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**Physio-chemical study of a medicinal plant of the Aster family (Chamomile) cultivated in the Algerian desert**

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In front of the jury composed of:

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## **Dedication**

**I give this research to every science student seeking knowledge and providing his scientific and cultural wealth.**

**Who did you support in her prayers and invite her to the light of night that lights my way**

**For those who share my joys and my faith, to the fountain of compassion and compassion, to the most beautiful smile of my life, my dear mother, a fine soldier, djendli kheira.**

**Who taught me the struggling world, and who is good is science**

**To him who did nothing to me, to him who sought my rest and success, to the greatest man in the universe, my dear father, Dgaloul haoued mouissa.**

**Who stood with me in my college studies and consultations to my partner in my life and to my dear husband Abdulkhak and his family**

**To all the generous family who supported me and are still among the brothers Ahmed and Abdul Jalil**

**And my sisters Al-Zahra, Zubaida, Hafsa, Nawal, Iman, Awatif, and especially my sister-in-law Souad And to the buds of the family, Malak, Israa, Aliaa, Rabee, Abdullah, Arwa, Anas, Khadija, Muhammad Othman, Taha, Farouk, Abdel Nour**

**We dedicate this work to the chemistry department, the college administration, and to each class of 2023**

**And to the one who accompanied us throughout this research and provided us with valuable information and advice Framed Professor, Mekhlfi Tarek**

**AIDA**

## **Dedication**

**To my support and my safe refuge, my supporter  
And my constant fans when they call me by his name  
I am happier and prouder that I am his daughter and his fruit  
Who saw the reflection of my success and joy sparkle in his  
eyes**

**Here's to you, my dear father**

**My friend, my hope, my hero, and my first teacher  
Whose supplication and satisfaction were my compass on the  
path**

**Who taught me the meaning of love and kindness, the  
meaning of patience, strength and love**

**My beautiful mother**

**To those who were and still are my support in life and my  
refuge my brother and the joy of the house is the descendants  
of the family, each in his name**

**To whom I spent the most beautiful and best hours with, my  
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**Basma, Maryam, Walid, Yasser Iqbal, Randa, Taqiyah, Hadil,  
Amanne**

**And my neighbors are in the residence wing**

**Faiza**

## **Irfan**

**Praise and thanks be to God Almighty for the blessing of Islam**

**And prayers and peace be upon our master Muhammad, may**

**God bless him and grant him peace**

**We thank God who made it easy for us to complete this message**

**We also extend our sincere thanks to Prof. Dr. Zainab Rahmani**

**And Dr. Zohoor Rahmani and Professor Balfar Muhammad Al-**

**Akhdar**

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**Maryam Habibi, Asmaa Nassib, Abdul-Jabbar Masoudi**

**And the whole VRS Detective crew**

**Faiza & Aida**

## Résumé :

Dans cette étude, nous avons d'abord traité de la culture de la plante de camomille, qui appartient à la famille Asteracea et nous avons fait un certain nombre d'analyses à ce sujet, y compris des tests d'extraction et phytochimiques, et nous avons constaté qu'il existe les mêmes composés étudiés auparavant, nous avons ensuite estimé la quantité de polyphénols, qui variait entre(  $0.031 \pm 0.014$ ) a(  $1.05 \pm 0.64$ ) dans les fleurs Quant aux papiers, ils étaient entre(  $0.047 \pm 0.036$ ) a(  $1.277 \pm 0.395$ ) , Quant à l'estimation de la quantité de flavonoïdes, leur quantité variait entre(  $0.102 \pm 0.064$ ) a(  $1.197 \pm 0.49$ ) dans les fleurs, et (  $0.032 \pm 0.017$ ) a(  $1.082 \pm 0.248$ ) .

Où nous avons remarqué que les valeurs de quantification étaient très similaires pour les fleurs et les feuilles

Nous avons même étudié la capacité antioxydante du molybdate en utilisant la courbe standard de l'acide ascorbique, et nous avons trouvé une différence par rapport aux valeurs où dans les fleurs il était plus élevé et TAC = 0.4 par rapport au butanol, mais dans les fleurs l'acétate était la valeur la plus élevée TAC = 0.949

De plus, nous l'avons étudié DPPH à travers l'absorption et le pourcentage d'inhibition, et nous avons constaté que notre plante a l'efficacité de la capacité antioxydante, car le pourcentage d'inhibition dans tous les extraits augmentait avec l'augmentation des concentrations

Grâce à IC50, nous avons remarqué que pour les feuilles, le butanol avait le taux d'inhibition le plus élevé, alors que pour les fleurs, c'était pour l'acétate.

Enfin, nous avons calculé le rendement d'extraction, et le butanol avait le pourcentage le plus élevé dans les feuilles et les fleurs.

## Summary :

In this study, we first dealt with the cultivation of the chamomile plant, which belongs to the Asteracea family, and we did a number of analyzes on it, including extraction tests and phytochemicals, and we found that there are the same compounds studied before, we then estimated the amount of polyphenols, which varied between(  $0.031 \pm 0.014$ ) a(  $1.05 \pm 0.64$ ) in the flowers As for the papers, they were between (  $0.047 \pm 0.036$ ) a(  $1.277 \pm 0.395$ ) , As for the estimate of the amount of flavonoids, their amount varied between(  $0.102 \pm 0.064$ ) a(  $1.197 \pm 0.49$ ) in the flowers, and (  $0.032 \pm 0.017$ ) a(  $1.082 \pm 0.248$ ) .

Where we noticed that the quantization values were very similar for flowers and leaves

We even studied the antioxidant capacity of molybdate using the standard curve of ascorbic acid, and we found a difference compared to the values where in the flowers it was higher and TAC =0.4 compared to butanol, but in the flowers acetate was the highest value TAC = 0.949

Moreover, we studied it DPPH through absorption and percentage inhibition, and we found that our plant has the effectiveness of antioxidant capacity, because the percentage inhibition in all extracts increased with the increased concentrations

Using IC50, we noticed that for leaves, butanol had the highest inhibition rate, while for flowers, it was for acetate.

Finally, we calculated the extraction yield, and butanol had the highest percentage in leaves and flowers.

## الملخص:

في هذه الدراسة تناولنا أولاً زراعة نبات البابونج الذي ينتمي إلى عائلة Asteraceae، وقمنا بإجراء عدد من التحليلات عليه، بما في ذلك اختبارات الاستخلاص والمواد الكيميائية النباتية، ووجدنا أن هناك نفس المركبات التي تمت دراستها من قبل، ثم قمنا بتقدير كمية البوليفينول والتي تراوحت بين  $(0.031 \pm 0.014)$  أ  $(0.64 \pm 1.05)$  في الأزهار أما بالنسبة للأوراق فقد تراوحت بين  $(0.036 \pm 0.047)$  أ  $(0.014 \pm 0.014)$ ، أما بالنسبة لتقدير كمية مركبات الفلافونويد، تفاوتت كميتها بين  $(0.064 \pm 0.102)$  أ  $(0.49 \pm 1.197)$  في الأزهار، و  $(0.017 \pm 0.032)$  أ  $(0.248 \pm 1.082)$ . حيث لاحظنا أن قيم التكميم كانت متشابهة جداً للأزهار والأوراق لقد درسنا أيضاً قدرة موليبيدات المضادة للأكسدة باستخدام المنحنى القياسي لحمض الأسكوربيك، ووجدنا فرقاً مقارنة بالقيم التي كانت أعلى في الأزهار و TAC = 0.4 مقارنة بالبيوتانول، ولكن في الأزهار كانت أسيتات أعلى قيمة TAC = 0.949 علاوة على ذلك، قمنا بدراسته DPPH من خلال الامتصاص والتثبيط المئوي، ووجدنا أن نباتنا لديه فعالية في القدرة المضادة للأكسدة، لأن النسبة المئوية للتثبيط في جميع الخلاصات تزداد مع زيادة التراكيز. باستخدام IC50، لاحظنا أنه بالنسبة للأوراق، كان للبيوتانول أعلى معدل تثبيط، بينما بالنسبة للزهور، كان للأسيتات. أخيراً، قمنا بحساب محصول الاستخلاص، وكان للبيوتانول أعلى نسبة في الأوراق والزهور.

## List des contents

Dedication .....	I
I give this research to every science student seeking knowledge and providing his scientific and cultural wealth.....	I
Irfan .....	III
Résumé :.....	IV
Summary : .....	V
List des contents.....	VII
List of tables:.....	XIII
List of figures: .....	XIV
List of abbreviation: .....	XVI
Chapter (I): .....	4
<b>Medicinal and aromatic plants .....</b>	<b>4</b>
I.1. Entrance: .....	5
I.2. Brief-history:.....	5
I.3. History of Medicinal plants in Algeria .....	6
I.4. Definition of medicinal plants .....	9
I.5. Definition of Aromatic Plants .....	10
I.6. Sources of medicinal and aromatic plants .....	10
I.7. Multiplying medicinal and aromatic plants .....	11
A) Seed propagation: .....	11
B) Green propagation:.....	12
I.8. Stages and dates of collecting plant parts .....	12
I.8.1. Collection of Medicinal Plants: .....	12
I.8.2. Timing of Plant Collection: .....	12
1.8.4. Process of Plant Collection: .....	13
I.9. Plant parts are collected as follows: .....	13



I.9.1. Roots and Rhizomes: .....	13
I.9.1. Bulbs: .....	14
I.9.2. Tubers: .....	14
I.9.3. Bark: .....	14
I.9.4. Wood: .....	14
I.9.5. Leaves and Herbaceous Stems: .....	14
I.10. Drying : .....	15
I.11. Natural drying methods .....	15
I.12. Dehydration .....	15
I.13. preservation: .....	16
I.14. The most important areas of use of medicinal and aromatic plants: ..	16
References .....	18
Chapter (II):.....	20
Medicinal and aromatic plants commercially and economically .....	20
II.1. entrance : .....	21
<b>II.2. The economic importance of medicinal and aromatic plants commercially and economically :</b> .....	21
II.3 . Brief General on Cultivator the plants medical and aromatic in north Africa: .....	21
II.4. Examples of products made from thyme oil : .....	22
Sweet Orange Deodorant .....	22
Lip Balm .....	22
Bath Salts or Soaks .....	23
Bath Bombs.....	23
Lotion Recipes .....	23
Tea.....	23
Soap.....	24
Rose Cleansing Grains .....	24

Rose Lip Balm .....	24
II.5. Conditions for exporting medicinal and aromatic plants:.....	24
REFERONCE .....	26
Chapter (III): .....	27
Common Chamomile .....	27
III.1. entrance .....	28
III.2 . Plants of the asteraceae family .....	28
III.2.1. General classification:.....	28
III .2.2. General characteristics characteristic of the labial family :29	
III.3. CHAMOMILE:.....	29
III.3.a. Definition of chamomile: .....	29
III.3.b. Description of chamomile:.....	30
III.3.c. The history of chamomile: .....	30
III.3.d. Botanical classification Chamomile: .....	31
III.4. Geographical distribution of Chamomile plant in the world:.....	32
III. 5. Chamomile cultivation climate:.....	32
References:.....	34
Chapter (IV):.....	36
Compounds of Secondary Metabolites .....	36
IV.1. Entrance : .....	37
IV.2. Définition of secondary metabolites.....	37
IV.3. The alkaloids .....	38
IV.3.1. Definition of alkaloids :.....	38
IV.3.2. Classification of alkaloids .....	38
IV.3.2.a. True alkaloids : .....	38
IV.3.2.b. Primary alkaloids :.....	38
IV.3.2.c. Pseudo alkaloids : .....	38

