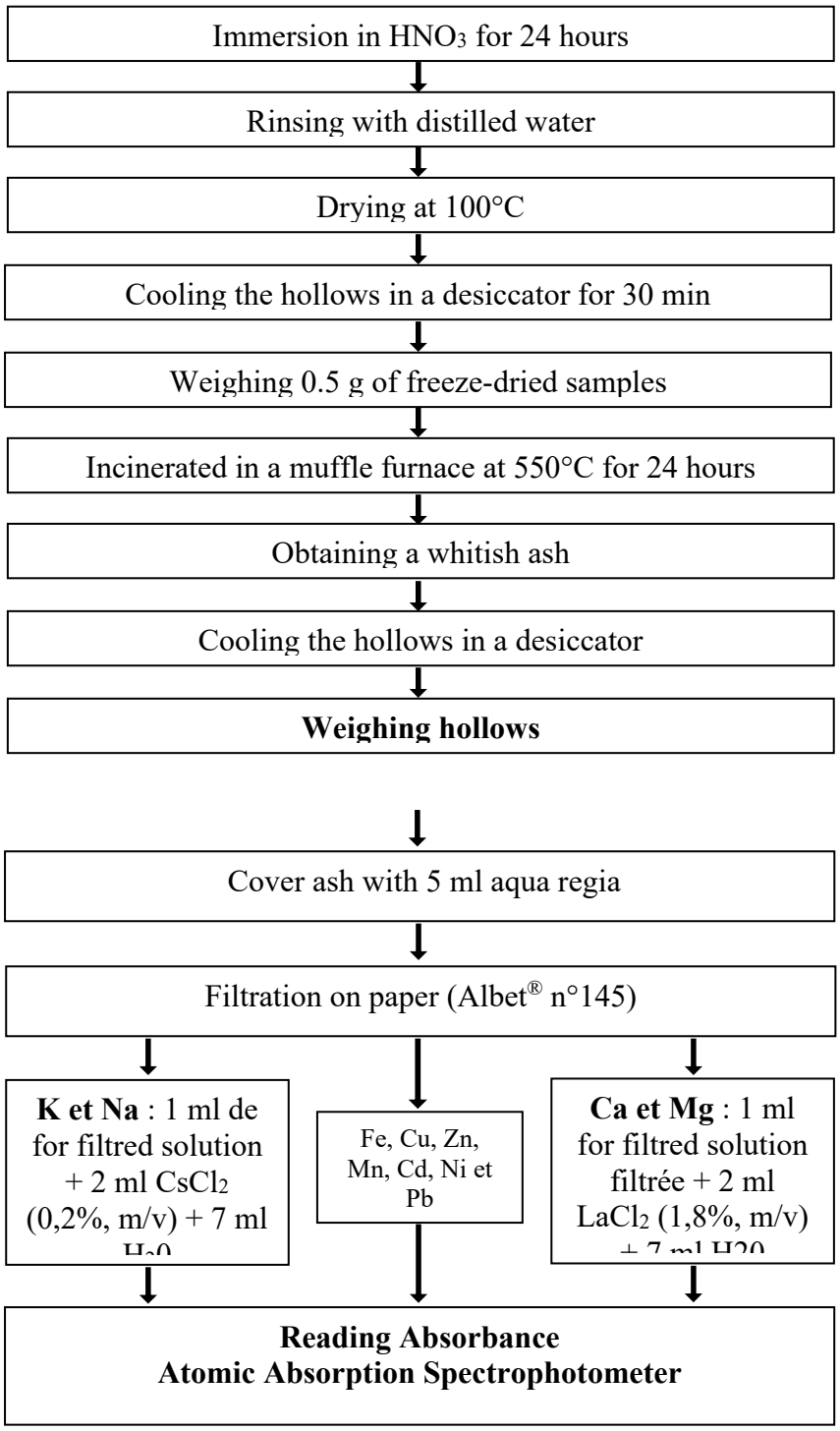

Figure 5: AAS mineral analysis diagram

III. Extraction total phenolic compounds

III.1. Ultrasound assisted extraction optimization using response surface methodology

Use ultrasound energy and solvent to extract target compounds from the plant matrix. Ultrasound is a mechanical wave with a frequency (> 20 kHz) (Périno-Issartier et al. 2013). These waves consist of a series of compression and rarefaction cycles that can propagate through the plant matrix inducing the displacement of the molecules from their original positionsIn this technique, the main objective is to optimize the response surface that is influenced by various process parameters. RSM also quantifies the relationship between the controllable input parameters and the obtained response surfaces (Aslan and Cebeci 2007). RSM was used to study the simultaneous effects of parameters (single factors) for maximizing yield of TPC in extraction using an ultrasonic apparatus.

III.1.1. Equipment and procedure

For the UAE experiments, an ultrasonic system was used as an ultrasound source (SONIC Vibra cell, VCX 130 PB Stepped microtips and probes, No 0.630-0422 Newtown, CT, USA), frequency of this apparatus was fixed at 20 kHz.

For the extraction, one gram of paprika was placed into a 100 mL beaker containing the extracting solvent and sonicated at the required temperature (25 ± 2 °C). After extraction, the sample was recovered by filtration in a Buchner funnel At the end of extraction, mixtures were filtered through N°1 Whatman filter paper and extracts were collected in a volumetric flask and kept in the dark at 4°C until TPC analysis.

III.1.2. Choice of extraction conditions

For the optimization of the UAE procedure, the influences of the process parameters were initially separately investigated in single–factor experiments to limit the upper and lower limits for these factors. Firstly, the most suitable solvent for the extraction (ethanol, methanol, and acetone) was selected. Then, the influence of sonication time *X1* (5–30 min), amplitude

X2 (20–100), solvent concentration *X3* (20–80%) and liquid-to-solid ratio *X4* (10–50 mL/g) were investigated.

III.1.3. Box-Behnken experimental design (BBD)

The design of experiment method is a technique for the optimal organization of experiments in order to obtain a maximum of information in a minimum number of tests with the best possible accuracy. The RSM is experimental design which has been established for optimizing various processes to investigate the relationship between the extraction conditions and the yield of TPC (mg GAE/g DP). Four factors which are coded at three levels (− 1, 0, + 1) were employed (Table 7).

Table 7: Experimental values and coded levels of the independent variables used for the BBD

Independent variable	**Factor Levels**		
	-1	**0**	**+1**
X_1: Sonication time (min)	5	12.5	20
X_2: Amplitude radiation (%)	40	60	80
X_3: Ethanol concentration (%)	20	50	80
X_4: Solvant-solid ratio (ml/g)	10	30	50

This design require an experiment number according to N = k2 + k + cp, where k is the factor number and cp is the replicate number of the central point hence 27 runs were resulted (Souza,

Santos, and Ferreira 2005) (Table 8). This model utilizes the second order polynomial equation (Eq. 4) which was used to predict the optimum condition of extraction process.

$$Y = B_0 + \sum_{i=1}^{K} B_i X_i + \sum_{i=1}^{K} B_{ii} X^2 + \sum_{i\rangle j}^{K} B_{ij} X_i X_j + E \qquad \text{(Eq. 4)}$$

Where Y is the predicted response (TPC yield), β0 is a model constant βi, βii, and βij are the coefficients of the linear, quadratic and interactive terms, respectively, *Xi* and *Xj* represent The coded independent variables.

Table 8: BBD experimental design with the independent variables and the experimental data for the response

Run	Extraction conditions				TPC yield (mg GAE/g DW)	
	***X*₁: Sonication time (min)**	***X*₂: Amplitude radiation (%)**	***X*₃: Ethanol concentration (%)**	***X*₄: Solvant-solid ratio (ml/g)**	**Experimental results**	**Predicted results**
1	12.5	80	20	30	10.79±0.69	10.36
2	20	60	80	30	8.78±0.38	8.57
3	12.5	40	50	10	6.51±1.11	6.65
4	20	60	50	50	9.40±1.12	9.35
5	12.5	80	50	50	8.75±0.32	8.86
6	12.5	40	50	50	9.45±0.16	9.75
7	5	60	80	30	8.60±0.75	8.92
8	12.5	60	20	10	8.78±0.10	9.03
9	5	60	20	30	10.04±0.11	9.94
10	20	80	50	30	8.47±0.18	8.96

11	12.5	60	20	50	9.83±0.05	10.27
12	12.5	60	50	30	8.17±0.29	8.05
13	5	80	50	30	7.07±0.28	7.70
14	12.5	60	80	50	10.10±0.70	10.21
15	12.5	80	50	10	7.38±0.32	7.33
16	5	40	50	30	9.29±1.22	9.29
17	12.5	80	80	30	7.76±0.16	7.96
18	5	60	50	10	7.10±0.23	7.26
19	12.5	60	50	30	8.00±0.23	8.05
20	5	60	50	50	9.75±0.46	9.47
21	20	60	20	30	9.63±0.17	9.83
22	12.5	60	50	30	8.17±0.21	8.05
23	12.5	40	80	30	9.22±0.38	9.33
24	12.5	60	80	10	6.80±0.32	6.82
25	12.5	40	20	30	9.32±0.06	9.20
26	20	40	50	30	7.76±0.15	7.58
27	20	60	50	10	6.89±0.44	6.93

III.1.4. Optimum yield of extraction

After developing the polynomial equation for the response with the independent variables, optimization was performed to find out the level of independent variables (*X1*, *X2*, *X3,* and *X4*) that would yield a maximum value of TPC. To verify the adequacy of the models, additional extraction trials were carried out at the optimal conditions predicted with the RSM and the obtained experimental data were compared to the values predicted by the regression model.

III.1.5. Spectrophotometric determination of phenolic compounds of paprika extract

The Folin-Ciocalteu assay was used to determine the TPC of the obtained extracts according to the method described by (Georgé et al. 2005). In tubes, 2.5 mL of Folin-Ciocalteu reagent (1/10) were added to 0.5 mL of paprika extracts. After 2 min, 2 mL of 7.5 % (w/v) sodium carbonate (Na_2CO_3) solution was added to each tube, and the tubes were incubated for 15 min/50 °C. After cooling, the absorbance was measured at 750 nm. The TPC was expressed as gallic acid equivalent on dry weight basis (mg GAE/g DW). Gallic acid was used to prepare a calibration curve with different concentrations (20 to 100 µg/mL) using the equation 2:

$$y = 0.12x \quad (R^2 = 0.998) \qquad \text{(Eq.5)}$$

The optimized extract was also analyzed for the content of flavonoids:

The amount of ***total flavonoids*** in the extracts was estimated using the methodology of (Quettier-Deleu et al. 2000) Concentration of flavonoids was expressed as mg of quercétine equivalent per g of dry weight (mg QE/g_{DW}) and obtained from a standard curve prepared with quercétine (5 to 50µg/mL) using :

$$y = 28.66x + 0.019 \qquad (R^2 = 0.982) \qquad \text{(Eq. 6)}$$

IV. Statistics analysis

All experiments were carried out in triplicate. The experimental results were expressed as means ± S.D (n = 3). Influence of each factor on the TPC yield in the single factor experiment for the UAE was statistically assessed by ANOVA and Tukey's post hoc test with 95% confidence level.