IV.3.3. DETECTION OF ALKALOIDS :.....	41
IV.3.4. The medical importance of alkaloids : .....	41
IV.4. Flavonoids.....	41
IV.4.1. Define it : .....	41
IV.4.2. Classification of flavonoids :.....	42
IV.4.2.a. Flavanols,flavan-3-ols or catechins : .....	42
IV.4.2.b. Flavanones :.....	42
IV.4.2.c. Isoflavanoids :.....	42
IV.4.2.d. neoflavonoids : .....	43
IV.4.2.e. Anthocyanins : .....	43
IV.4.2.f.Chalcones : .....	43
IV.4.3. Properties of flavonoids :.....	44
IV.4.4.Flavonoids in the matricaria chamomilla : .....	45
IV.4.5.Detecting of flavonoids : .....	46
IV.5. Coumarins:.....	46
IV.5.1. Define it : .....	46
IV.5.2. Detecting of coumarins :.....	47
IV.6. Phenolic acids :.....	47
IV.6.1. Define it : .....	47
IV.6.2. Biological and therapeutic properties of phenolic acids : .....	48
IV.6.3. Phenolic acid in the Matricaria chamomilla :.....	48
IV.7. The terpenoids : .....	49
IV.7. 1. Define it : .....	49
IV.7. 2.Classification of terpenoids :.....	49
IV.7. 3.Detecting of terpenoids :.....	50
IV.8. glycosides : .....	50
IV.8.1. define it :.....	50

IV.8.2. detecting of glycosides : .....	50
IV.9. saponins : .....	50
IV.9. 1. define it : .....	50
IV.9. 2.detecting of saponins : .....	51
IV.10. Resins : .....	51
IV.10.1. Define it .....	51
IV.10.2. SOME RESINS : .....	51
IV.11. Volatile oils.....	52
IV.11.1. Detecting of volatile oils : .....	52
IV.11.2. Thin layer chromatography (TLC) : .....	52
IV.11.3. Extraction : .....	53
IV.12. Essential oils : .....	53
IV.12.1. Define of essential oils : .....	53
IV.12.2. Locations of concentration of essential oils : .....	53
IV.12.3. Methods of extracting essential oils : .....	54
IV.12.4.Method of extraction with volatile organic solvents : .....	56
IV.12.5. Chemical composition of essential oils : .....	58
IV.12.6. Physical and chemical properties of essential oils : .....	59
IV.12.7. Benefits and uses of essential oils : .....	61
IV.12.8. Benefits of essential oil in Matricaria chamomilla : .....	61
References : .....	62
V.PRACTICAL PART .....	64
V.1.ENTRANCE .....	<b>Erreur ! Signet non défini.</b>
V.2.Ways and means:.....	65
V.3. Plant material: .....	65
V.3.1.Sample drying: .....	65
V.3.2.Soaking: .....	65

V.3.3.Phytochemical examination:.....	68
V.3.4.Essential oil extraction:.....	71
V.4.Polyphenol Dosage: .....	72
V.4.1.Principle : .....	72
V.4.2.We Avon prepare: .....	73
V.5.Estimation of total polyphenols intake TPC :.....	74
V.5.1.Flavonoids Dosage:.....	76
V.5.2.Principle .....	76
V.5.3.We have prepared: .....	77
V.6.Estimation of flavonoid intake TFC: .....	77
V.7.Determination of total antioxidant capacity: .....	80
V.7.1.We prepared: .....	80
V.7.2.Chloroform.....	<b>Erreur ! Signet non défini.</b>
V.7.3.Butanol .....	<b>Erreur ! Signet non défini.</b>
V.7.4.Acetat .....	<b>Erreur ! Signet non défini.</b>
V.7.5.Alternator return test : .....	81
V.8.Standard curve for DPPH: .....	84
V.9. Oil analysis by GC/MS : .....	89
REFERENCE.....	94
Conclusion .....	96

## List of tables:

Table	Title of tables	Page
1	The percentage of moisture of plant parts after drying	16
2	Examples of products made from thyme oil	22
3	selected examples of each groups of Alkloids	37
4	Flavonoids in the Matricaria chamomilla	42
5	Phenolic acid in the Matricaria chamomilla	44
6	Classification of terpenoids	45
7	SOME RESINS	50
8	chemical composition of essential oil of Matricaria chamomilla from Nepal	53
9	chemical composition of chamomile flowers essential oil from	54
10	Matriels used	64
11	weight after drying of flowers and leaves(g)	66
12	tests Phytochimiques	67
13	Extraction yield	69
14	Series volumes taken in the quantification of flavonoids	72
15	DOSAGE of polyphenol ( flowers) (c mg /g)	74
16	DOSAGE of polyphenol ( leaves) (c mg /g)	75
17	DOSAGE of flavonoids (flowers) (c mg /g)	78
18	Dosage of flavonoid(leaves) (c mg /g)	78
19	MOLYBDATE FLOWER TAC	82
20	MOLYBDATE LEAVES TAC	83
21	IC50 of leaves	87
22	IC50 of flowers	88
23	device of GC /MS	89 -90- 91

## List of figures:

	Title of figures	Page
1	CHAMOMILE PLANT	27
2	GEOGRAPHICAL DISTRIBUTION	28
3	The basic unit of coumarin	39
4	Ascorbic acid	40
5	Gallic acid	40
6	Method of Hydrodistillation	46
7	Method of extraction with volatile organic solvents	47
8	a schematic illustrating the experimental work in the laboratory	65
9	picture of rotavap	66
10	Extraction yield of flowers	69
11	Extraction yield of leaves	69
12	Extraction yield of leaves and flower	69
13	Laboratory image	71
14	the Clevenger apparatus	71
15	Standard curve for Galic acid	73
16	Dosage of Polyphenol ( flowers) (c mg /g)	74
17	Dosage of Polyphenol ( leaves) (c mg /g)	74
18	Standard curve for Quercitine	77
19	DOSAGE of FLAVONOIDS (flowers) (c mg /g )	77
20	Dosage Of flavonoid(leaves) (c mg /g)	78
21	Molybdate preparation	79
22	A picture of the prepared series of molybdate after reading it	80
23	Standard curve for ascorbic acid	80
24	Molybdet flower	81

25	Molybdate leaves	81
26	Molybdate flower TAC	82
27	Molybdate leaves TAC	83
28	Root oxidation equation	84
29	DPPH of flower	84
30	DPPH of leaves	85
31	DPPH flowers with ascorbic acid	85
32	DPPH of leaves with ascorbic acid	86
33	IC50 of leaves	86
34	IC50 of flowers	87
35	device of GC /MS	87
36	Result of GC /MS	89



## **List of abbreviation:**

**Abs:** Absorbance

**AEAC:** Ascorbic acid equivalent antioxidant capacity

**CI<sub>50</sub>:** Concentration at inhibition of 50%

**DPPH:**  $\alpha, \alpha$ -diphenyl- $\beta$ -picrylhydrazyl

**UV:** ultra violet

**GC:** chromatographe of gas

**TFC:** total flavonoids

**TPC:** total phenolic compounds

**mg EAG/g:** milligram of equivalent of galic acid frochem g of extract of matiere se

**%:** percentage

**(-):** negative,

**(+):** positif, present

**C:** concentration

# *General introduction*

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The human interest in medical web site begins to pretend the human behavior and directs the same nature by sealing his interests, and diseases created by god and the human life and their awareness went through many evolutionary stages that included all aspects of his needs, including medicinal and aromatic plants. It is known that the medicines currently used in the world to treat many diseases are mostly due to plant Origin, and the percentage of that reaches 60%. In addition to that, other natural sources constitute a good percentage of the use in treatment. Through his experiments, man began to retrace these active elements, purify them, determine their atomic weight and chemical composition, and manufacture similar ones, but the results from these absolute substances were not as effective as those given by natural drugs, and therefore it proved beyond any doubt that man is unable to produce drugs similar in composition and effect as well produced by natural plants.

The flow of the history of drugs and herbal medicine notes that the number of known medicinal plants is very large in the world, but the benefit that man has obtained so far is still below the required minimum limits from these plants, so the need imposed more known and greater attention to medicinal plants, which comes through in-depth studies that through which the basic goal of these plants is achieved.

The use of plants in pharmaceutical preparations depends mainly on chemical analyses of these plants. These analyses are often very complex. In addition, all experiments for the extracted materials are carried out on animals and on cellular samples of different bacterial materials and molds. As a result, the medical prescription consists of dried plants, processed and prepared to suit the description, or extracted from basic materials of pure plant origin, taken directly or in the form of a simple preparation as a treatment, or to be prepared and added by the pharmaceutical industry and its laboratories. In the past, medicines were prepared and produced in small quantities, manufactured and marketed in the form of pills, paste or emulsion. Sometimes this is done in each pharmacy separately and personally, but now the procedure is done by isolating the active substance first with mentioning the percentages of its presence in the plant and verifying in the pharmacopoeia and the method of using the drug (external-internal), the amount used in children and the elderly. All medicinal treatments allowed to be circulated in countries of the world.

This is what prompted us in the course of our research to contribute to the integration of plant wealth and to study a plant that belongs to the Compositae (Asteraceae), chamomile is an

annual herbaceous plant that reaches 50-60 cm in height, with many branching branches, its leaves are lobed, feathery and its branches bear whorls of disc-yellow flowers; it is distributed on a hollow bed, and is surrounded from the outside by one or two rows of white radial flowers. The plant is grown in order to obtain its inflorescences which give off the scent of apples, so it was called earth apple, the original home of chamomile is southern and eastern Europe and from there it spread to nearby areas, in the whole sea basin. Mediterranean in the south America and the USA its cultivation has spread in Egypt for a long time and Germany, Hungary, Yugoslavia and Russia produce large quantities of it, this is through previous studies that show various secondary metabolic compounds as well the biological effectiveness of chamomile. The axes of our study were divided as follows:

- **The first chapter** : medicinal and aromatic plants and their economic importance.
- **Chapter two** : Medicinal and aromatic plants commercially and economically
- **Chapter three** : Common Chamomile
- **Chapter four** : Secondary metabolite
- **Chapter five** : practical part

# **Chapter (I):**

## **Medicinal and aromatic plants**

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## **I.1. Entrance:**

Since humans existed on Earth, they turned to the plants surrounding them to obtain the food necessary for their survival. When they needed shelter to protect themselves from the weather and predators, they sought refuge in shrubs or trees, using them for their protection. When they fell ill, they tried to use certain plants for treatment, while others were used for beauty and adornment due to their beautiful shapes and fragrances. When they wanted to mature their food or repel predators, they lit fires from plants, and they used plants as clothing to protect themselves from heat and cold. The plant kingdom contains many plant families that include a large number of plants with medicinal properties known as medicinal and aromatic plants. It also includes plants that contain essential oils known as aromatic plants. These medicinal and aromatic plants have been important since ancient times and still retain their status in all civilizations. The ancient Egyptians, Indians, Chinese, Persians, Japanese, African tribes, Gypsies in Europe, Native Americans in the Americas, and Australian Aboriginals all knew about these plants and used them for treatment, food flavoring, and cosmetics.

## **I.2. Brief-history:**

Medicinal plants have been discovered since prehistoric times and used in traditional medicine. Hundreds of chemical components can be synthesized from plants to be used to control insects, fungi, diseases, and herbivorous mammals. Many plant chemicals with proven or potential biological activity were known, but the possession of a single plant for a large number of diverse chemicals made the effect of using the whole plant less effective. And prevented the evaluation of the activities related to these substances

present in many plants in accurate scientific research aimed at deterring their effectiveness and safety. [1]

Medicinal plants, were mentioned for the first time historically in the Sumerian civilization, as hundreds of medicinal plants, including opium, were listed on clay tablets in the thirtieth century BC. The Ebers papyrus in ancient Egypt around 1550 BC described more than 850 medicinal plants. The Greek physician Dioscorides, who served in the Roman army, documented more than 1000 prescriptions based on more than 600 medicinal plants in the book of the five Articles and by AD 60, three books formed the basis of the pharmacopoeia for nearly 1500 years. [2]

Pharmaceutical research relied on popular botanical science to discover pharmacologically effective plants and this led to the discovery of hundreds of beneficial ingredients that included aspirin, digoxin, quinine, and opium, these compounds are found in different types of plants, but in most of them belong to four main biochemical classes: alkaloids, glycosides, polyphenols, and terpenoids. [3]

Medicinal plants are widely used in non-industrial societies as they are more readily available and cheaper than modern medicine. The global annual export value of this and of plant species having medicinal properties was estimated at 2.2 billion in 2012, and in 2017, the value of the global market for medicines and plant extracts reached hundreds of billions of dollars. Many countries apply some kind of regulation to traditional medicine, but the World Health Organization organizes a network that encourages the safe and risk factors of climate change and environmental destruction, and qualitative factors such as excessive collection to meet market needs. [4]

### **I.3. History of Medicinal plants in Algeria**

Nassim Jabou, who is also an international expert in agriculture and nutrition at the United Nations Agriculture Organization, adds that medicinal and aromatic

oils received special care after independence, as they are specially destined for export and have an important position at the global level. A strategy was drawn up to develop this manufacturing industry, by valuing what the state inherited from colonialism .And the construction of large industrial trains, such as those in the city of "Sebdo" in Tlemcen, which are dedicated to lavender oil, and which were used to produce one of the most luxurious perfumes in the world at the time, with an Algerian brand called "Al Hoggar" perfumes

On February 24, 1971, the late President Houari Boumediene announced the nationalization of hydrocarbons, a decision that resulted in the imposition of an undeclared commercial embargo by France and its allies, who boycotted Algeria's products of medicinal and aromatic oils, and oil exports declined dramatically, which precipitated the decision to stop these The manufacturing industry, and the allocation of most of the lands that were exploited in this division, for food cultivation, after the new directions at the time that gave priority to food production, and yet some large trains remained operating, such as the Jasmine factory in Al-Shafah in the state of Blida, but in decent quantities, so that Algeria lost its position, in the international market This was the first break with the development of agriculture directed to the manufacturing industry ,according to Professor Nassim Gabo. As for the second break, it occurred during the national tragedy, when the security conditions during that period prevented many from harvesting these wild plants in the forests, so the remaining distilleries were closed, thus announcing the end of the story. Production of medicinal and essential oils in Algeria

Experiences and studies indicate that the benefits generated by the manufacturing industry of medicinal and aromatic oils are tens of times greater than the profits reaped when they are marketed as raw materials. In the field of conversion, the latter imports the raw material , converts it and then exports it in the form of oils, thus exceeding one billion dollars annually from these



conversion operations. The same is the case for France, which allocates an area of 40,000 hectares only for the cultivation of medicinal and aromatic plants and extracted oils, but its good use of this area in a professional and dberatemannereli brings it more than 400 million euro in profits from the raw material, which it does not export but rather exploits in the manufacturing industry, where it is involved in the production of many products and preparations, the value of its exports is estimated at 34 billion euro from the revenues of finished products that are exported around the world

At the regional level, the Arab countries in North Africa outperformed Algeria in the production of essential oils, despite benefiting from the country's expertise in this cultivation. However, its interest in this field qualified it to be among the largest producers in the world, such as Egypt. Its production is not limited to medicinal and aromatic oils only. Rather, this extends to dried plants, as Egypt is one of the largest producing countries, and it was able to extract the fame of the "Algerian thirst" label, thus becoming the most famous and popular "Egyptian thirst" in the international market Professor Nassim Jabou believes that Algeria, at the current stage, is interested in producing everything that is a raw material, and at the same time it is working to develop manufacturing industries, especially since we are witnessing activity and movement at the national level in the field of perfumes and cosmetics, which imports all its raw materials from abroad, knowing That we can provide some of them, and contribute to the government's action plan, which aims to reduce the import bill Jabou explains that some institutions have started acquiring the raw material from some companies that produce the raw material locally, such as the "Olésens" company in Constantine ,which specializes in the production of cosmetics ,and which used to import all its raw materials from abroad, and now it acquires several raw, materials from Algeria with high quality, higher rand<sup>[7]</sup>

## **I.4. Definition of medicinal plants**

The world-renowned botanist Dragendorff defined any plant of vegetal origin used for medicinal purposes as a medicinal plant. More precisely, a plant is considered medicinal if at least one of its organs possesses therapeutic properties. A medicinal plant is defined as a plant that contains one or more active chemical substances, in low or high concentrations, in one or more of its different organs. These substances have physiological ability to treat a particular disease, or at least reduce the symptoms of the disease if given to the patient in their pure form or in the form of fresh or dried plant material or partially extracted. Medicinal plants have the ability to produce one or more types of active substances, but this does not mean that everything produced by the plant is active. There are also inactive substances, such as cellulose and most of the plant cell components (Alabed, 2009). [9]

Around 35,000 plant species are used worldwide for medicinal purposes, representing the widest range of biological diversity used by most people. Medicinal plants continue to meet the urgent need for healing in many people despite the development of modern healthcare systems (Elsa et al, 2007).

Medicinal plants are used in two forms: (1) raw form, which can take various forms such as infusions, essential oils, and pigments extracts, and (2) pure form, in which the active element responsible for the therapeutic effect is chemically defined and identified. Pure compounds are generally used when the active ingredients have strong and specific effects (Hamburger, 1991). [12]

### **Definition of medicinal plants:**

Medical plants are those that have therapeutic capabilities that can be obtained from nature or cultivated. These medicinal plants can be used in various forms, such as fresh or dried herbs, or the raw material can be used in the manufacture of various liquid and solid extracts. A medicinal plant is one that contains one or more chemical substances in one or more of its various organs or mutations, regardless of the nature of these substances, whether they are in low or high concentrations, and have physiological ability to treat a specific disease or at least reduce the symptoms of the disease if given to the patient in its pure form after extraction from the plant material or if used partially. [9]

### **I.5. Definition of Aromatic Plants**

Aromatic plants are defined as those that contain volatile essential oils in one or more of their plant organs, whether in their free form or in other forms that are converted or decomposed into volatile essential oils with an acceptable aroma, and can be extracted using conventional methods. [14] They are used in various aromatic fields. There is nothing that can be used to distinguish between medicinal and aromatic plants, as some essential oils have medicinal uses, such as cinnamon, and some plants classified as aromatic plants contain medicinal chemicals in addition to essential oils, such as roses. [11]

### **I.6. Sources of medicinal and aromatic plants**

These plants can be obtained from two main sources:

1. Plants grown under controlled conditions in valleys and deltas and newly cultivated lands such as Mentha , Origanum majorana , basil, aromatic grains, hibiscus, chamomile, Chrysanthemum ,Pelargonium graveolens, jasmine, and others. [9]

2. Plants that grow wild in deserts, forests, mountain slopes and peaks, plateaus, swamps, lakes, and seas. In Egypt, they are collected from the Egyptian deserts, the eastern and western north coast, and on the edges of canals and drains such as Artemisia judaica, Horehound, Cactsa ,Ambrosia, Ammi majus, Ammi visnaga , Calotropis, Senna, Citrullus, Colocynthis, Colchicum Datura, Hyoscyamus, Plantago, Mustard, Urginea maritima and others. [11]

## **I.7. Multiplying medicinal and aromatic plants**

Multiplying medicinal and aromatic plants: Medicinal and aromatic plants are propagated in the same way as other plants. They can either be planted using seeds or one of the green propagation methods (cuttings, grafting, offshoots, layering, and runners.... etc). [5]

### **A) Seed propagation:**

The seed contains the embryo resulting from the process of fertilization, which occurs when the pollen nucleus of the male gamete fuses with the egg nucleus of the female gamete. Some seeds contain embryos called "green embryos" that arise from the tissues of the ovary or the new shoot. These green embryos produce plants that are similar to the mother plant, as they did not result from the process of fertilization. In addition to the embryo, seeds contain stored food and one or more coverings called the

seed coat. Many medicinal and aromatic plants are propagated by seeds, such as plants in the Apiaceae family (coriander, caraway, cumin, anise, fennel, etc.), castor oil plant, hibiscus, belladonna, tomato, opium poppy, Indian hemp (hashish), carnation, chili, cloves, nutmeg, camphor, cocoa, chamomile, and others. [9]

## **B) Green propagation:**

Many medicinal and aromatic plants can be propagated using green propagation methods such as cuttings, grafting, and offshoots, or underground parts that grow beneath the surface of the soil, such as rhizomes, tubers, and corms, or by layering or through cankers, or by planting organs, tissues, and plant cells, etc. Green propagation is considered easier, cheaper, and faster than seed propagation, and it may be necessary to use it when the plant does not produce seeds in Egypt or when there is a fear of genetic isolation in seed propagation. [9]

## **I.8. Stages and dates of collecting plant parts**

### **I.8.1. Collection of Medicinal Plants:**

Nature represents a rich source of medicinal plants, where the process of collecting them is both useful and enjoyable. Harvesting or collecting medicinal plants is not a major problem. The important thing knows the suitable plants and the ability to distinguish between them. [6]

### **I.8.2. Timing of Plant Collection:**

It is always better to harvest plants when the weather is dry, as wet plants due to rain or dew can change, rot, and ferment, and may lose any therapeutic value they have. Therefore, morning is considered the most suitable time for plant collection, and it can also be done in the evening before the temperature drops. [8]

#### **1.8.4. Process of Plant Collection:**

It is better to collect wild plants, if possible, from a place with little traffic. Plants intended for drying should not be washed (only the roots should be washed thoroughly with clean water to remove any traces of soil). Therefore, it is advisable to avoid picking plants growing on the edges of roads or on the edges of cultivated fields, which may be contaminated with recently used chemical fertilizers. Only peaceful plants should be selected and wilted plants, those with spots and unusual colors, as well as plants attacked by insects that grow near fungi should be discarded [10]. It is very easy to get rid of various residues, such as algae, leaves, and branches, when collecting the required plant to keep only the plant that interests us. Getting rid of these residues becomes difficult after the collection process is completed. [9]

#### **I.9. Plant parts are collected as follows:**

"Completing the harvesting process requires careful attention to ensure that the plant you wish to collect is not mixed with other plants. If different types of plants are being collected together, they should not be mixed with each other during the collection process. It is also essential to avoid crushing or pressing the plants during collection and careless stacking, which can lead to decay and loss of freshness. This can also lead to early fermentation. It is better to use a large basket specifically designed for collecting plants. [13]

##### **I.9.1. Roots and Rhizomes:**

Collection is done during the plant's growth rest period, in autumn or in spring before the start of plant growth. Digging usually occurs in the second or third year for perennial plants, and in autumn of the first year for annual plants. Before drying, the roots and rhizomes are washed and freed from soil and sand with normal water. Peeling roots is not allowed except when harvested in spring. For

roots harvested in autumn, their skin is stored with active ingredients. They have a thick structure consisting of layers of shells that originally were leaves, and they are most commonly used in medicine.

#### **I.9.1. Bulbs:**

The popular name for bulbs is onions.

#### **I.9.2. Tubers:**

The tuber is swollen and grows underground. The most commonly used is the African potato tuber."

#### **I.9.3. Bark:**

The bark is usually collected in the spring, which is when the sap flows in the plant due to its vegetative growth, and due to the flow of sap in the bark vessels, it is easy to remove the bark during this period. The collection time is chosen after a period of high humidity, which also helps to separate the bark layer from the wood, making the collection process easier, such as in the case of cinnamon .

#### **I.9.4. Wood:**

It is rarely used and is usually shredded by carpenters or cut into chips

#### **I.9.5. Leaves and Herbaceous Stems:**

Leaves and growing plant tops are collected when they are rich in active ingredients, and this time is when photosynthesis is most active, which is in the spring season. The period before flower formation or before its full formation is considered the stage when the leaves are rich in active ingredients, and this is the most suitable stage for collecting leaves rich in their components .Leaves are collected in the afternoon, where their active substance content has increased, and leaves or branches that are moist should not be collected as they become easily rotten .Leaf collection is usually done by hand, avoiding collecting all of them so that the plant is not deprived of all its green area. Sometimes, entire branches are

cut with scissors, and the leaves are later collected from these branches after the drying process. It is important to avoid rubbing the leaves or piling them in a basket or bag

- ✓ Flowers differ from other parts of the plants in that their collection period is very short and requires precision and care in choosing the appropriate time to collect them, and in general flowers are collected before as . so on as flowering begins, such as camomille and jasmine. [9]

### **I.10. Drying :**

It is the process of removing moisture from the material to be dried. This process should be applied immediately after a collecting the plant .the plants are placed distributed in well ventilated room, placed on a jut or cotton fabric, where the different species are separated from each other. It should also not be exposed to radiation. [9]

Direct sun , unless. It is actually mentioned during that exposing it to sunlight may lead it to lose some of its properties, due to the volatilization of many materials.