The JMP (Version 7.0, SAS) and Design-Expert (Trial version 8.0.7.1) softwares were used to construct the BBD to analyze all the results.

Chapter II:

RESULTS AND DISCUSSION

RESULTS AND DISCUSSION

I. Physicochemical analyses of paprika powder

I.1.Moisture, ash and pH of paprika

The results the determination of Physicochemical analyses content of paprika before storage are shown in the **Table 9**. Results of the moisture content are close to those reported by (B. Nagy and Simándi 2008) who reported 18% and the research of Tudomanyegyetem and Kar on 2006 the moisture ranged from 6.85 to 12.56% much lower than our results . in the work of (Lee et al. 2017)the moisture change from 6.94±0.81, 7.57±1.25 to 11.85±1.66 % depending on the drying red paprika method used for example vacuum they use freeze-drying of osmotic dried red paprika in sugar and corn syrup, vacuum freeze-drying of osmotic dried red paprika in sugar and vacuum freeze-dried respectively. the high moisture content of our powder means that it is made using traditional drying methods in more thermal treatment such as microwave treatment could reduce the moisture content sample due to heat removal of moisture from the food matrix (Shoqairan et al. 2023). Data of Jung and Hong (2017) found that the moisture content of dried paprika was 11.19% on the freeze-drying paprika and the amount of water was increased by 18.19% on the 15th day of the storage cycle.

According to Tudomanyegyetem and Kar (2006) They study moisture Hungarian paprika powders with an initial humidity between 6.85 and 7.56% then The moisture content of each was increased by 1, 2, 3, 4 and 5%; This affects the color of paprika powder with it becomes darker and clean red color, so perhaps it entailed better perception, acceptance and choice for the consumer as is the case with our powder, which has a high moisture content. In addition moisture content of paprika were developed by Shirkole and Sutar (2018) to estimate the stability of the paprika. The estimated shelf life of low moisture paprika (4.40% dry basis) was found to be 101 and 31 days in HDPE and LDPE packages, respectively when stored in

domestic condition. In industrial storage condition, the shelf-life prediction was 5.47 years in HDPE and 1.68 years in LDPE packages.

Codex Alimentarius limited moisture content of red pepper powder to 11%. Moisture content in dried products is of great importance for satisfactory microbiological stability and for a reasonable shelf life. The initial moisture content of pepper powder is very important because it is strongly correlated with the stability of ascorbic acid and pigments as well as microbial aspects (Cankaya, Hayoglu, and Turkoglu 2017)

On the other hand, the results obtained of the ash content seem the same than reported by Štursa, Diviš, and Pořízka 2018 and (Lee et al. 2017) whom recovered 5.8 % and 5.14%, respectively. which are below the maximum permissible limit value (10%) according to ISO 7540 standard. In paprika Serbian (Vasić et al. 2021) the total content of ash shown that values ranged from 5.40 to 7.80% therefore slightly higher than the ash content of our powders.

Molnár, Kónya, et al. (2018) founds that average content of ash in the paprika samples was 5.8 ±0.6 of different region; but he found significant differences between samples from more distant regions (Hungary, Spain, Turkey, Bulgaria…) can be differentiated according to their different chemical composition, while samples from similar regions (Hungary, Slovakia, Romania) is more difficult to differentiate; so geographic origin influences ash content and chemical composition.

paprika have the value of pH 6.03±0.03 and was slightly acidic and close to neutrality. The pH value found is slightly higher for freeze-dried paprika from the Korea region, which is more acidic, with values between 5.05 and 5.35 (Lee et al. 2017) as the same are those reported by Hong et al. 2013. Zaki et al. (2013) found the pH of the paprika ranged from 5.1 to 6.3; generally, pH value indicates enzymatic activity and paprika's shelf life.

Table 9: Physicochemical analysis of Paprika

	Moisture content (%)	**Dry mater (%)**	**Ash content (%)**	**pH**
Paprika	16 ± 1	84±1	5.76±0.05	6.03±0.03

I.2.Color determination

The values for the apparent colors of paprika powder are shown in **table 10.** The apparent color was expressed as the *L*, a*,* and *b** values of the CIE-LAB color system.

For the apparent color, the *L**= 34.66, *a**= 49.90, and *b**=25.34, these values mean that our powder is darker; this may be due to oxidation of the sugars during traditional drying; however, the red parameter is higher than the yellow parameter, which means that there are more capsanthin then β carotenoids.

Kang et al. 2015 reported that the color values of paprika were measured as *L* value of 30.75, *a* value of 23.27, *b* value of 11.38. Ha et al. 2012 as L value of 36.4, *a* value of 34.3, *b* value of 18.9. Compared with our results, the degree of clarity is the same, but the colour of our samples is more important than their results.

however, according to the Lee et al. 2017 study based on the color of paprika samples, which has also undergone osmotic dehydration is clearer and brighter *L**= 55.98. When developing processed foods of paprika, research and development considering this part will lead to products with better merchandise; so, the method of drying can affect the color of the paprika. For example The effect of drying method on color changes of paprika powders was also investigated, with freeze drying reported as the best method for retention of paprika color (Topuz, Feng, and Kushad 2009).

Table 10: The L*, a*, and b* values of the CIE-LAB color system of paprika

Color		
L*	***a****	***b****
34.63±1.02	49.90±0.98	25.34±1.30

I.3. Determination of Concentration of mineral element

Elemental composition of paprika powder has been determined by using Atomic Absorption Spectrophotometer (AAS). A total of 29 elements macro-elements and trace elements; have been measured. Their concentrations are expressed in ppm.

The results of elemental analysis obtained by comparator method of AAS techniques are shown in **Table 11** in mg/g dry weight of the samples. It is to be noted that each result is an average of at least three independent measurements with a precision of about ± 1%.

The results show that paprika powder contains a very high level of rubidium (Rb) with a concentration of 355.851 ppm, Rubidium is a metallic chemical element found in group 1 (alkalis) of the periodic table. Its name comes from the Latin word "Rubidius" which means "red", In humans, the daily Rb requirement was estimated to be 10 µg Rb/kg body weight, whereas the recommended daily intake was proposed to attain 20µg Rb/kg BW (Seixas and Pierce 2005).

followed by iron (Fe^{+2}) with 1.237 ppm, then lithium Li^{+1} with 0.505 ppm, Zn^{+2} with 0.349 ppm, Mn^{+2} with 0.214 ppm, Sr with 0.187 ppm and finally copper (Cu^{+2}) with 0.128 ppm representing trace elements. However, our powder does not contain any of these following macro-elements: Ca^{+2}, Mg^{+2}, K^{+}, Na^{+2}, however the remainder is found with minimal concertation which varies from 0.057 to 0.001 ppm.

According (Zaki et al. 2018) the mineral composition of paprika powder from the three Moroccan regions. The results show that this spice is rich in potassium, magnesium and

relatively low in sodium. As for calcium its average content is 88 to 110 mg/100g PS. Thes results are in line with those obtained for Serbian Serbian paprika (Poór et al. 2018) contrary to our results.

Comparing our data with the literature, a lower Fe content was observed in chili pepper fruits from Turkey than in our samples and sweet pepper fruits from Poland, Spain and hungry (Poór et al. 2018).

The Zn content of investigated Spanish sweet paprika samples was significantly higher than then our powder it varied from 11,3 to 33,4 µg g-1 d.m. and higher than processed samples of European, African and Asian origin (Poór et al. 2018).

The heavy metals analyzed were either not detected (lead (Pb) and cadmium) or are present in low concentrations (copper and zinc), which is in line with current regulations in force (< 5 ppm). Same results are observed with Hungarian paprika simples, Turkish sweet red pepper and in Russian sweet pepper fruits. Palacios-Morillo and co-workers. (2014) revealed a higher content of Pb in Spanish paprika samples compared to our results (70.63-267.90 mg kg-1 d.m.). however, our results that concentrations of toxic elements were below the legal threshold limit in foodstuff.

We can conclude that the investigated paprika were of good quality and contained only very small amounts of toxic elements and the higher level of Rb, can be used for the determination of geographical origin of paprika.

Table 11: Concentrations of elements (ppm) mg/Kg in paprika powder.

Cd	**Co**	**Cu**	**Cr**	**Fe**	**Pb**
0,001	0,035	0,128	0,011	**1,237**	0,009
Mn	**Hg**	**Ni**	**Ag**	**As**	**Ba**
0,214	0,001	-0,004	0,017	0,002	0,057
Ca	**Ga**	**In**	**Li**	**Mg**	**K**
N/A	0,001	0,001	0,505	N/A	N/A
Rb	**Se**	**Na**	**Sr**	**Zn**	**Tl**
355,851	0,002	N/A	0,187	0,349	0,003

II. Extraction methodology for phenolic compounds

II.1. Ultrasound assisted extraction optimization using response surface methodology

II.1.1. Choice of extraction conditions

A preliminary study was previously conducted to determine the range of each independent variable would influence TPC pepper recovery. Selection of the most suitable solvent was the initial step. We tested three solvents; ethanol, methanol and acetone at 80% (*v/v*). Ethanol and methanol offered a maximum TPC yields, and since many reports suggested that ethanol was the most appropriate solvent for the extraction of various phenolic compounds from different plant materials (Chen, Zhao, and Yu 2015; He et al. 2016) and it is categorized as GRAS (Generally Recognized as Safe) for application on food system, it was chosen as extraction solvent.

Selection of UAE conditions such as time, amplitude, ethanol concentration, and solid–liquid ratio was based on the previous single factor experiments. Table 7 shows the variables and their levels (− 1, 0, + 1) used in the experimental design. A BBD model analyzed randomly the experimental data obtained from the 27 trials (Table 8).