Medicinal plants are dried at temperature between (90-60°) if the plats is aromatic the collection process takes place in the mining and dried at a temperature mot exceeding (50°). [10]

### **I.11. Natural drying methods:**

It relies on the use of natural energy sources (sun andair) to evaporate moisture form plants. [12]

### **I.12. Dehydration :[9]**



The rate of drying at the same time, these factors are called properties of drying rate in industrial drying leads to rapid evaporation of moisture from the plants. [9]

Table 1 : the percentage of moisture of plant parts after drying

Final moisture content	Dry plant part
%12-10	Leaves
%14-12	Flowers
%14-12	Grass
%14-12	Roots and rhizomes
%14-10	Seed
%20-11	Fruit

### **I.13. preservation:**

After the plants have dried, they must move to preservation stage directly, to prevent dust from accumulating on them. To this end, we use paper bags with a long period of time, plastic bags (except for types that contain essential oils). And glass containers, it should always be checked that water does not condense on the walls of the container, which indicates a problem in the drying process.

### **I.14. The most important areas of use of medicinal and aromatic plants:**

The most important areas of use of medicinal and aromatic plants:

There are many fields in which medicinal and aromatic plants can be used

The fields are:

- ✓ Preparing some medicines, such as medicines to relieve joint pain and rheumatic infections.

- ✓ And medicines for high blood pressure and atherosclerosis and as an antiseptic.
- ✓ Production of fixed oils, as the seeds of some of these plants contain fixed oils
- ✓ interfering in the installation of some medical preparations.
- ✓ Food processing for the treatment of atherosclerosis and angina pectoris, such as oil.
- ✓ Hohoya , sunflower, flax, and castor seeds.
- ✓ Preparing cosmetics such as powders, hair creams, and soaps.
- ✓ It is used in the manufacture of scents and perfumes, and among these plants are the rose and jasmine.
- ✓ Manufacture of insecticides, which depend on what is found in medicinal and aromatic plants.
- ✓ Of deadly poisons, whether for insects or fungi, such as these plants (Al-Baid them, and Alders , henna and smoke).
- ✓ It is used as seasonings, spices, drinks, or flavorings or aromas. [11] [14]

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**Chapter (II):**

**MEDICINAL AND AROMATIC PLANTS COMMERCIALY AND  
ECONOMICALLY**

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## **II.1. entrance :**

Students are increasingly interested in medicinal and aromatic plants because of their economic importance in the manufacture of medicines, cosmetics and other fields, and interest in them has begun on the commercial side and their contribution to the exports of some countries.

## **II.2. The economic importance of medicinal and aromatic plants commercially and economically :**

Medicinal and aromatic plants have a bright global market, as the value of exports in the first twenty countries amounted to 609.9 US dollars, representing 80.23 million dollars of the total world exports for the year 2001. Among the most important countries exporting medicinal and aromatic plants in the world: China, India, France, and the United States of America , Singapore, Chile, and among the most important countries exporting medicinal and aromatic plants in the Middle East: Egypt, Iran, Syria, Morocco, Tunisia, and among the most important of these countries is the Arab Republic of Egypt out of a total of 2.33%.<sup>[1]</sup>

## **II.3 . Brief General on Cultivator the plants medical and aromatic in north Africa:**

North Africa has one of the oldest and richest traditions related to the use of medicinal plants. These plants are important to the people of the region, especially in the rural areas, where they are in some places the only available source of medicine, as well as in the cities, the prices of modern medicines are on the rise, and people are returning to the use of traditional phytotherapy.<sup>[2]</sup>

Egypt: More than 30 species of medicinal plants are grown as export plants, on an area of 80,000 acres, and the Egyptian flora contains more than 2,500




medicinal and aromatic plants between the amoecious and labial family, as well as compound . [3]





Algeria.:

It used to count more than 3,000 species of aromatic and medicinal plants, which made it occupy the forefront at the level of the great Arab population, but this position has declined due to human neglect as well as climate changes. [4]




#### II.4. Examples of products made from thyme oil :

Tabel 2: Examples of products made from thyme oil<sup>[5]</sup>

THE PRODUCT	PICTURE
Bobong oil	
Sweet Orange Deodorant	
Lip Balm	

<p>Bath Salts or Soaks</p>	 <p>CHAMOMILE <i>bath salts</i></p> <p>A glass jar filled with white bath salts, garnished with chamomile flowers, sits on a wooden surface. A white cloth bag and more flowers are scattered around.</p>
<p>Bath Bombs</p>	 <p>Two round, white bath bombs are shown on a wooden surface, surrounded by fresh chamomile flowers and greenery.</p>
<p>Lotion Recipes</p>	 <p>CHAMOMILE &amp; ELDER <i>lotion</i></p> <p>A blue jar of white lotion is displayed on a wooden surface, accompanied by a bouquet of chamomile and elder flowers.</p>
<p>Tea</p>	 <p>A glass of golden chamomile tea is served in a clear glass, garnished with several chamomile flowers floating on top. More flowers are scattered on the wooden table.</p>



<p>Soap</p>	
<p>Rose Cleansing Grains</p>	
<p>Rose Lip Balm</p>	

## II.5. Conditions for exporting medicinal and aromatic plants:

- Application of most quality standards and availability of the product to the required specifications in the manufacturing and production stages as well as export
- Obtaining the approval of the US Food and Drug Administration and the Pharmacopoeia
- Obtaining approval from the Climate Change Authority after export, in order to preserve the safety of the product, and the conditions differ between countries<sup>[6]</sup>

- The plant must be grown using a biological or organic method (verified by analyzing the plant before obtaining quality documents).
- Continuous development of the quality of the product to be exported
- The trend towards organic production (environmentally friendly plants), as the demand for them increased, especially in the European market . [7]

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**Chapter (III):**  
**Common Chamomile**

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### **III.1. entrance**

Plants are divided according to the family, and each family is also divided in turn, and this is according to international classification standards, as these plants may belong to the same family, but they differ in shape, taste, or others, and this is according to the terrain and climate diversity from one region to another in the world, and this diversity may play a role in the difference of elements effective with it.

### **III.2 . Plants of the asteraceae family**

The compound family is known as compound<sup>[1]</sup> or asteraceae<sup>[2]</sup> and has the scientific name asteraceae or the sunflower family to which a large number of secondary vegetable crops belong, as it includes about 1620 genera and 23000 species (according to the Royal British Kew Gardens) <sup>[3]</sup>. It is the most widespread plant, as its plants are found in all parts. <sup>[4]</sup>

#### **III.2.1. General classification:**

The range : eukaryotes

The kingdom: plants

Upper band: embryonic plants

Section : vascular plants

Division : eukaryotes

Saa : by seed

Architecture: star

Sect :       dicotyledons

Rank:       asters

Platonon:   asterisk

Family:   stars

Tribe:   stellate

Al Amira:   star

Gender:   star <sup>[5]</sup> <sup>[6]</sup>

### **III .2.2. General characteristics characteristic of the labial family :**

Flowers are often complete, but some plants of the family are monoecious and monoecious, while others are monoecious and dioecious. And the light in the compound family is important Capitulum (or Head Head). The flower consists of five scaly sepals, five fused petals in the form of a tube that bears the top of the ovary, five stamens that bear the corolla, a lower ovary, and one pen that ends with two stigmas. Pollination is either self- or mixed.

The fruit in the compound family consists of one chamber, and it is dry when ripe. It is metaphorically called the “seed”, but it is a real fruit with poor achene, which is seated, and has a long tip that is sometimes withdrawn. The seeds are not endosperm <sup>[7]</sup>.

### **III.3. CHAMOMILE:**

#### **III.3.a. Definition of chamomile:**

Chamomile is considered an annual plant with straight, smooth branches, with regularly divided leaves, and its smell resembles apples and pineapples. It grows to a height of 60-91 centimeters, and its flowers are about 2.5 centimeters in

diameter. It has a hollow conical center, and it is covered with small yellow flowers, surrounded by silver flowers up to White in color, [8] Many people enjoy drinking chamomile tea as an alternative to black or green tea, because it is caffeine-free, and has a distinctive, somewhat sweet taste [9]

### **III.3.b. Description of chamomile:**

Annual plant glabrous, aromatic, 2-5 dm erect or ascending, branching; leaves bipinnatisect with almost filiform segments; involucre with oblong-obovate inner leaflets broadly scarious; very small (about 1 mm) yellowish-white achenes, subcylindrical, slightly arched, with 5 filiform ribs on the internal face, smooth on the back with a very oblique epigynous disc provided with a very short rim; hollow receptacle with conical and pointed end; flower heads mediocre (about 2 cm in diameter) corymbose; center flowers yellow, ligules white. [10]

### **III.3.c. The history of chamomile:**

The name chamomile is derived from a Greek word meaning “apple of the earth”, because the Greek chamomile has a scent similar to the smell of apples, and chamomile originated in Europe and Western Asia since ancient times, and it was highly valued by the Egyptians, Romans and Greeks for its medicinal properties, to the extent that the ancient Egyptians symbolized it with a god The sun, Ra, because they believed it helped treat fevers according to the Eber Papyri dated 1550 BC. The ancient Egyptians also used the herb to honor the gods, mummify the dead, and treat the sick. [11]

**III.3.d. Botanical classification Chamomile:**

*Matricaria chamomilla*)<sup>[12]</sup>

Scientific classification

Kingdom: Plantae

Clade: Tracheophytes

Clade: Angiosperms

Clade: Eudicots

Clade: Asterids

Order: Asterales

Family: Asteraceae

Subfamily: Asteroideae

Tribe: Anthemideae

Genus: *Matricaria* L.

Type species

*Matricaria recutita* L.

Synonyms<sup>[13]</sup>

*Lepidothea* Nutt.

*Cotulina* Pomel

*Achyloopsis* Lehm.

*Gama La Llave*

*Cenocline* K.Koch

*Lepidanthus* Nutt.

*Camomilla* Gilib.



Figure 1 : Chamomile plant



Courrantia Sch.Bip.

Sphaeroclinium (DC.) Sch.Bip

### III.4. Geographical distribution of Chamomile plant in the world:

Chamomile is widespread throughout Europe, originating in Eastern Europe and the Orient the Middle East, and also increasingly in India, North America, and Australia, and is known or common on Wide range in North and East Africa as well as in Hungary and Croatia..

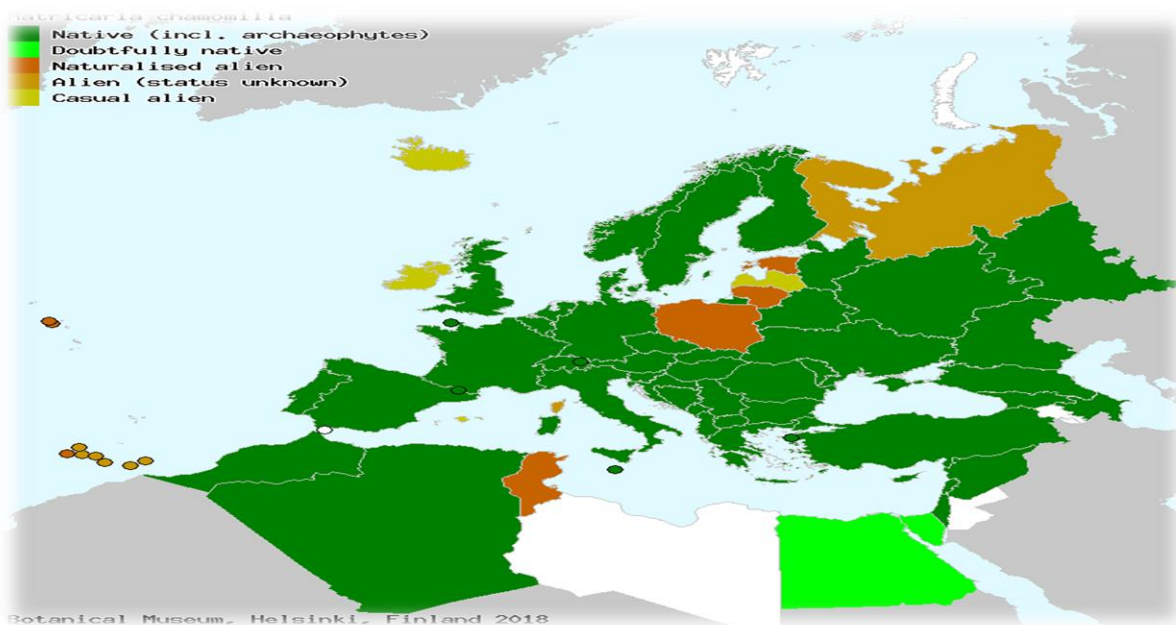


Figure 2: Geographical distribution

### III. 5. Chamomile cultivation climate:

Chamomile grows well in temperate climates from Eastern Europe, to Mediterranean countries and subtropical climates in South Africa. It is a resilient plant that can survive Nighttime temperatures as low as 14 degrees Fahrenheit (10 degrees Celsius). The best medium climates for chamomile plants are those with long warm days (sufficient hours of sunshine and cool nights). Full sun exposure is necessary for essential oil production. During active growth (late

winter to spring, plants thrive in a day temperature of 60 degrees Fahrenheit (15-20°C). [13]

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**Chapter (IV):**  
**Compounds of Secondary Metabolites**

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## **IV.1. Entrance :**

Plants are capable of producing and synthesizing diverse groups of organic compounds and are divided into two major groups : primary and secondary metabolites . secondary metabolites are metabolic intermediates or products which are not essential to growth and life of the producing plants but rather required for interaction of plants with their environment and produced in response to stress . some secondary metabolites such as phenylpropanoids protect plant from UV damage . the biological effects of plant secondary metabolites on humans have been known since ancient times .

## **IV.2. Définition of secondary metabolites :**

The secondary metabolites are usually characterized by being complex molecules with a large molecular weight compared to the primary metabolites

They are produced in large quantities and are usually excreted outside the cells

They have certain roles in relation to the producing cells ,for example ,they help the cells in the formation of plaques <sup>[1]</sup>

Each group of organisms may specialize in the production of a specific group of secondary metabolites ,but there are some organisms that produce different groups of secondary metabolite differ from one organism to another ,and most of them are related to primary metabolites that may

Act as precursors for them ,and the pathways for the synthesis of secondary metabolites are specific for one substance . <sup>[2]</sup>

## **IV.3. The alkaloids**

### **IV.3.1. Definition of alkaloids :**

the term alkaloids was first coined by the scientist (Meisner) in 1809 AD they are defined as nitrogenous organic compounds composed of (CH), (N) many of which contain in the structural structure one or more heterocyclic rings, formed from security acids. [10]

### **IV.3.2. Classification of alkaloids**

#### **IV.3.2.a. True alkaloids :**

Which are alkaloids that contain one or more nitrogen atoms in heterocyclic rings, and they are derivatives amino acids, for example : colchicine.

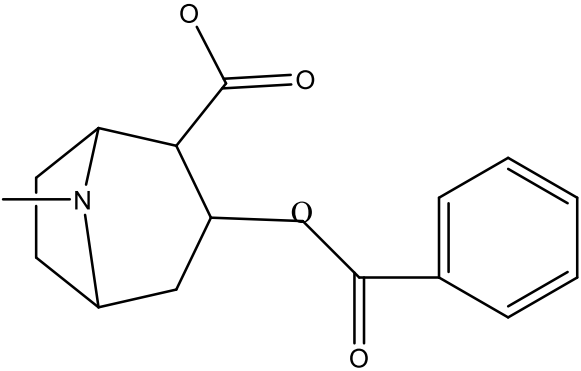
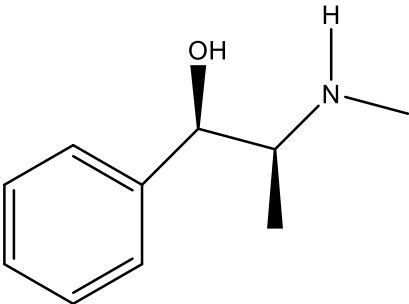
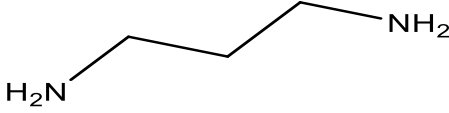
#### **IV.3.2.b. Primary alkaloids :**

They are alkaloids in which the nitrogen atom is not in a heterocyclic ring (mescaline, ephedrine)

#### **IV.3.2.c. Pseudo alkaloids :**

That are not derived from amino acids, such as steroidal alkaloids (solanidine in potato varieties) and purine alkaloids.

Table3 : selected examples of each groups of alkloids [11]

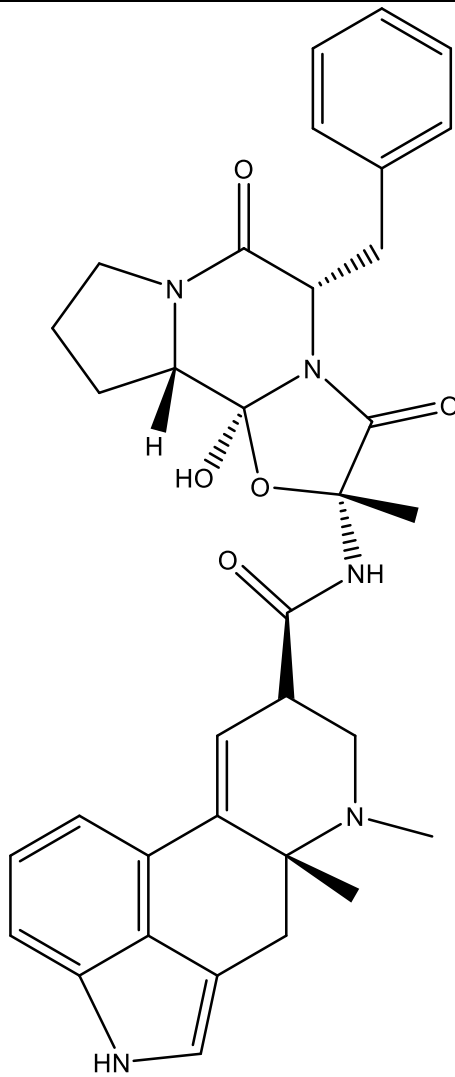
<p><b>Heterocyclic alkaloids</b></p> <p>These are alkaloids that have nitrogen as part of their cyclic ring system ,they are</p> <p>The most common group of alkaloids in Nature(cocaine)</p>	
<p><b>Alkaloids with exocyclic nitrogen atom</b></p> <p>Also called proto alkaloids or biological amines ,these alkaloids have nitrogen that is not part of any ring system,they are less common in nature (ephedrine)</p>	
<p><b>Polyamine alkaloids</b></p> <p>These are aliphatic molucules with one ormore amino</p>	



groups, an example is putrescine

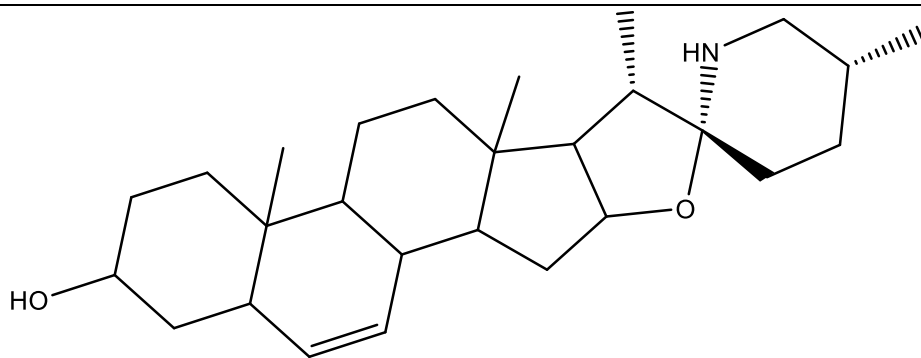
**Peptide alkaloids**

Peptide alkaloids are composed of amino acid monomers linked by peptide bonds (ergotamine)



**Terpene and steroidal alkaloids**

Terpene alkaloids are made of a mono, sesqui, di, and tri terpenoid skeleton while steroidal alkaloids



as the name indicates have a steroid skelton (solasodine)	
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### **IV.3.3. DETECTION OF ALKALOIDS :**

3ml of extract was stirred with 3ml of 1 HCL on steam bath, 1ml of mixture was taken separately in two test tubes, few drops of Dragendorff's reagent were added in one tube and occurrence of orange red precipitate was taken as positive.

In the second tube Mayer's reagent was added and appearance of buff colored precipitate was taken as positive test for presence of alkaloids. [21]

### **IV.3.4. The medical importance of alkaloids : [12;20]**

Substances considered analgesics or reduce activity at the level of nerve centres such as morphine, dilating the pupils such as atropine, anti-cancer such as vinca alkaloids, stimulants such as caffeine, diuretics such as xanthine, antibacterial and anti-fungal glycosides. [12] [20]

## **IV.4. Flavonoids :**

### **IV.4.1. Define it :**

Flavonoids are an important class of natural products; particularly, they belong to class of plant secondary metabolites having a polyphenolic structure, widely found in fruits, vegetables and certain beverages, they have miscellaneous favourable biochemical and antioxidant effects associated with various diseases such as cancer, they are associated with a broad spectrum of health-promoting effects and are an indispensable component in a variety of nutraceutical, pharmaceutical, medicinal and cosmetic applications. [13] [14]

#### **IV.4.2. Classification of flavonoids :**

##### **IV.4.2.a. Flavanols,flavan-3-ols or catechins :**

flavanols,also called dihydroflavonols or catechins, are the 3-hydroxy derivatives of flavanones,they are a highly diversified and multisubstituted subgroup,flavanols are also referred to flavan-3-ols as the hydroxyl group is always bound to position 3 of the C ring ,unlike many flavonoids , there is no double bond between position 2 and 3 ,flavanols are found abundantly in bananas ,apples, blueberries , peaches and pears. [14]

##### **IV.4.2.b. Flavanones :**

Flavanones are another important class which is generally present in all citrus fruits such as oranges , lemons and grapes Hesperetin, naringenin and eriodictyol are examples of of this class of flavonoids,Flavanones are associated with a number of health benefits because of their free radical-scavenging properties ,These compounds are responsible for the bitter taste of the juice and peel of citrus fruits ,Citrus flavonoids exert interesting pharmacological effects as antioxidant,anti-inflammatory , blood lipid-lowering and cholesterol –lowering agents,Flavanones , also called dihydroflavones, have the C ring saturated ;therefore , unlike flavones , the double bond between positions 2 and 3 is saturated and this is the only structural difference between the two subgroups of flavonoids , Over the past 15 years , the number of flavanones has significantly increased. [15]

##### **IV.4.2.c. Isoflavonoids :**

Isoflavonoids are a large and very distinctive subgroup of flavonoids .Isoflavonoids enjoy only a limited distribution in the plant Kingdom and are predominantly found in soybeans and other leguminous plants ,Some isoflavonoids have also been reported to be present in microbes ,They are also found to play important role as precursors for the development of phytoalexins

during plant microbe interactions, Isoflavonoids exhibit tremendous potential to fight a number of diseases. Isoflavones such as genistein and daidzein are commonly regarded to be phyto-oestrogens because of their oestrogenic activity in certain animal models, Szkudelska & Nogowski reviewed the effect of genistein inducing hormonal and metabolic changes, by virtue of which they can influence various disease pathways. [16]

#### **IV.4.2.d. neoflavonoids :**

neoflavonoids are a class of polyphenolic compounds, While flavonoids have a 2-phenylchromen-4-one backbone, neoflavonoids have a 4-phenylchromen backbone with no hydroxyl group substitution at position 2, The first neoflavone isolated from natural sources in 1951, It is also found in the bark and timber of the Sri Lankan endemic plant. [17]