II.1.2. Optimization of UAE conditions

The BBD was used to optimize the UAE conditions such as sonication time, amplitude radiation, ethanol concentration and solvent-solid ratio for pepper TPC recovery, and also to evaluate interaction effect between all factors. The TPC yields obtained from the BBD trials are reported in Table 8, they ranged from 6.51 to 10.79 mg GAE/g DP, which means that the tested factors influenced the recovery of phenolics. The data from 27-runs of experiments was analyzed using ANOVA (Table 12). A second order polynomial model (Eq. 7) fitted well the experimental data ; it is highly significant ($P < 0.0001$) and indicates that the quadratic experimental model proposed was in good agreement with the experiment results. Additionally, determination coefficient ($R2 = 0.967$) revealed that approximately 96% of the total variations were well explained by the experimental data model which means that only 4% of total variations were not explained by the model. To determine whether the goodness of the statistical quadratic model, the adjusted R2 value should be close to 1 (0.929), and explained a high correlation degree as shown in Table 12. The coefficient of variation value (CV = 8.58%) was less than 10% and root mean square error (RMSE = 0.305) was very low, that displayed also the perfect precision and a better reliability of the experimental values and the predicted ones. The non significance level of lack of fit, $11.46 > 0.05$, confirms the validity of the model. Therefore, the model is adequate and can be used to optimize the extraction conditions using ultrasound technique.

The statistical analysis (Table 12) showed that the independent variables have a significant effect on TPC recovery from pepper sample. The two linear terms, X3 : ethanol concentration ($p = 0.0001$) and X4 : liquid solvent to solid ratio ($p = 0.0001$) affect more significantly compared to others without significant effect. In addition, different interactions between factors were observed such as X1X2 ($p < 0.0004$), X2X3 ($p = 0.0004$), X3X4 ($p = 0.0032$) and the less interaction was observed between X2 and X4 ($p = 0.02$). Finally, a quadratic effect was

observed just with *X3 2* (p = 0.0001). Hence, by removing the insignificant terms, the predictive quadratic equation (Eq. 7) is :

$$Y(TPC) = 8.11 - 0.593X3 + 1.152X4 + 0.732X1X2 - 0.731X2X3 - 0.39X2X4 + 0.56X3X4 + 1.01X_3^2 \quad (\text{Eq.7})$$

Table 12: Estimated regression coefficients for the quadratic polynomial model and the analysis of variance (ANOVA) for the experimental results of TPC.

Term	Estimate	Sum of squares	DF	F-Value	Prob>F
Intercept Model	8.11	33.456559	14	25.5226	<.0001
Linear					
X_1	-0.077333	0.071765	1	0.7665	0.3985
X_2	-0.11125	0.148519	1	1.5862	0.2318
X_3	-0.593583	4.228094	1	45.1561	<.0001
X_4	1.152	15.925248	1	170.0818	<.0001
Interaction					
X_1X_2	0.732	2.143296	1	22.8904	0.0004
X_1X_3	0.147	0.086436	1	0.9231	0.3556
X_2X_3	-0.73125	2.138906	1	22.8435	0.0004
X_1X_4	-0.0345	0.004761	1	0.0508	0.8254
X_2X_4	-0.3905	0.609961	1	6.5144	0.0254
X_3X_4	0.5625	1.265625	1	13.5169	0.0032
Quadratic					
X_1^2	0.1449583	0.112069	1	1.1969	0.2954
X_2^2	0.0203333	0.002205	1	0.0235	0.8806

X_3^2	1.0043333	5.379656	1	57.4548	<.0001
X_4^2	-0.108542	0.062834	1	0.6711	0.4287
Lack of fit		1.1043281	10	11.4636	0.0828
Pure error		0.0192667	2		
C.V.%	8.584926				
RMSE	0.305995				
Corr total		34.580154	26		<.0001
R^2	0.967508				
R^2 Adjusted	0.9296				

II.1.3. Optimisation by RSM

The response surface is used to find the experimental region that gives a higher TPC value. They play a principal role to identify the optimum condition of the output variables efficiently, under which dependent variable could reach the maximum response. In the response surface plot (Figure 5), the extraction yield of TPC was obtained along with two continuous variables, while the other two variables remain constant. From these dimensional profiles, among the plotted surfaces, two surfaces showing an interaction effect between the two parameters; sonication time and amplitude (X1X2) and solvent concentration and ratio (X3X4) represented in figure 6.A and 1.F, respectively, have an effect that is both positive and significant on the efficiency of TPC extraction. While the areas showing an interaction effect between the two parameters; the amplitude and solvent concentration (X2X3) and the amplitude and ratio (X2X4) represented in Figure 5D and E, respectively, have an effect that is both positive and significant on TPC extraction efficiency. Additionally, from these dimensional profiles, we noticed that the most influential factor governing TPC yield was ethanol concentration and solvent-solid ratio. The TPC extraction values increased with the increase in both ethanol

concentration (Figures 5B, D, and F) and solvant-solid ratio (Figures 5C, E, and F) ; for sonication time it appears that 5 min gives the highest extraction rate of TPC (Figures 5A, B, and C), and the amplitude was situated at 40 MHz.

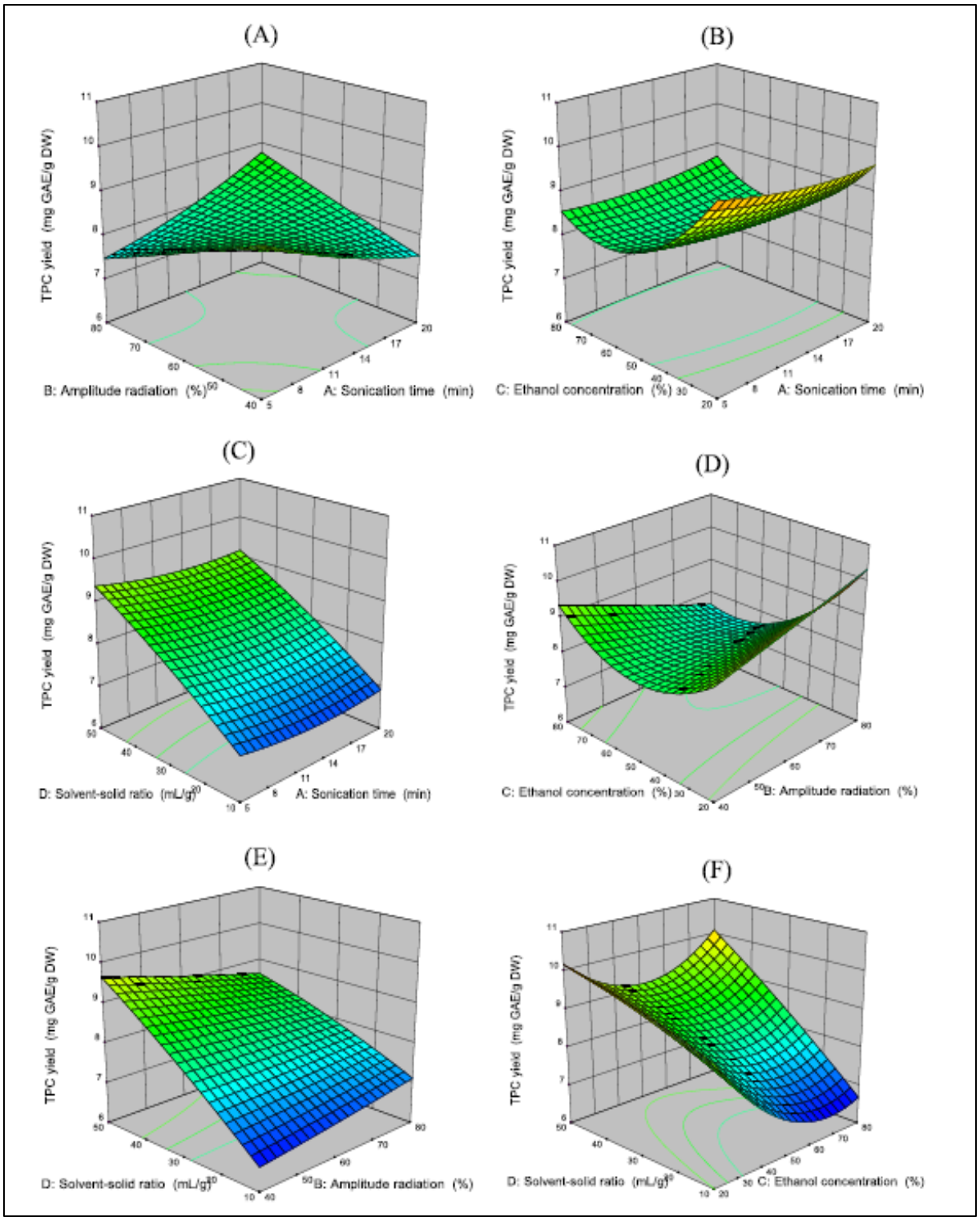

Figure 5: Ultrasound assisted extraction parameters effect on total phenolic yield from paprika powder: (**A**) Sonication time and amplitude radiation, (**B**) Sonication time and ethanol concentration, (**C**) Sonication time and solvent-solid ratio, (**D**) Amplitude radiation and ethanol concentration, (**E**) Amplitude radiation and solvent-solid ratio and (**F**) Ethanol concentration and solvent-solid ratio

II.1.4. Validation of the developed models

The UAE conditions that can give maximal yield of TPC are : ultrasonic time : 5 min, amplitude : 40%, ethanol concentration : 80% and solid–liquid ratio : 50 mL/g (Figure 6). Under these conditions, the quadratic model predicts a value of 12.23 ± 1.01 mg GAE/g DP for TPC recovery which is very close to experimental assay (12.33 mg GAE/g DP) occurred within the same conditions. This confirms the effectiveness of the proposed model to reflect the expected optimization.

*

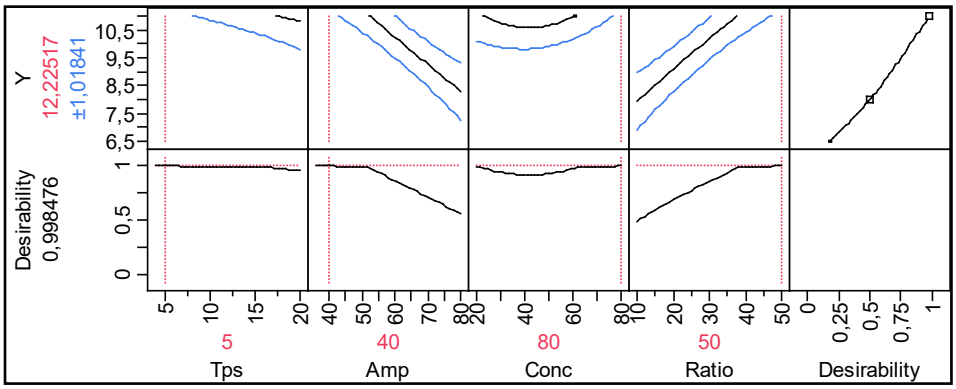

Figure 6: optimal conditions for extracting phenolic compounds by UAE

Table 13: Predicted and experimental values of response under Optimum conditions

Response	**Optimum constraction conditions**				**Result values (mgGAE/g DW**	
	Sonication time min	**Amplitude %**	**Ethanol concentration%**	**Ratio ml/g**	**Experimental**	**predicted**
TPC yield	5	40	80	50	12.3±0.25	12.2±1.01

SECOND PART:

effect of storage on polyphenols content and biological activity

Chapter I:

MATERIALS AND METHODS

MATERIALS AND METHODS

I. Determination of the total phenolic content (TPC) and the total flavonoids content (TFC)

The conditions for extracting the phenolic compounds were optimized in the first part of the work, which will then be used to monitor changes in the quantity of total polyphenols and their biological activity over time.

The protocols used to quantify the phenolic compounds are also described in the first part.

II. Determination of DPPH radical scavenging activity

There are many methods to measure the antioxidant activity, in our case we choose the scavenging activity of the radical 1,1-diphenyl-2-picrylhydrazyl (DPPH), this radical has an absorption maximum at 517 nm, which disappears (bleaching of the purple-colored solution) after it's reduction by an antioxidant. 2 mL of DPPH solution in methanol (0.1 mM) were added to 1 mL of extract. Absorbance was measured at 517 nm after incubation during 30 min at darkness. Since mature peppers contain pigments, we used a blank without DPPH and the absorbance of the blank was subtracted from the sample reading at 517 nm(Vega-Gálvez et al. 2009). The antioxidant capacity of the extract was expressed as inhibition percentage and calculated according to the following equation Eq 8:

$$\%Inhibition = (A_{DPPH} - A_{samples}) \times 100 / A_{DPPH} \qquad \text{(Eq 8)}$$

Where A_{DPPH} is the absorbance of DPPH radical solution; $A_{samples}$ is the absorbance of extract after addition of DPPH at 30 min. The effective concentration of sample required to scavenge DPPH radical by 50% (IC_{50} value) is obtained by linear regression analysis of dose-response curve plotting between % inhibition and concentration.

III. Antibacterial activity

Antibacterial capacity test can be used for drug discovery, epidemiology and prediction of therapeutic outcome(Balouiri, Sadiki, and Ibnsouda 2016). In the present study, we tested the

antibacterial capacity of phenolic extracts of paprika powder during storage (1-6 months) at three different temperature levels against four pathogenic bacteria (two Gram+: *Staphylococcus aureus ATCC 25923* and *Bacillus subtilis ATCC 6633* and two Gram$^-$: *Escherichia coli ATCC 25922* and *Pseudomonas aeruginosa ATCC 27853*). These strains were provided by Pasteur institute. The most methods used for the antibacterial activity evaluation are: discs diffusion, agar dilution and broth dilution. These methods are relatively rapid, inexpensive and do not require sophisticated laboratory equipment. All assays were performed in triplicate in two independent experiments to ensure reproducibility.

III.1. Diffusion method

The antibacterial activity of phenolic extracts was determined by agar well diffusion method as adopted by (Palladini et al. 2023), bacterial culture inoculums grown previously on nutrient agar medium was prepared for each strain in sterile physiological water with an optical density of 0.50 McFarland. 6 mm diameter whatman paper discs were soaked with 20 µL of phenolic extracts (50 mg/mL) and then placed on the surface of Mueller Hinton agar, formerly inoculated with an inoculum containing approximately 10^6 CFU/mL. After incubation at 37°C for 24 hours, the microbial sensitivity was evaluated by measuring the diameter of the inhibition zone (DIZ) (mm) figure 8.

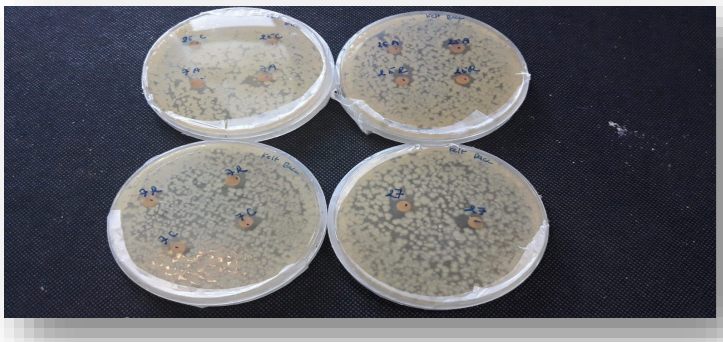

Figure 8: Antibacterial activity by diffusion method of phenolic extracts from paprika

III.2. Broth microdilution method

This method is appropriate to determine the minimum inhibitory concentration (MIC) values also minimum bactericidal concentration (MBC) it known as the minimum lethal concentration (MLC) (Balouiri et al., 2016). Bacterial culture is prepared in the same way as in the diffusion method. The procedure involves preparing two-fold dilutions of the antibacterial agent (e.g.1, 2, 4, 8, 16 and 32 mg/mL) in a liquid growth medium dispensed with smaller volumes using 96-well microtitration plate (microdilution). Then, each well is inoculated with a bacterial inoculum prepared in the same medium after dilution of standardized bacterial suspension adjusted to 0.5 McFarland scale. After well-mixing, the inoculated 96-wells microtitration plate is incubated (mostly without agitation) under suitable conditions depending up on the test microorganism.

III.2.1. Determination of MIC and MBC

Broth microdilution method (Agrawal and Shevade 2014) was used to determine the Minimum Inhibitory Concentration (MIC) and the Minimum Bactericidal Concentration (MBC) values for paprika extract exhibiting antibacterial activity toward test pathogens. The crude extract concentration of 100 mg/mL was prepared in DMSO, and then diluted serially in Mueller Hinton Broth (MHB), ranging from 1/2 to 1/1024. 100 µL of each extract dilution was combined with 100 µL bacterial inoculum (10^6 CFU/mL in fresh MHB) in each well. The 96-wells microplate was incubated for 24h at 37 °C. MIC was defined as the lowest concentration of the plant extract at which the bacteria will display no growth. 20 µL of each well with no visible growth was sub cultured since the last positive well, into MHA plates and incubated as described before. The minimum bactericidal concentration was considered the lowest concentration of the extract which did not show any bacterial growth. All measurements were performed in triplicate, The diagram above shows the steps involved in the microplate process (**figure 9).**

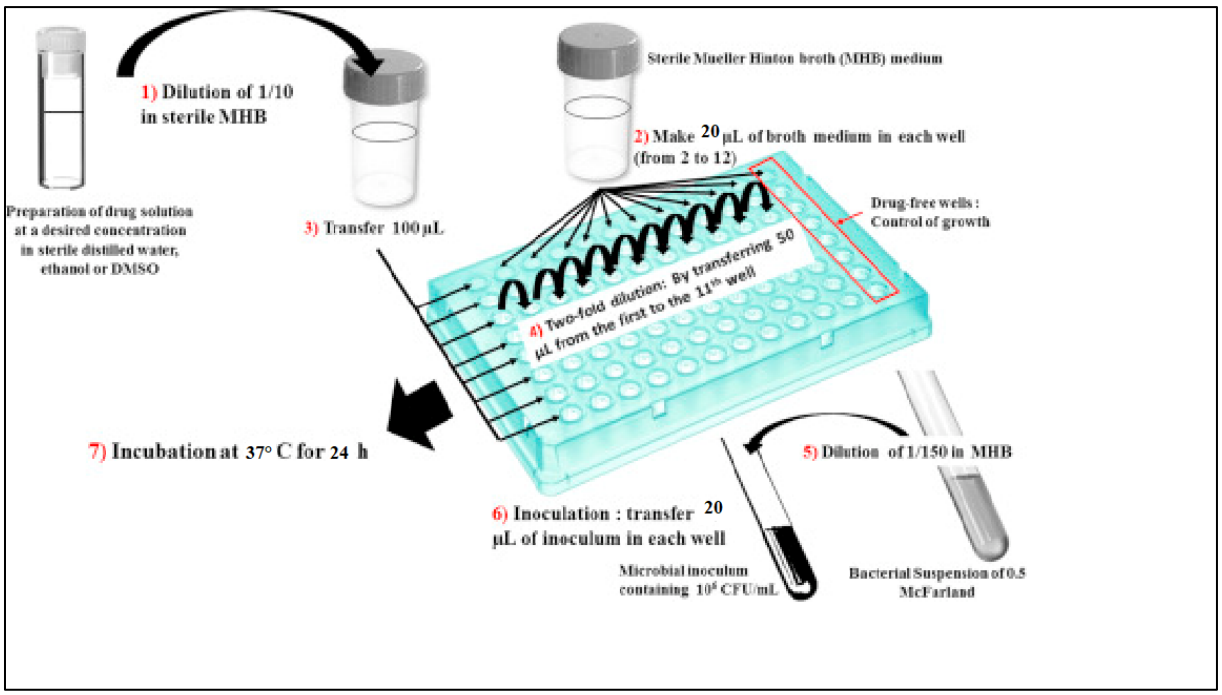

Figure 9: Diagram of broth microdilution method for determination of MIC and MBC of phenolic extracts

IV.Statistics analysis

The analysis was carried out in three replicates for all determinations. The mean and standard error of means were calculated. The data were analyzed by one way analysis of variance ANOVA). Significance of the differences was defined as $P < 0.05$.

Chapter II:

RESULTS AND DISCUSSION

RESULTS AND DISCUSSION

I. Total phenolic and flavonoid contents

I.1.Total phenolic content (TPC)

General pattern of change in TPC is observed in figure 10 over the 06 months of storage. We carried out the analysis in the first month just after obtaining the powder, in the third month and in the sixth month of storage. We observe a superposition of curves representing the TPC of the powder stored at refrigeration temperature (4°C) and freezing temperature (-18°C) during the entire storage period, so the polyphenols behave in the same way at low temperatures. There is a decrease in TPC during the third month of storage and then a further increase in this content, unlike the TPC of powders stored at room temperature, in the third month there is an increase in TPC, However, in the sixth month of storage, the TPC decreased considerably for the two powders stored at 4°C and -18°C, compared with the powder stored at room temperature, which remained more or less stable.

The TPC were better preserved at ambient temperature, it increased significantly ($p < 0.05$) in the stored sample during 3 months (from 12.33 ± 0.25 to 13.91 ± 0.56 mg GAE/g DP) and decreased significantly ($p < 0.05$) after the 6th month of preservation (from 13.91 ± 0.56 mg GAE/g DP to 12.83 ± 0.24 mg GAE/g DP) but remaining constant with respect to its initial content. These results are in agreement with the findings of other authors (Moldovan, Popa, and David 2016) who studied the Cornelian cherry (*Cornus mas* L.) fruits room temperature without significant loss of their bioactive compounds (TPC) and those obtained by (Iqbal et al. 2015), who indicated that the increase in temperature and storage time induce a gradual and regular decrease in TPC, respectively.