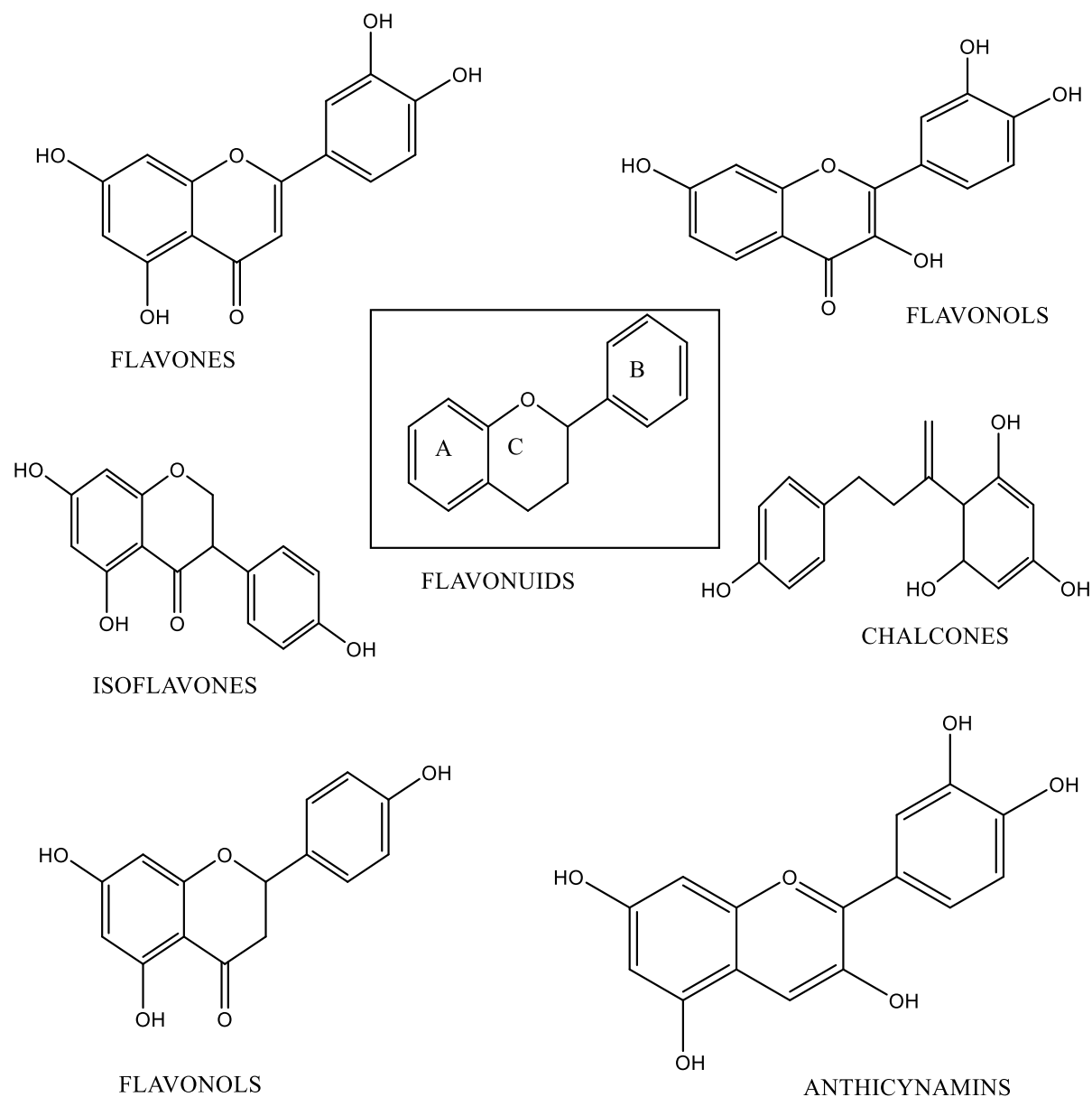
#### **IV.4.2.e. Anthocyanins :**

Anthocyanins are pigments responsible for colours in plants, flowers and fruits, Cyanidin, delphinidin, malvidin, pelargonidin and peonidin are the most commonly studied anthocyanins, They occur predominantly in the outer cell layers of various fruits such as cranberries, black currants, red grapes, merlot grapes, raspberries .....ect, Stability coupled with health benefits of these compounds facilitate them to be used in the food industry in a variety of applications, The colour of the anthocyanin depends on the pH and also by methylation or acylation at the hydroxyl groups on the A and B rings. [16]

#### **IV.4.2.f. Chalcones :**

Chalcones are subclass of flavonoids, they are characterised by the absence of ring C of the basic flavonoid skeleton structure shown in Fig 1. Hence, they can also be referred to as open-chain flavonoids, Major examples of chalcones include

phloridizin, arbutin, phloretin and chalconaringenin, Chalcones and their derivatives have garnered considerable attention because of numerous nutritional and biological benefits. [18]



#### IV.4.3. Properties of flavonoids :

Flavonoids are water –soluble pigments found in the vacuoles of plant cells, they can also be divided into three groups ;anthocyanins, flavones and

flavonols, They are widely distributed in plants, fulfilling many functions such as flower coloration, producing yellow, red or blue pigmentation in petals designed to attract insects involved in UV filtration, symbiotic nitrogen fixation and floral pigmentation. [19]

#### IV.4.4. Flavonoids in the matricaria chamomilla :

Table 4 : Flavonoids in the matricaria chamomilla [14]

Extract compound	Type Matricaria	Extraction method
Kaempferol- 3 -O - rutinoside	Matricaria recutita	EPI, PI, NL and MRM
	Matricaria recutita	UV MS, std
	Matricaria pubescens	HPLC, PDA, MS chromatogram
Luteolin- 7- O - Glucoside	Matricaria recutita	LC, PDA, MS chromatogram
	Matricaria chamomilla	UHPLC, HESI, MS MS
	Matricaria chamomilla	LC, DAD, MS
	Matricaria pubescens	HPLC, PDA, MS chromatogram
6-hydroxy-7-O- glucoside	Matricaria recutita	LC- PDA- MS chromatogram
	Matricaria recutita	LC- PDA- MS chromatogram
Apigenin-4'-acetyl hexoside (tentatively identified)	Matricaria recutita	LC- PDA -MS chromatogram
Apigenin-7-acetyl- hexoside Apigenin-7-O-(6- malonyl)-glucoside	Matricaria recutita	LC PDA MS chromatogram
	Matricaria recutita	LC PDA MS chromatogram

Apigenin-8-C-glucoside	Matricaria recutita	EPI PINLandMRM
Apigenin-7-O- $\beta$ -d-glucoside	Matricaria recutita	UHPLC- UV chromatogram
Apigenin-7-O-(6''-O-acetyl)- $\beta$ -d-glucoside	Matricaria recutita	UHPLC –UV chromatogram
MAB apigenin-7-O- $\beta$ -d-glucoside	Matricaria aurea	UPLC- MS- MS- UV –visible (NMR)
Apigenin 7-glucoside	Matricaria chamomilla	HPLC- DAD- MS
Apeginin -7-O-acetyl glucoside	Matricaria chamomilla	LC- DAD- MS
	Matricaria chamomilla	UHPLC- HESI- MS MS
Kaempferol-3-O-glucoside	Chamaemelum nobile	UV-vis
Kaempferol-1-O-pentosylhexoside		
Kaempferol	Matricaria chamomilla	UHPLC- HESI MS -MS
	Matricaria chamomilla	HPLC- DAD -MS
Kaempferol-7-neohesperidoside 6,8 Dihydroxy Kaempferol 3-rutinoside	Matricaria chamomilla	UHPLCQ- TOF- MS
	Matricaria chamomilla	UHPLCQ- TOF- MS

#### IV.4.5. Detecting of flavonoids :

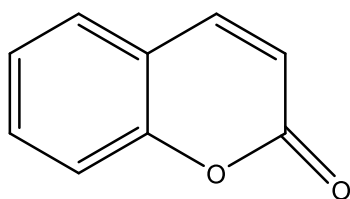
To 1ml of extract , 1ml of 10% lead acetate solution was added , the formation of a yellow precipitate was taken as a positive test for presence of flavonoids.

#### IV.5. Coumarins:

##### IV.5.1. Define it :

Coumarins(2H-1-benzopyran-2-one) consist of a large class of phenolic substances found in plants and are made of fused benzene and alpha pyrone rings. More than 1300 coumarins have been identified as secondary metabolites

from plants, bacteria, and fungi, The prototypical compound is known as 1,2-benzopyrone or, less commonly, as o-hydroxy cinamic acid lactone, and it has been well studied, coumarins were initially found in tonka bean (*Dipteryx odorata* wild) and are reported in about 150 different species distributed over nearly 30 different families, of which a few important ones are Rutaceae, Umbelliferae. [21]



COUMARINS

Figure 2: The basic unit of coumarin

#### IV.5.2. Detecting of coumarins :

Exposing the muzzle → color yellow-greenish

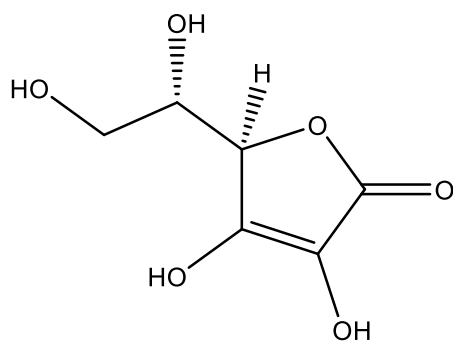
Alcoholic x- → greenish [21]

#### IV.6. Phenolic acids :

##### IV.6.1. Define it :

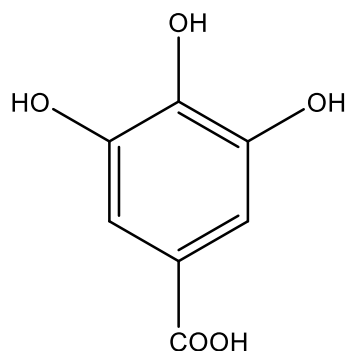
Phenolic acids are plant secondary metabolites widely spread in plant kingdom (Buneton, 1993), they are derivatives of benzoic or cinamic acid (Mchansho et al, 1987), the main subclasses of phenolic acids include Hydroxy benzoic acids and Hydroxycinnamic acids (Cheynier, 2005). [19]





Ascorbic acid

Figure 3: Ascorbic acid



Gallic Acid

Figure 4: Gallic acid

#### IV.6.2. Biological and therapeutic properties of phenolic acids :

- ✓ Heat preservation.
- ✓ anti inflammatory.
- ✓ both gallic acid and chlorogenic acid are compounds it has antioxidant activities.
- ✓ acid cafeique is very effective against viruses, bacteria and fungi.
- ✓ acid gallique and acid cafeique, wich showed anti-cancer effects in the lungs of mice in vitro. [12]

#### IV.6.3. Phenolic acid in the Matricaria chamomilla :

Table 5 : Phenolic acid in the Matricaria chamomilla [19]

Chlorogenic	Matricaria chamomilla	HPLC- DAD –MS
Chlorogenic acid	Matricaria recutita	UV- MS ,std
	Matricaria recutita	HPLC -PDA –MS chromatogram
	Matricaria pubescens	HPLC- PDA- MS-chromatogram
	Matricaria recutita	LC-MS- TIC
	Matricaria recutita	UHPLC- UV chromatograms
4,5-O-dicaffeoylquinic acid	Matricaria recutita	UV- MS ,std
	Matricaria recutita	LC- PDA -MS chromatogram

Trans-2-hydroxy-4-methoxycinnamic-oxo—2-O-β-D-glucopyranoside	Matricaria recutita	UV MS, std , NMR
4-hydroxy-3-Methoxycinnamic acid (ferulic acid)	Matricaria recutita	HPLC chromatogram

## IV.7. The terpenoids :

### IV.7. 1. Define it :

Plants tissues are related to adaptation to both abiotic and stressors such herbivores and pathogens, however, the high volatility and reactivity of some terpenoids may also effect the atmosphere composition , volatility of terpenoids provids for sessile plants, a tool for communication with other organisms such as neighbring plants , pollinators and foes of herbifores, via air-bone infochemicals. [6]

### IV.7. 2. Classification of terpenoids :

Table6: Classification of terpenoids [7]

Number of carbone	Name	Example
C5	Hemiterpene	Isoprene, prenol, isovaleric
C10	Monoterpene	acid,lmonene,eucalyptol,
C15	Sesquiterpene	pinene, giberellin
C20	Diterpene	Squalen
C25	Sesterterpene	lanosterol, lycopene,
C30	Triterpene	vitamine e
C40	Tetraterpene	
C 40	Polyterpene	

### **IV.7. 3. Detecting of terpenoids :**

2ml of the organic extract was dissolved in 2ml of CHCL<sub>3</sub> and evaporated to dryness, 2ml of conc H<sub>2</sub>SO<sub>4</sub> was then added and heated for about 2minutes, developement of a grayish color indicates the presence of terpenoids.