For storage at low temperature, the TPC was affected negatively and varied from 8.75 ±0.13 to 13.91 ± 0.56 mg GAE/g DP (Table 14). No significant difference ($p > 0.05$) was noticed between the samples stored at low temperatures (Table 14), whether refrigerated or frozen, after

3 and 6 months of preservation (13.22 ± 0.58 mg GAE/g DP, 13.62 ± 0.18 mg GAE/g DP and 8.99 ± 0.83 mg GAE/g, 8.75 ± 0.13 mg GAE/g DP), respectively. More significant decrease ($p < 0.01$) was noticed during the refrigeration and freezing modes from 3 to 6 months (from 13.22 ± 0.58 to 8.99 ± 0.83 mg GAE/g DP and from 13.62 ± 0.18 to 8.753 ±0.129 mg GAE/g DP), respectively.

It is to highlight that no significant differences ($p > 0.05$) were observed in TPC of the powders stored at low and ambient temperatures (13.91 ± 0.56 mg GAE/g DP (ambient temperature) vs. 13.22 ± 0.58 mg GAE/g DP (refrigeration) and 13.62 ±0.18 mg GAE/g DP (freezing)) for the 3rd month of preservation. Indeed, the TPC at ambient temperature, and also the study conducted by (Oranusi et al. 2013) showed that among some imported spices in Nigeria, paprika powder contains a considerable amounts of TPC and TFC. According to (Dubey et al. 2015), indigenous chilli genotypes from North East India showed variation in TPC and it is the *Capsicum frutescens* genotype which contains the highest content, the other genotypes showed lower levels but which exceed those found in this study. Similarly, (Škrovánková et al. 2017) studied paprika spices (*Capsicium annuum*) from Czech, Austrian, and Slovak producers, they recorded generally considerable quantities of TPC but it was the hotter samples of paprika spices that have slightly higher values of TP than sweet types, decreased with longer storage time and the rate of decrease at low storage temperature was larger than that at room temperature which is confirmed by (Cheng et al. 2017). Indeed, the storage of vegetable products at a temperature that is too low, below the "chiling injury" temperature, causes cell dislocation, leading to the enzymatic browning reaction and degradation of phenolics. The chilling injury is also considered as oxidative stress, it is leading to an increase in enzyme activity (Tomás-Barberán and Espín 2001).

With the exception for the results obtained after the 6th month of storage at ambient temperature (where we have noticed a decrease), our results are in disagreement with those reported by other

researchers (Park and Kim 2007), who reported that paprika samples stored at ambient temperature retain less phenolics than those stored at 30 °C after 3 months. In the same way, the decrease in TPC has been reported in dry hot peppers after 5 months of storage at 30 °C (Iqbal et al. 2015), and this degradation has been related to polyphenol oxydase (PPO) activity (M. Deng et al. 2018).

According to Dubey et al. (2015), indigenous chilli genotypes from North East India showed variation in TPC and it is the *Capsicum frutescens* genotype which contains the highest content, the other genotypes showed lower levels but which exceed those found in this study. Similarly, Škrovankova et al. 2017 studied paprika spices (*Capsicium annuum*) from Czech, Austrian, and Slovak producers, they recorded generally considerable quantities of TPC but it was the hotter samples of paprika spices that have slightly higher values of TPC than sweet types. According (IFN et al. 2012) Similarly, polyphenols are stable to freezing (as in the case of Raspberry juice), but storage time can have an impact on the decline in these compounds.

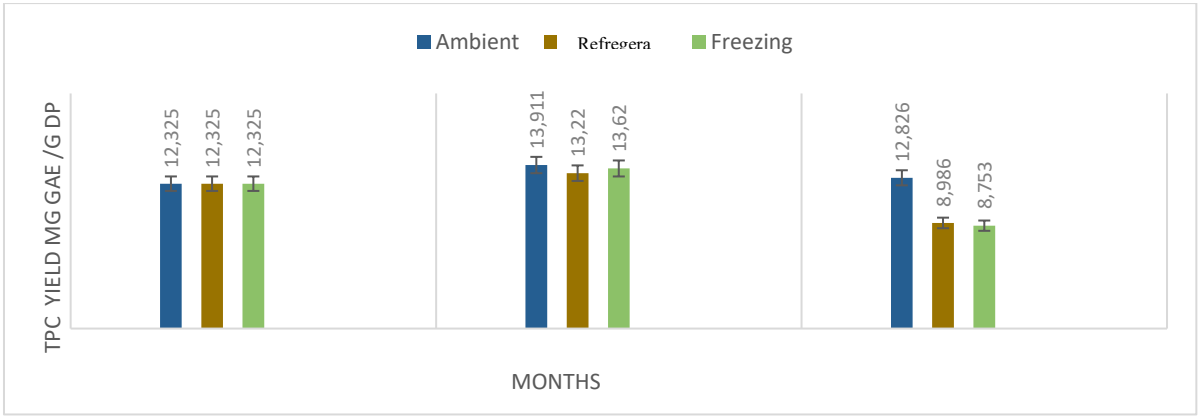

Figure 10: Change in TPC of paprika powder over the storage period at three different temperatures (Ambient temperature, Refrigeration, Freezing).

Table 14: Total phenolics contents of paprika during six months of storage at three different temperatures**.**

Evaluated parameters	Months	T° storage		
		Ambient	**Refrigeration**	**Freezing**
TPC	**1**	12.325 ±0.25^{b}	12.325 ±0.25^{b}	12.325 ±0.25^{b}
	3	13.911 ±0.56^{a}	13.220 ±0.576^{ab}	13.62 ±0.18^{ab}
	6	12.826 ±0.24^{b}	8.986 ±0.832^{c}	8.753 ±0.129^{c}

I.2. Total flavonoid content (TFC)

Figure 11 shows changes in the flavonoid content of paprika powder during storage. Flavonoid content increase in powders stored at room temperature, but falls for other types of storage (refrigeration and freezing). We note that flavonoid content rises in powders stored at -18°C during the third month of storage, showing a very different behavior to that of TPC.

TFC were affected by the mode and the time of preservation, the freezing better preserved their content after 3 months of conservation (1.39 ± 0.02 mg QE/g DP) whereas ambient temperature being the best mode which preserves them either at the 6th Months (1.29 ± 0.01 mg QE/g DP) **(Table 15).**

We observed that the contents were lower than that reported by (Kevers et al. 2007) (4.8 ± 0.5 mg QE/g), being practically stable within 6 months of storage. (Camargo, Dunoyer, and García-Zapateiro 2016) stated that cold storage conditions after 3 months significantly increased TFC, which can be attributed to changes occurring in phenols metabolism during storage. After the 6th month of storage there is no difference between refrigeration and freezing in terms of TFC. We tried to enrich the discussion of our results by comparing them to those reported by the previous works but unfortunately, the data are completely different (both the non-controlled external factors : e.g., sweet pepper varieties, agro-climatic conditions, soil compositions,

harvest time periods, among others; and the controlled conditions: extraction conditions used in the experiments).

For example, the study conducted by (Hamed et al. 2019), revealed the effect of ripening stage and cooking methods on the capsaicinoids, polyphenols and antioxidant activities of various *Capsicum annuum* cultivars. (Howard et al. 2000) reported that phytochemical including phenolic compounds and antioxidant activity of selected pepper cultivars (*Capsicum* species) depend on the type of pepper and its maturity, and also the study conducted by Oranusi et al. 2013 showed that among some imported spices in Nigeria, paprika powder contains a considerable amounts of TPC and TFC.

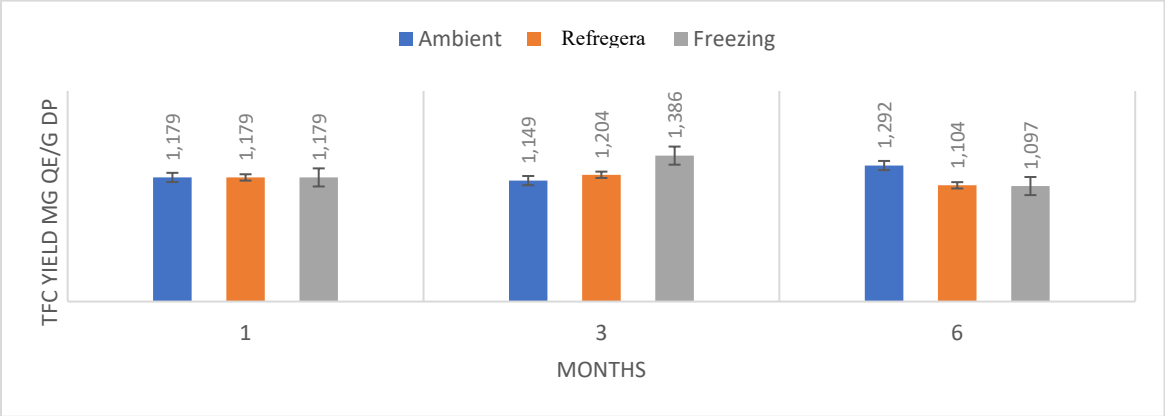

Figure 11: change in TFC of paprika powder over the storage period at three different temperatures (Ambient temperature, Refrigeration, Freezing).

Table 15: Total flavonoids content of paprika during six months of storage at three different temperatures**.**

Evaluated parameters	Months	T° storage		
		Ambient	**Réfrigération**	**freezing**
TFC	**1**	1.179 ±0.017^{c}	1.179 ±0.017^{c}	1.179 ±0.017^{c}
	3	1.149 ±0.0052^{d}	1.204 ±0.001^{c}	1.386 ± 0.015^{a}
	6	1.292 ±0.0091^{b}	1.104 ±0.0064^{e}	1.097 ±0.0169^{e}

Each value in the table is the mean ± standard deviation (n = 3)

Values sharing different letters in the same raw are significantly different ($p < 0.05$)

Results are ranked in ascending order ; a > b

II. Antioxidant activity

There are strong evidences that additive and synergistic interactions of phytochemicals present in pepper significantly strengthen the protective effects against oxidative damage thus, chillies could provide both nutritional and non-nutritional antioxidants to the human diet (Dubey et al. 2015) and (Guilherme et al. 2020).

In figure 12 the polyphenols in our powder have an excellent antioxidant effect in all three storage modes, we find that refrigeration and freezing preserve better polyphenols but ambient temperature decreases this effect with storage time; except powder stored at -18°C we, which have the greatest antioxidant power.

The antioxidant capacity of a compound is higher when its IC50 is lower, indeed the results of the present study (Table 14) revealed that paprika powder has a powerful anti-radical activity with low IC_{50} values ($3.469\pm0.099 < IC_{50} < 6.762\pm0.112$ µg/ mL) even during the storage period and all the modes used.

IC_{50} increased significantly in the powder stored at room temperature (from 4.367 ±0.12 to 6.762 ±0.112 µg/mL $p<0.01$), remains stable in the refreeze samples (from 4.367 ±0.12 to 4.659 ±0.185 and 5.327±0.885 µg /mL), and decrease in the powder stored in the freezer in the 3rd month of storage (from 4.745 ±0.016 µg/ mL to 3.367±0.12).

The refreezing mode did not affect the antioxidant activity of the stored powder during the 3rd and the 6th months (4.659 ±0.185 and 5.327 ±0.885 µg/ mL, respectively). The same trend is noticed at ambient storage at the 1st and the 3rd months (4.367 ±0.12, 4.982 ±0.06 µg /mL, respectively), by contrast, a negative effect was observed during the 6th month. Concerning the freezing mode, it did not affect the antioxidant activity in the 3rd month (4.745 ±0.016 µg/ mL), but had a positive effect in 6th month of storage (3.469 ±0.09 µg/ mL), confirming the findings

of Iqbal et al. (2015), who reported that the freezing mode preserves more the antioxidant activity of bioactive substances.

The stability of the antioxidant power of paprika during its storage may be linked to the increase in the TPC in the 3rd month regardless of the storage temperature; which is in line with the results reported in the literature (Moldovan, Popa, and David 2016).

The results of this study indicated that freezing storage of pepper powder is the best mode for preserving its antioxidant capacity (DPPH). Which confirms the results previously reported in the literature (Fernández-Pachón et al. 2006; Stratil, Klejdus, and Kubáň 2006).

Several works have been carried out in order to evaluate the antioxidant activity of pepper varieties and their products. But, as we pointed out above, the data are completely different. Therfore, the IC_{50} value of 150.40± 8.07 µg /mL and 0.03, 0.127 and 0.021mg/mg in extracts was reported for the Korian Paprika and *Capsicum chinense* varieties, respectively(Kim et al. 2011; Dubey, Singh, Upadhyay, Pandey, & Prakash, 2015). Also, the study of Hamed et al. (2019), on the scavenging activity of three variety of red pepper, reported 79%, 75%, 74% of DPPH radical-scavenging activity for red *Pueblo Chile*, red Mosco, red Fresno extracts, respectively; (Hu et al. 2021) have reported that 1mg/mL of extract of *Capsicum annuum* reduce 82% of DPPH radical and Oranusi et al. (2013) reported a reduction of 32.760±0.40% of the same radical by 25 mg/mL of paprika powder. Škrovánková et al. (2017) have reported an IC 50 values ranged from 7.35 to 17.325 mg/mL for powder of sweet paprika.

The works of (Kim et al. 2011; Hu et al. 2021) found that red paprika harvested in Korea showed the strongest antioxidant activity and 182.77 to 31.74 g/mL in a DPPH assay, so they're much more powerful than our powders; compared with Moroccan paprika (Zaki et al. 2013) , our powders showed the highest radical scavenging activity with DPPH assay IC50 of 6.762µg/ml to 3.469 µg/ml vs IC50 of 260µg/ml. Likewise Other studies have found that the drying method affects antioxidant potential (Velázquez et al. 2014). The evaluation of the antioxidant activity showed

higher DPPH radical-scavenging capacity in the smoked samples compared with oven-dried and sun-dried paprikas. The profiles of volatile phenolic compounds of the smoked samples are responsible for the differences in the antioxidant capacity.

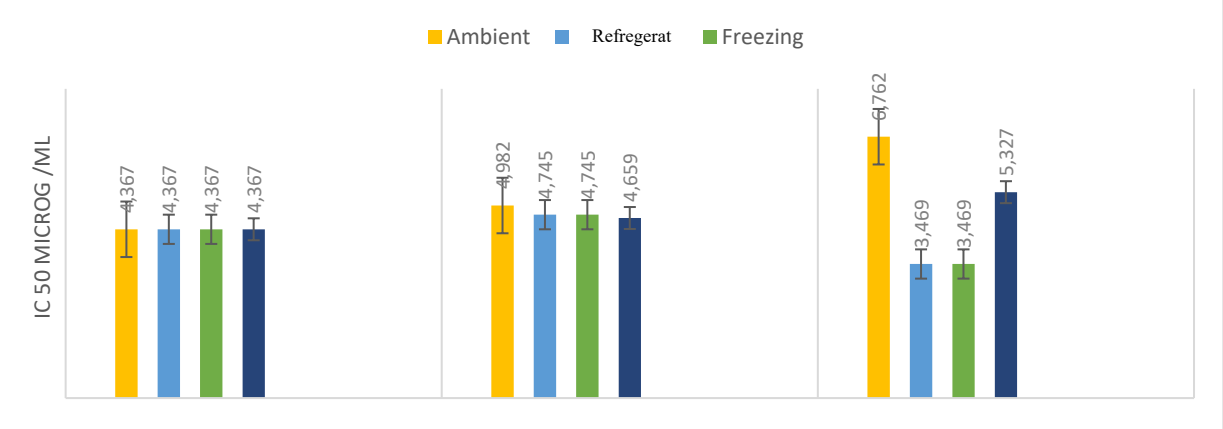

Figure 12: change in IC_{50} DPPH µg /mL of paprika powder over the storage period at three different temperatures (Ambient temperature, Refrigeration, Freezing). .

Table 15: IC_{50} DPPH of paprika during six months of storage at three different temperatures**.**

Evaluated parameters	Months	T° storage		
		Ambient	**Refrigeration**	**Freezing**
IC_{50} DPPH	**1**	4.367 ± 0.12^{bc}	4.367 ± 0.12^{bc}	4.367 ± 0.12^{bc}
	3	4.982 ± 0.06^{b}	4.659 ± 0.185^{b}	4.745 ± 0.016^{b}
	6	6.762 ± 0.112^{a}	5.327 ± 0.885^{b}	3.469 ± 0.099^{c}

Each value in the table is the mean ± standard deviation (n = 3)

Values sharing different letters in the same row are significantly different ($p < 0.05$)

Results are ranked in ascending order ; a > b

III. Antibacterial activity

III.1. Diffusion method

The results of the antibacterial activity of the phenolic extract from stored paprika powder are summarized in table 15 and figure 7. Different levels of activity can be distinguished on the basis of the diameter of the inhibition zone (DIZ: 2 < d < 3 mm indicates low activity; 4<d<5 mm, intermediate activity; 6< d < 9 mm, strong activity and finally > 9 mm means very strong activity. Paprika extract showed a good inhibitory activity and most of the extracts remains active for the entire storage period against the selected food borne pathogens (table 15). Surprisingly, Gram $^{-}$ bacteria are more sensitive to different extracts than Gram $^{+}$ one. It should be noted that, the different modes and time of storage had no effects on the activity of different extract against Gram $^{+}$ strains with a DIZ of 4.5 ±0.50 mm for *B. subtilis* and 2.5 ±0.50 mm for *S. aureus*, contrary to their recorded effect on Gram $^{-}$ which varies from 5.5 ±0.50 mm to 8.5 ±0.50 mm for *P. Aeruginosa and* 3.5 ±1.00 mm to 6 ±1.00 mm for *E. Coli*.

Indeed, *P. aeruginosa* is more sensitive to the powders stored at room temperature, however, for the extracts of powders stored at low temperature, the activity remains always strong 6mm<d<9mm, but for *E. coli* the activity decreases with the storage time, it passes from a strong activity to an intermediate activity whatever the temperature.

Capsicum annuum extracts were tested (Inés Molina et al. 2022)for their antimicrobial activity against several pathogenic microorganisms with inhibition percentages between 22 and 88% for a concentration of 100 µg/mL. These extracts were potent inhibitors of *Staphylococcus aureus* and *Pseudomonas aeruginosa*, Paprika extracts capable of controlling bacterial virulence represent a promising alternative as a natural preservative to restrict contamination or spoilage of food.

Dorantes et al. 2000 found that coumaric and cinnamic acids issues from three different chili peppers (habanero, serrano, and pimento) inhibits bacteria; also Extracts from *Capsicum annuum* fruit have been investigated by (Bacon et al. 2017) , and antimicrobial properties have

been reported. Extracts from several different *C. annuum* varieties have inhibited growth of species of *Bacillus*, *Clostridium*, *Pseudomonas*, *Listeria*, *Salmonella*, *Staphylococcus*, and *Streptococcus*, Extract from jalapeno fruit, specifically, has inhibited *Streptococcus pyogenes*, *Listeria monocytogenes*, *Clostridium sporogenes*, and *Clostridium tetani*.

Plants and herbs contain many different classes of phytochemicals These phytochemicals include terpenoids, alkaloids, lectins, polypeptides, quinones, phenolics, flavonoids, coumarins, and others (Cowan 1999). There is an abundance of research in microbiology focused on plant and their ability to inhibit spoilage and pathogenic food bacteria (Viazis et al. 2011) .

Bacon et al. 2017 found in Disk diffusion assays were performed using extractuion from jalapeño pepper (*Capsicum annuum* var. *annuum*) against *L. monocytogenes, E. coli* and *Salmonella anatum.* They found that the only bacterium with visible zones of inhibition was *L. monocytogenes*. while there is a study (Nurjanah et al. 2014) done on paprika oleoresin from chilli planted in Indonesia , they found inhibition zones between 1-3 mm on gram positive bacteria *Staphylococcus aureus*, and gram negative bacteria *Escherichia coli, Pseudomonas aeruginosa*. In addition study of (Mokhtar et al. 2017) Antimicrobial activity of pepper polyphenols (Coumarin, caffeic acid, narangin, kaempferol, rutin, quercetin) against pathogen bacteria using the disk diffusion the highest inhibition zones obtained with caffeic acid (3.5–20.5 mm), quercetin (4.75–3.5 mm), and kaempferol (7–14 mm) results that are almost in line with our results.