### **IV.8. gly cosides :**

#### **IV.8.1. define it :**

glycosides may be phenol, alcohol or sulfur compounds, they are characterized by a sugar portion or moiety attached by a special bond to one or non-sugar portions , many plants store chemicals in the form of inactive sugar, which can be activated by enzyme hydrolysis, for this reason, most glycosides can be classified as produgs since they remain inactive until they are hydrolyzed in the large bowel leading to the release of the glycone, the right effect of membranes and skin. [12]

#### **IV.8.2. detecting of glycosides :**

to 2ml of extract with dilute HCL and 2ml sodium nitropruside in pyridine and sodium hydroxyde solution were added , formation of pink to blood red color indicates the presence of glycosides. [17]

### **IV.9. saponins :**

#### **IV.9. 1. define it :**

saponins are compounds whose active portions form colloidal solutions in water , which produce lather on shaking and percipitate cholesterol .They occur as glycosides whose aglycone tripenoid or steroidal structures, the combination of lipophilic sugars at the end gives them the ability to lower surface tention, producing the detergent characteristic or soap-like effect on membranes and skin

They are largely distributed in plant kingdom, which have many physicochemical (foaming, emulsification, solubilization, sweetness and bitterness) and biological properties (haemolytic, antimicrobial, antioxidant, molluscicide, ichthyocide), exploited in many applications in food, cosmetics, pharmaceutical, industries and soil bioremediation. Among the saponins properties, CMC (critical micellar concentration), maximum surface density and aggregation number (number of monomers in a micelle) are of great importance for application as surfactants and foaming agents, these are influenced by variables such as temperature, salt concentration, aqueous phase pH, solvent concentration and type, such as ethanol or methanol. [9]

#### IV.9. 2. detecting of saponins :

5ml of extract was shaken vigorously with 5 ml of distilled water in a test tube and warmed, the formation of stable foam was taken as an indication of the presence of saponins. [21]

#### IV.10. Resins :

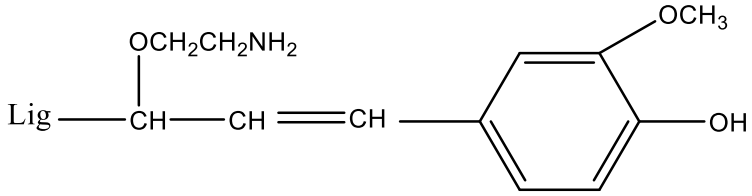
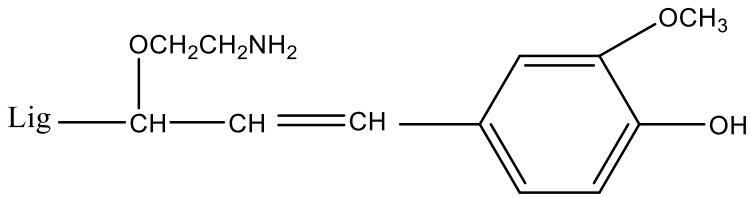
##### IV.10.1. Define it

They are solid or semi-solid organic compounds, their composition is different and chemically complex resulting from the oxidation of 1/6 of the volatile oils. Resins are defined as plant secretions produced from plant tissues, either medically when the plant is exposed to physiological damage as a result of infection with a pathogen or mechanical damage as a result of the influence of factors: infection with an insect pest, resins can be produced industrially as a by-product of formaldehyde or by freezing the resins after mixing it with glycerin, such as pinegum resin.

##### IV.10.2. SOME RESINS :

Table 7: some resins

1	
---	--

2	
3	

## IV.11. Volatile oils

### IV.11.1. Detecting of volatile oils :

Detection of volatile compounds present in essential oils is possible and even the confirmation of identity of such compounds can easily be achieved, to deal with adulteration based on the addition of synthetic versions of compounds present in the essential oils, isotope-ratio mass spectrometry and chiral GC analysis have demonstrated great efficiency, in cases in which the adulteration with a non-volatile substance is suspected, the use of liquid chromatography (high performance liquid chromatography, HPLC) could be useful. However, this technique is not routinely used for quality control in industries and methods based on GC are still predominant with the consequent limitations. [21]

### IV.11.2. Thin layer chromatography (TLC) :

Is a chromatography technique used to separate mixtures, they was discovered by M-Tswett in 1906, thin layer chromatography is performed on a sheet of glass, plastic, or aluminum foil, which is coated with a thin layer of adsorbent material, usually silica gel, aluminum oxide, or cellulose (blotter paper), This layer of adsorbent is known as the stationary phase, after the sample has been applied on the plate, a solvent or solvent mixture (known as the mobile phase) is drawn up the plate via capillary action, Because different analytes ascend the TLC plate at different rates, separation is achieved.

### **IV.11.3. Extraction :**

Extraction, as the term is used pharmaceutically, involves the separation of medicinally active portions of plant or animal tissues from the inactive or inert components by using selective solvents in standard extraction procedures. The products so obtained from plants are relatively impure liquids, semisolids or powders intended only for oral or external use. These include classes of preparations known as decoctions, infusions, fluid extracts, tinctures, pilular (semisolid) extracts and powdered extracts. Such preparations popularly have been called galenicals, named after Galen, the second century Greek physician.

The purposes of standardized extraction procedures for crude drugs are to attain the therapeutically desired portion and to eliminate the inert material by treatment with a selective solvent known as menstruum, the extract thus obtained may be ready for use as a medicinal agent in the form of tinctures and fluid extracts. [18]

## **IV.12. Essential oils :**

### **IV.12.1. Define of essential oils :**

Essential oils alcoholic are used in a wide variety of consumer goods such as detergents, soaps, toilet products, cosmetics, pharmaceuticals, perfumes, confectionery food products, soft drinks, distilled beverages (hard drinks) and insecticides. [18]

### **IV.12.2. Locations of concentration of essential oils :**

It was found that the location of the essential oils in the plant (which plant organs it is found) greatly affects the method of obtaining the oil, for example, the oils found in flower petals are sensitive and obtained by extraction with organic solvents or grease. As for the oils found in fruits, leaves or roots, they can be obtained by different distillation methods. Also, the location of the essential oils in the tissues of the plant, whether inside or outside the cells, affects the thickness

of their cells in the first case, For example, the oil of strife is found inside thick cells in the flowers, so it is extracted with hot grease, while the jasmine oil is found in the flowers outside the cells, so it is obtained by extraction with grease, On the cold, and purpose of heating in the first case is to detonate the cells or oil glands to get the oil out and extract it. [18]

#### **IV.12.3. Methods of extracting essential oils :**

##### **Distillation :**

##### **A-water distillation :**

In this method, the material is completely immersed in water, which is boiled by applying heat by direct fire, steam jacket, closed steam jacket, closed steam coil or open steam coil, the main characteristic of this process is that there is direct contact between boiling water and plant material.

When the still is heated by direct fire, adequate precautions are necessary to prevent the charge from overheating, when a steam jacket or closed steam coil is used, there is less danger of overheating; with open steam coils this danger is avoided, But with open steam, care must be taken to prevent accumulation of condensed water within the still, Therefore, the still should be well insulated, the plant material in the still must be agitated as the water boils, otherwise agglomerations of dense material in the still must be agitated on the bottom and become thermally degraded, Certain plant materials like cinnamon bark, which are rich in mucilage, must be powdered so that the charge can readily disperse in the water; as the temperature of the water increases, the mucilage will be leached from the ground cinnamon, this greatly increases the viscosity of the water-charge mixture, thereby allowing it to char. [20]

Consequently, before any field distillation process, from this laboratory trial, the yield of oil from a known weight of the plant material can be determined, the laboratory apparatus recommended for trial distillations is the Clevenger system.

### **Water and steam distillation :**

In water and steam distillation, the steam can be generated either in a satellite boiler or within the still, although separated from the plant material, like water distillation, water and steam distillation is widely used in rural areas, moreover, it does not require a great deal more capital expenditure than water distillation, also, the equipment used is generally similar to that used in water distillation, but the plant material is supported above the boiling water on a perforated grid, in fact, it is common that persons performing water distillation eventually progress to water and steam distillation.

It follows that once rural distillers have produced a few batches of oil by water distillation, they realize that the quality of oil is not very good because of its still notes (subdued aroma). As a result, some modifications are made, using the same still, a perforated grid or plate is fashioned so that the plant material is raised above the water, this reduces the capacity of the still but affords a better quality of oil, if the amount of water is not sufficient to allow the completion of distillation, a co-habitation tube is attached and condensate water is added back to the still manually, thereby ensuring that the water, which is being used as the steam source, will never run out.

It is also believed that this will, to some extent, control the loss of dissolved oxygenated constituents in the condensate water because the re-used condensate water will allow it to become saturated with the dissolved constituents, after which more oil will dissolve in it. [20]

### **Hydrodistillation :**



In order to isolate essential oils by hydrodistillation, the aromatic plant material is packed in a still and a sufficient quantity of water is added and brought to a boil ; alternatively, live steam is injected into the plant charge , due to the influence of hot water and steam, the essential oil is freed from the oil glands in the plant tissue, the vapor mixture of water and oil is condensed by indirect cooling with water, from the condenser , distillate flows into separator, where oil separates automatically from the distillate water. [20]

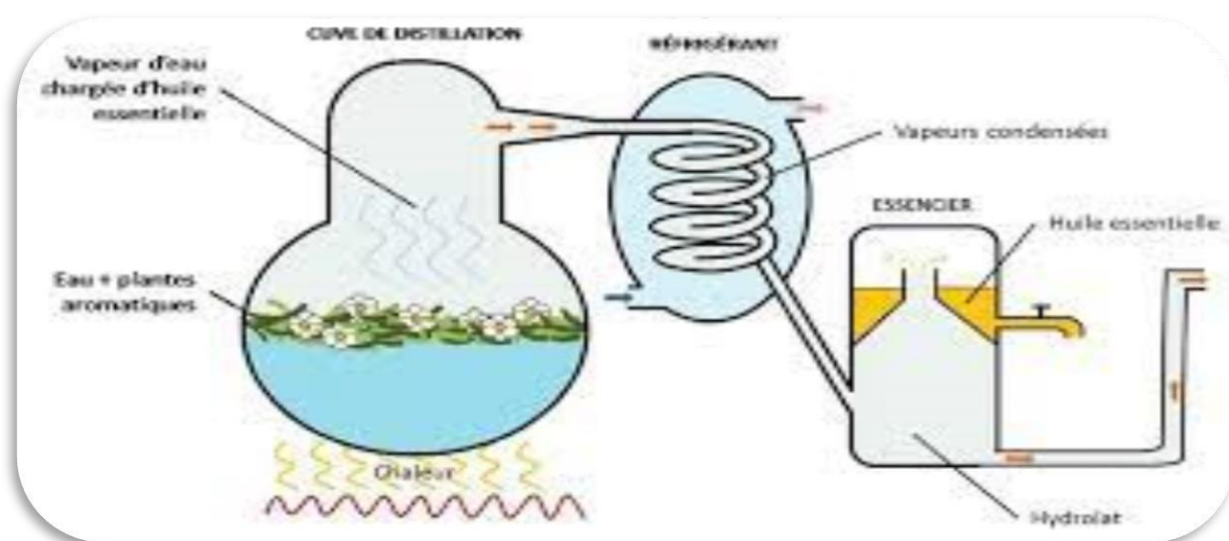


Figure 5: Method of Hydrodistillation [20]

#### IV.12.4.Method of extraction with volatile organic solvents :

Extraction by volatile organic solvents this method is used in the case of plants whose aromatic oils are affected by temperature (the boiling point of water) such as jasmine, lilly and violet flowers, where they are extracted by volatile organic solvents, and used in the manufacture of expensive perfumes, and it is customary not to use this type of essential oil in food or medical purposes because of the traces that may remain from organic solvents. [9]

The idea in this method is to extract the oil from the flowers using an organic solvent such as hexane, benzene, petroleum ether, or carbon tetrachloride, as the boiling point of these solvents is usually lower than the boiling point of water, and this is done in special devices that are cylinders that have a top cover placed in them, so it is filled with flowers, and the solvent is pushed into the cylinder after closing the lid, and the flowers are useful.