Table 16: Diameters of inhibition halos of different paprika extracts during storage at different temperatures

Evaluated parameters	Microbial strain		M 1	M 3			M 6		
				A	**R**	**F**	**A**	**R**	**F**
Diameters of inhibition halos (mm)	***BG+***	*B. subtilus ATCC 6633*	4.5 ±0.50^{a}	4.5 ±0.50^{a}	4.5 ±0.50^{a}	4.5 ±0.50^{a}	4.5 ±0.50^{a}	4.5 ±0.50^{a}	4.5 ±0.50^{a}
		S. aureus ATCC 25923	2.5 ±0.50^{a}	2.5 ±0.5^{a}	2.5 ±0.5^{a}	2.5 ±0.5^{a}	2.5 ±0.5^{a}	2.5 ±0.5^{a}	2.5 ±0.5^{a}
	BG-	*P. aeruginosa ATCC 27853*	8.5 ±1.00^{a}	6.5 ±0.50^{b}	6.5 ±0.50^{b}	5.5 ±0.50^{b}	8.5 ±0.50^{a}	6.5 ±0.50^{b}	6.5 ±0.50^{b}
		E. coli ATCC 25922	5.5± 1.00^{a}	6 ±1.00^{a}	2 ±1.00^{b}	4.5 ±1.00^{ab}	3.5 ±1.00^{ab}	3.5 ±1.00^{ab}	4 ±1.00^{ab}
A: Ambient, R: Refreezing and F: Freezing									

M1 first month of storage, *M3* : third month of storage and *M6* sixth month of storage. *A* : ambient, *R* refrigeration and *F* freezing

Each value in the table is the mean ± standard deviation (n = 3)

Values sharing different letters in the same row are significantly different ($p < 0.05$)

Results are ranked in ascending order ; a > b

III.2. MIC and MBC

The MIC and MBC values are shown in figure 13, the antibacterial activity of an extract is higher when its MIC and MBC are lower. The MIC values are between 1.56 and 12.5 mg/mL, the best activity was observed against *E. Coli* by the powder stored during 3 months in refreezing and freezing modes (MIC=1.56 mg/mL), these values significantly increased ($p<0.01$) at all storage processes; ambient temperature as well as refreezing and freezing modes during the 6 months (MIC = 3.12 mg/mL).

All extracts have exhibited lower activity against *B. Subtilis* with an MIC of 12.5 mg/mL, than towered other tested strains. For *S.aureus*, the best activity is exhibited by the extract from powder stored at ambient temperature during both 3 and 6months (3.12 mg/mL).

Variability in the effect of the different extracts is observed against *P. Aeruginosa* strain, the best effect (MIC= 3.12 mg/mL) is exhibited by the powder stored during 3 months at ambient and refreezing temperatures, when a moderate activity (MIC= 6.25 mg/mL) was observed during 6 months at ambient temperature and the lowest one (MIC= 12.5 mg/mL) was detected during the first month and freezing mode during 3 months.

As for MBC values, except for *E. coli* which exhibited the highest value (50 mg/mL), they are equal for all other strains (12.5 mg/mL) during all storage modes and temperatures. MIC and MBC are similar for *B. subtilis* for all storage modes and temperatures and for *P. Aeruginosa* during the 1st month and the freezing mode during 6 months.

The antibacterial activity of the extracts of *Capsicum chinense* against the tested microorganisms has been reported. Results found in the present study do not agree with those reported in the literature about the sensitivity of Gram- bacteria, indeed, these strains are usually more resistant to antibacterial agents than gram Gram+ one (Gayathri, Gopalakrishnan, and Sekar 2016; Naeim et al. 2020). Also, higher antibacterial activity was observed with acetone

and acetonitrile extract of *Capsicum chinense* against *Staphylococcus aureus,* while minimum antibacterial activity was observed in *E. coli* (Gayathri et al., 2016).

In the other hand, the antibacterial activity of the studied samples is much lower compared to that reported by X. Hu et al. (2020) on red bell pepper (paprika), who found an MIC value of 0.20±0.04, 0.50 ± 0.02, 0.30 ± 0.02, 0.60 ± 0.03 mg/mL for *Bacillus cereus*, *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas aeruginosa*, respectively. By contrast, it is much higher than reported by(Naeim et al. 2020), who found an MIC value of 25 and 12.5 mg/mL, for *Capsicum annum* fruits ethanolic extract, against *E. coli* and *S. aureus* was; respectively.

Work by (Hu et al. 2021) shows that the extracts showed an excellent antibacterial activity. The minimal inhibitory concentration (MIC) of Korean paprika was 0.20 mg mL^{-1} for *Bacillus cereus*, 0.30 mg mL^{-1} for *Escherichia coli*, 0.50 mg mL^{-1} for *Staphylococcus aureus* and 0.60 mg mL^{-1} and for *Pseudomonas aeruginosa*, values are lower than our results.

Stady of (Koffi-Nevry et al. 2012) that effect of *Capsicum annuum* and *Capsicum frutescens* (of Côte d'Ivoire) methanol and aqueous extracts were found to be effective against *Vibrio cholerae, Staphylococcus aureus,* and *Salmonella typhimurium,* The minimal inhibitory concentrations of methanol and aqueous extracts were 0.20 mg mL^{-1} and 0.25 mg mL^{-1}, respectively. Minimal bactericidal concentrations values of both extracts ranged from 1 to 2.5 mg mL^{-1} . Phytochemical assay revealed the presence of alkaloids, flavonoids, polyphenols, and sterols. Thus, *Capsicum* fruits may serve as a source of natural bactericidal agents to be used in food and medicinal systems. Mokhtar et al. 2017 found in the determination of the minimal inhibitory concentrations, The clinical strains *Staphylococcus aureus* was more sensitive to quercetin (0.00195–0.0078 mg L^{-1}) and *Pseudomonas aeruginosa* ATCC 27853 (0.0156–0.125 mg L^{-1}) so these extracts seem more effective than our own.

study of Ciulu et al. 2015 that polyphenols of Romanian Capsicum annuum extracts were also investigated for their antimicrobial was significantly higher towards Enterococcus faecalis, E.

coli and Bacillus subtilis strains and similar for Staphylococcus aureus strain. so, these biological properties indicate the potential of the obtained extracts to be used as antimicrobial agents and confirm our results.

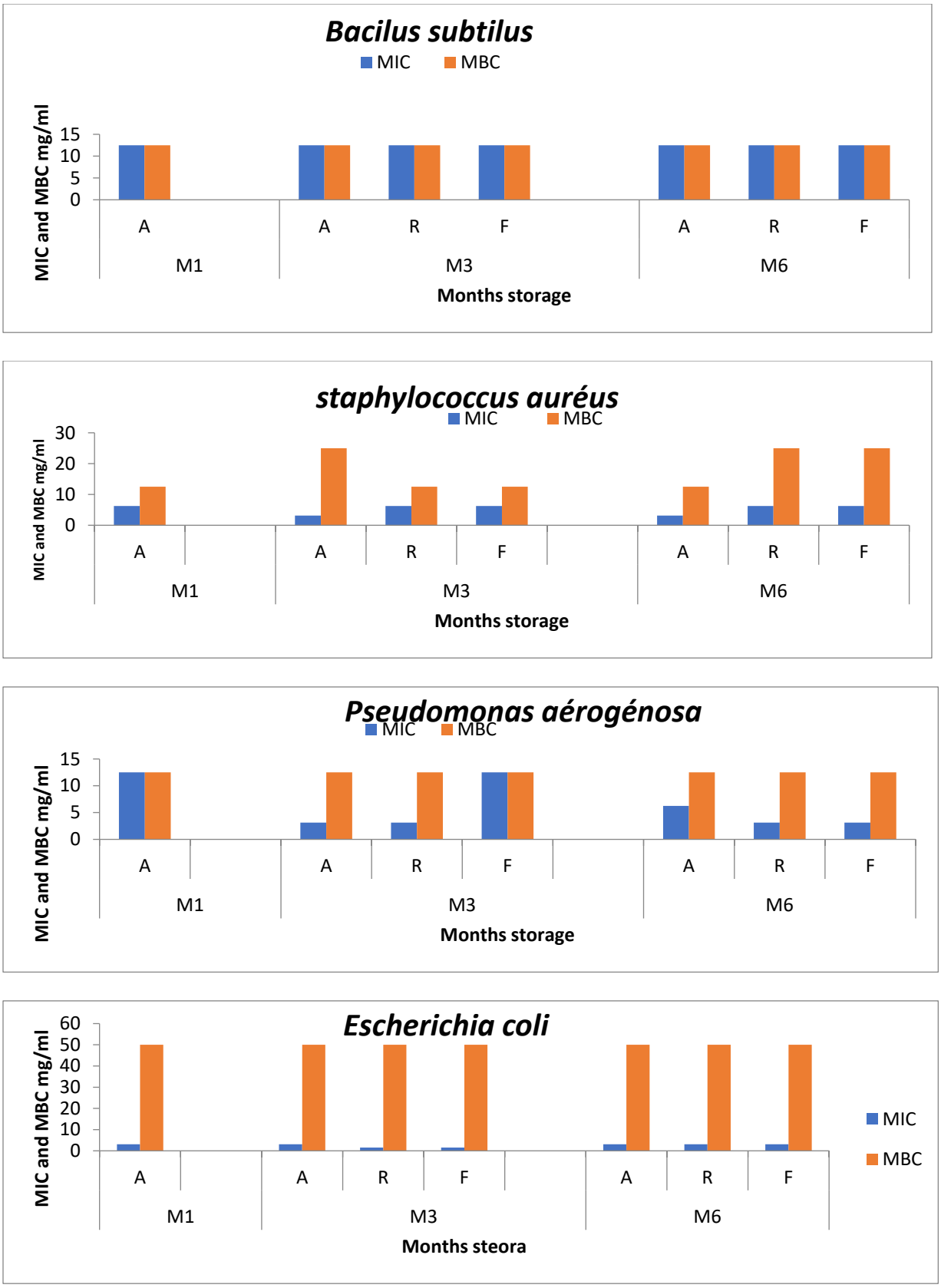

Figure13: MIC and MBC of different phenolic extract of paprika for six months of storage at different temperatures.

General conclusion and perspectives

General conclusion and perspectives

Paprika is an excellent local product from Bejaia area (Algeria), the powder obtained with basic sun-drying methods and grinding with traditional millstones; it has a high initial moisture content compared with other powders, which determines its quality and his shelf life. In addition, geographic origin influences ash content and chemical composition. Colour measurements shows that CIE-LAB *L** low luminosity index, paprika has a roch mineral content; it a good quality and contained only very small amounts of toxic elements and high level of Rb, can be used for the determination of geographical origin of paprika. It have a long shelf -life. Paprika resist against the attacks of pathogenic bacteria and it is an excellent agent that resists to the oxidation due to its phenolic compounds they have an excellent antioxidant effect in all three storage modes, and this effect increases with storage time especially for powders stored at -18°C. The use of the response surface methodology facilitated the process of traceability of the paprika phytochemical (phenolic content) stored for six months at three temperature modes. The storage of paprika in ambient temperature 25° C is able to maintain its quality in terms of phenolic compounds and antioxidants. The TPC are better preserved at ambient temperature and the TFC are more vulnerable, they are affected by both the temperature and the time of preservation, the freezing and ambient temperature being the best modes preserve them. The antibacterial activity of paprika extracts showed a good inhibitory activity and the most of the extracts remain active for the entire storage period against the selected food borne pathogens, and the strains tested, Gram– bacteria are more sensitive to different extracts than Gram+ ones. Thus, powder of *Capsicum* fruits paprika may serve as a source of natural bactericidal agents to be used in food and medicinal systems and these biological properties indicate the potential of the obtained extracts to be used as antimicrobial agents.

It is important to promote this product of local product for that we can envisage as prospects:

- Comparisons can be made with powders obtained by microwave and/or oven drying.
- Study other compounds in the powder such as vitamins, carotenoids (capsanthin) and look for possible activities.
- Assessment of colour change during storage in correlation with carotenoid content.
- Test the application of the powder as a preservative on perishable products such as charcuterie and check the various parameters physico-chemical, sensory, etc.

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Annexes

Annexe 1 :

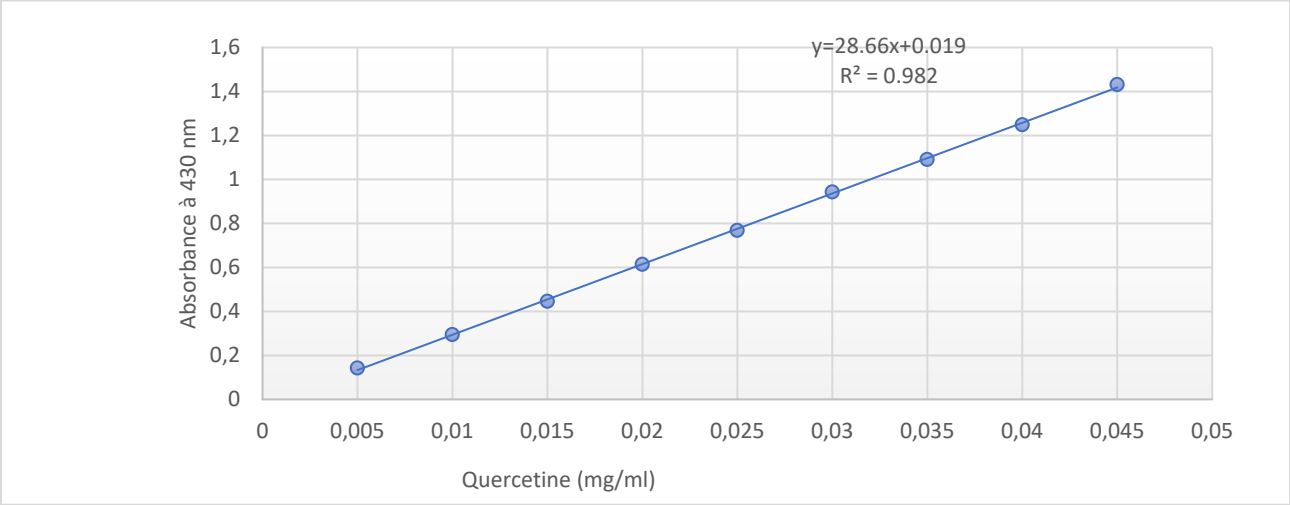

Figure 1: Calibration curve for the flavonoids assay.

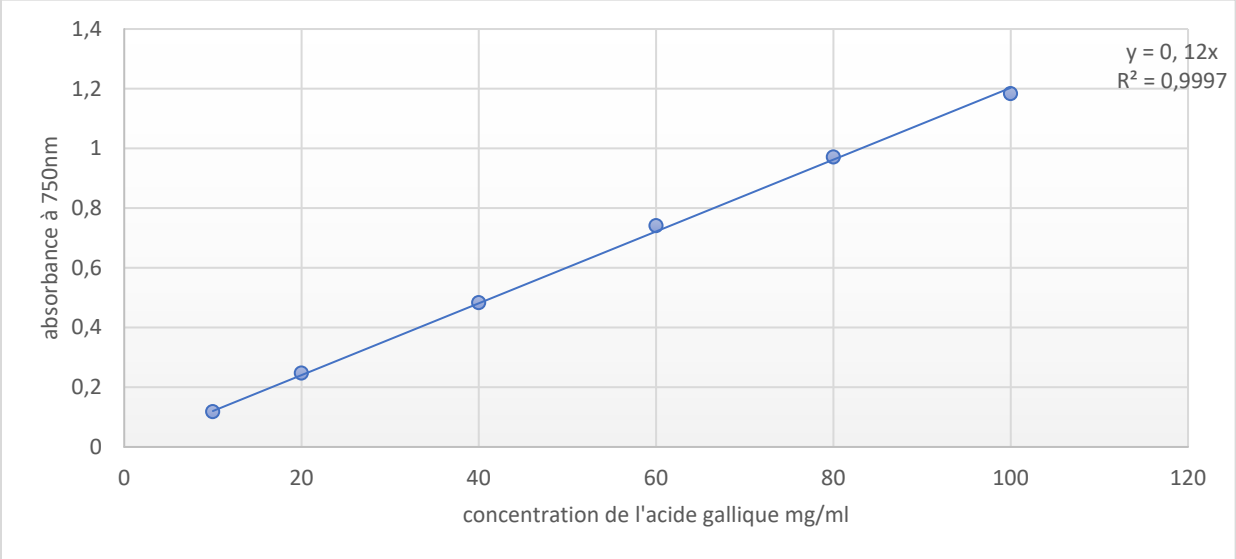

Figure 2: Calibration curve for the determination of total polyphenols

Annexe 2 :

Tableau 1: Composition des milieux préparés

Gélose nutritive

Peptone…… 10 g

Extrait de viande……. 5 g

Chlorure de sodium……. 5 g

Agar……. 15 g

pH = 7,2

Bouillon nutritif

Peptone……. 10 g

Extrait de viande…….. 5 g

Chlorure de sodium…….. 5 g

pH= 7,2

Eau physiologique

Chlorure de sodium……..... 9 g/l

Dispense into test tubes and autoclave for 20 minutes at 120°C

Abstract

The effect of different storage methods (ambient temperature 20-30 °C (A), refrigeration at 4 °C (R) and freezing at − 18 °C (F)), on the phytochemistry of an Algerian spice (paprika powder) who was prepared using traditional methods, was assessed. The optimized extract was obtained under the optimum conditions of ultrasound-assisted extraction (UAE) using response surface methodology (RSM) coupled with a Box–Behnken Design (BBD). This extract was evaluated for its total phenolics content (TPC), total flavonoids content (TFC) and its antioxidant and antibacterial activities. Under the optimum conditions (5 min for the irradiation time, 40% for the amplitude, 80% for ethanol concentration and 50% for solid–liquid ratio) the TPC was 12.23 ± 1.01 mg Gallic Acid Equivalent/gram of Dried Powder (mg GAE/g DP) which is very close with experimental assay. the freezing and ambient temperature being the best modes which preserve the TPC and TFC. Paprika phenolic compounds have an excellent antioxidant effect in all three storage temperatures, the antibacterial activity of our extracts remains excellent throughout the storage period, with changes in the inhibitory effect and inhibitory and lethal doses depending on the strains tested and the storage temperature.

Key words : Paprika, Storage, Phenolic compounds, UAE, Box–Behnken design, Biological activities.

Résumé

L'effet de différentes méthodes de stockage (température ambiante 20-30 °C (A), réfrigération à 4 °C (R) et congélation à - 18 °C (F)), sur la phytochimie d'une épice algérienne (poudre de paprika) qui a été préparée selon des méthodes traditionnelles, a été évalué. L'extrait optimisé a été obtenu dans les conditions optimales d'extraction avec utilisation de l'extraction assistée par ultrasons (EAU) en utilisant la méthodologie de la surface de réponse (RSM) couplée à un plan de Box-Behnken (BBD). Cet extrait a été évalué pendant toutes la période de stockage pour sa teneur en composés phénoliques totaux (TPC), sa teneur en flavonoïdes totaux (TFC) et ses activités antioxydantes et antibactériennes. Dans les conditions optimales (5 min pour le temps d'irradiation, 40% pour l'amplitude, 80% pour la concentration en éthanol et 50% pour le rapport solide-liquide), le TPC était de 12,23 ± 1,01 mg d'acide gallique équivalent/gramme de poudre séchée (mg GAE/g DP), ce qui est très proche de l'essai expérimental. La congélation et la température ambiante sont les meilleures températures de conservation du TPC et du TFC. Les composés phénoliques du paprika ont un excellent effet antioxydant aux trois températures de stockage. L'activité antibactérienne de nos extraits reste excellente tout au long de la période de stockage, avec des changements dans l'effet inhibiteur et les doses inhibitrices et létales en fonction des souches testées et la température de stockage.

Mots clés : Paprika, Stockage, Composés phénoliques, EAU, Box-Behnken design, Activités biologiques.

الملخص

تم تقييم تأثير طرق التخزين المختلفة (درجة حرارة الغرفة(A) 20-30 درجة مئوية، والتبريد (R) عند 4 درجات مئوية، والتجميد (F)عند -18 درجة مئوية، على الكيمياء النباتية لتوابل جزائرية (مسحوق الفلفل الحلو) التي تم تحضيرها بالطرق التقليدية. تم الحصول على المستخلص الأمثل في ظل ظروف الاستخلاص المثلى باستخدام الموجات فوق الصوتية (UAE)باستخدام منهجية سطح الاستجابة (RSM) إلى جانب تصميم (Box-Behnken (BBD). تم تقييم هذا المستخلص من حيث محتواه الكلي من المركب الفينولي (TPC)، ومحتوى الفلافونويد الكلي (TFC) والأنشطة المضادة للأكسدة والمضادة للبكتيريا. في ظل الظروف المثلى (5 دقائق لوقت الإشعاع، و40% للسعة و80% لتركيز الإيثانول و50% لنسبة الصلب إلى السائل)، كان إجمالي مركب الفينول الكلي 12.23± 1.01 (مغ من حمض الغاليك/غرام من المسحوق المجفف)، وهو قريب جدًا من الفحص التجريبي. ويُعد التجميد ودرجة حرارة الغرفة أفضل الأوضاع لحفظ TPC و TFC. تتمتع مركبات الفينول البابريكا بتأثير ممتاز مضاد للأكسدة في جميع درجات حرارة التخزين. ويظل النشاط المضاد للبكتيريا لمستخلصاتنا ممتازًا طوال فترة التخزين، مع تغيرات في التأثير المثبط والجرعات المثبطة والمميتة اعتمادًا على السلالات المختبرة وطرق التخزين.

الكلمات المفتاحية : بابريكا، تخزين، مركبات فينولية، الاستخلاص بالموجات فوق الصوتية ، تصميم بوكس-بهنكن، أنشطة بيولوجية.