Thus, in the solvent with the presence of a lower stirrer, a continuous stir occurs until the oil is extracted from the flowers, and the process takes three times soaking (from 3 to 5 hours) depending on the temperature, which should not exceed 40 C, then these solvents are dripped after that under a severe vacuum to lower the boiling point of the solvent, and given the difference in the boiling point of the solvent from the oil, the oil remains in the distillation apparatus and the solvent is received in the receiving unit, where it is reused, it is noted that the organic solvent is extracted from the flower, in addition to the essential oil, some waxy and fatty materials and some vegetable dyes, therefore, the product after the distillation process is called the crude oil, fat, or paste, concrete oil, and it contains.

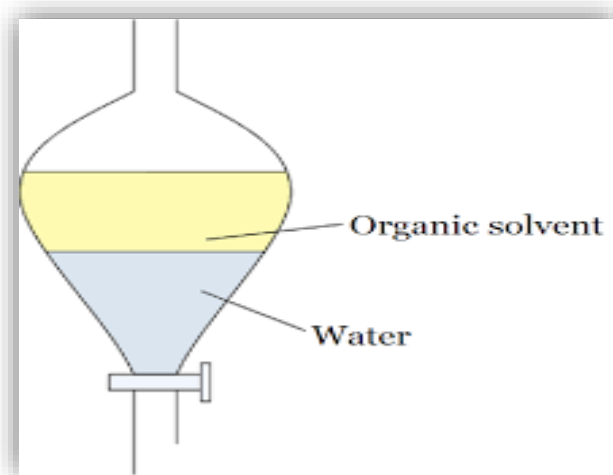


Figure 6: Method of extraction with volatile organic solvents [9]

**Cold pressing extraction :**

the extraction of essential oils by cold pressing is also called scarification by squeezing or coarse extraction, this method of extraction uses the oils of citrus fruits peels such as lemon , orange, bergamot, bitter orange and grape fruit, where the essential oils are found in the peels of these fruits within the oiliness of the flavido layer, due to their high content .

Very high percentage of terpenes up to 90% or more and they are very sensitive to heat and light, and usually it can be extracted with cold pressing method in any of the following ways :(the manual method and the automated method. [9]

#### **Extraction with liquid carbon dioxide :**

It is one of the most moder methods of extracting essential oils from plants, whose oils are sensitive to heat(such as various flower oils), in this method ,liquefied carbon dioxideis passed over the parts of aromatic plantsin special devices under high pressure, where it extracts essential oils from plants quickly and whith very high efficiency, and the oil and liquid carbon dioxide are transferred to the second unit of the device, where the pressure is reduced, and the carbon dioxide is transformed into carbon to the gaseous form and the essential oil is left separated at the bettom of the unit in a very pure form, and at the same time the gas is with drawn and pressurized again to convert to the liquid form and the process is repeated several times , and in the end you get the essential oil with a very high quality. [9]

#### **IV.12.5. Chemical composition of essential oils :**

Essential oils are mixture of serval compounds : terpenoids, aromatic (phenolic)derivatives of phenyl propane, which are the most present, and sometimes waste and carbon derivatives that differ from each other chemically and have different sources. [15] [17]

#### IV.12.6. Physical and chemical properties of essential oils :

- ✓ it does not dissolve in water or has little solubility in it, but rather floats on its surface due to its less specific density than the density of water and its specific density ranges between 0,75- 0,99 very soluble in ether, chloroform, alcohols, in fixed oils and in most organic solvents.
- ✓ colorless or yellowish white, the rare ones being blue or blue-green due to the presence of Azulene, as in chamomile oil.
- ✓ it evaporates or evaporates under natural and normal conditions, which distinguishes it from fixed oil.
- ✓ They are liquid substances after distillation or extraction by different methods of separation. [18]

#### A study of the components of the essential oil in *Matricaria chamomilla* :

Table 8 : chemical composition of essential oil of *matricaria chamomilla* from Nepal [18]

RI <sup>a</sup>	Compound b	%	
1010	- $\sigma$ -3-carene	$\beta$ -Elemene	Tr
1024	P-Cymene	(E)-Caryophyllene	0,07
1028	Limonene	(E)- $\beta$ Farnesene	Tr
1031	1,8-cineole	Germacrene D	Tr
1038	(z)- $\beta$ -	$\beta$ -selinene	Tr
1048	Ocimene	Bicyclogermacrene	0,30
1058	(E) $\beta$ -	(E,E) $\alpha$ -Farnesene	0,07
1062	Ocimene	$\sigma$ -Cadinene	0,22
1172	$\gamma$ -terpinene		0,14
1381	Artemisia		0,08
1385	ketone		0,48

1392	Menthol		0,12
1418	$\beta$ -patchoulene		0,27
1460	$\alpha$ -isocomene		42,21
1482			3,41
1486			0,25
1497			3,16
1510			8,33
1524			0,12

Table 9:chemical composition of chamomile flowers essential oil from

RIexp	Compound	%
925	$\alpha$ -Thujene	0,2
932	$\alpha$ -pinene	1,9
946	Camphene	Tr
971	Sabinene	0,6
974	$\beta$ -pinene	0,1
987	Myrcene	0,7
1002	$\alpha$ -phellandrene	Tr
1014	$\alpha$ -terpinene	0,2
1022	o-Cymene	0,4
1026	limonene	0,3
1029	1,8- $\beta$ Cineole	0,1
1035	(z)- $\beta$ –Ocimene	0,4
1046	(E)-ocimene	2,9
1057	$\gamma$ -terpinene	0,7
1059	Artemisia ketone	0,7
1088	Terpinolene	0,2

#### **IV.12.7. Benefits and uses of essential oils :**

- ✓ They are used as flavoring agents in many food and non-food industries, such as : sweets, bakery products, soda water, soap, and cosmetics, perfumes. [7]
- ✓ It is used in the manufacture of some medicines, as is the case in the use of essential oils for some plants of the umbellifera family(cumin,anise, and caraway) as a carminative gas repellent.
- ✓ They are used in the manufacture of mosquito repellents lotions,as they are applied to exposed areas.
- ✓ The inflorescences are used as a drink,like tea, and it has a stimulating effect on all parts of the body, a gastric repairer, and a treatment for stomach and intestinal colic . [9]

#### **IV.12.8. Benefits of essential oil in Matricaria chamomilla :**

- ✓ The inflorescences contain volatile oil ranging between 0,6- 1,2 , which is obtained by distillation with water vapor.
- ✓ Chamomile oil is used in the manufacture of the finest types of perfumes and cosmetics.
- ✓ It is used in the manufacture because oil contains bisabolol and bisabolone of ointments that are useful in treating skin diseases and inflammation. [13]

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# **PRACTICAL PART**

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## Methods and materials used :

This chapter includes the experimental study, which is the quantitative assessment of flavonoids and polyphenols, and the antioxidant activity, as well as the chemical detection of chamomile plant compounds at the level of a laboratory VPRS.

Table 10: Mataterials used

products	Company	purity
FeCl <sub>3</sub>	Biochem chemopherma	99%
AlCl <sub>3</sub>	Honeywell	-
Amonium molybdate	Biochem chemopherma	99 .5%
MeOH	Biochem chemopherma	99%
Galic acid		-
CHCl <sub>3</sub>	Biochem chemopharma	99.9%
Acetat	Biochem chemopharma	-
DPPH	Biochem chemopharma	-
Sulfiric acid	Biochem chemopharma	96%-98%

## Ways and means:

### Plant material:

The chamomile plant is from the compound family

Sample collection:

The sample was collected in late April from Al-Hedjira area

### Sample drying:

First, we separated the plant parts separately, the flowers alone, the leaves and the stems alone

Secondly, the sample was dried by placing it between newspaper leaves, spreading it out, and putting something heavy on it to dry well, in a dark room.

### Soaking:

Then soaked in a solution of methanol water (70%-30%) for 48 hours. Then the samples were filtered using filter leaves.

to leave the sample free of methanol. Extraction with increasingly polar solvents: petroleum ether, chloroform, and butanol. Then it is filtered and separated by phases .

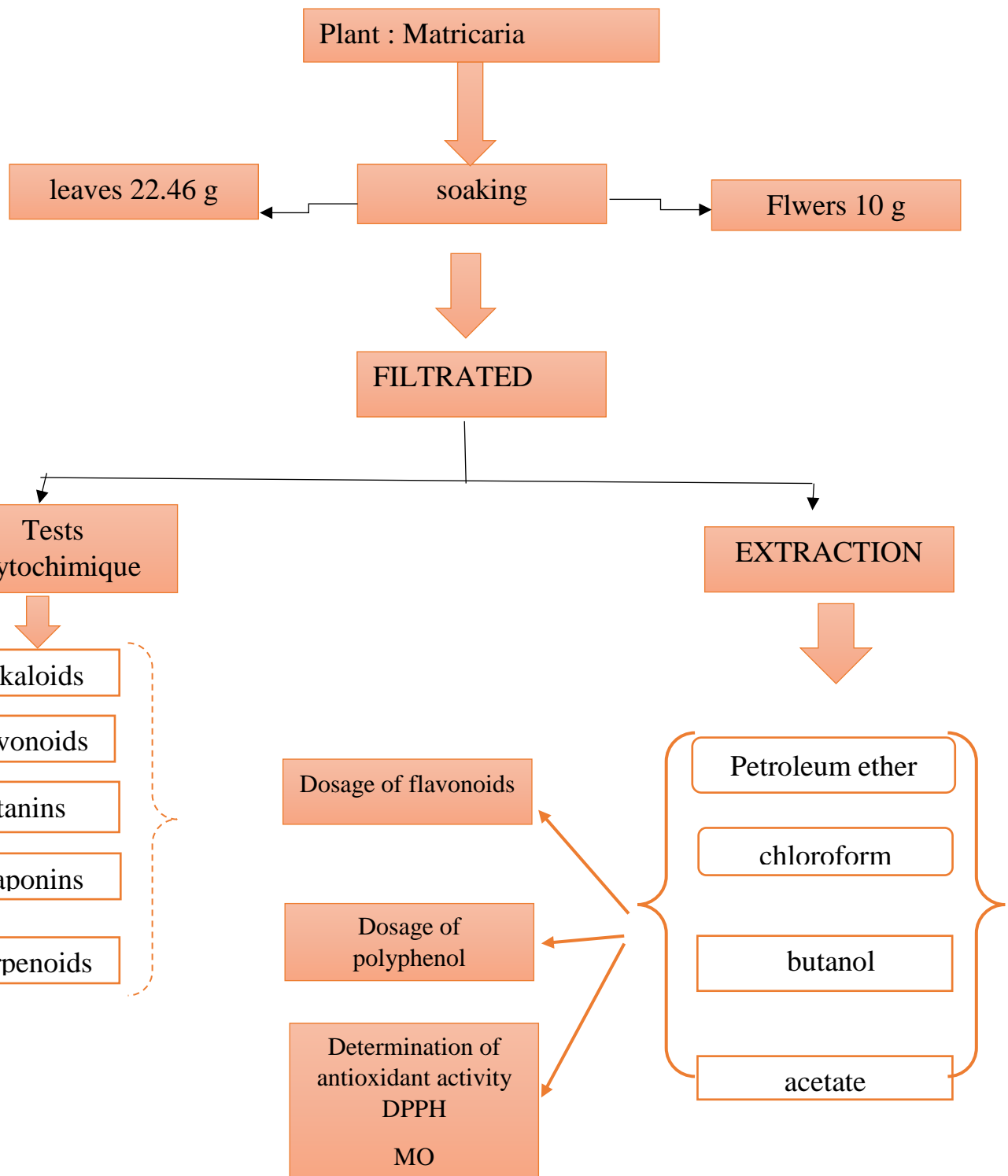


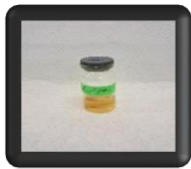



Figure 8 : a schematic illustrating the experimental work in the laboratory

After that evaporated for rotavap , we took samples extracted from each (extract + ether), (extract + butanol), (extract + chloroform), (extract + acetate)



Figure 9: picture of rotavap

Table 11: weight after drying of flowers and leaves(g)



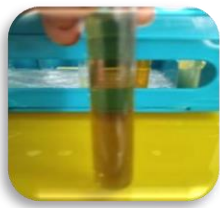
	weight after drying of flowers (g)	Flowers + 7ml Methanol	weight after drying of leaves (g)	Feuille + 7ml Methanol
Ether	0.014		0.071	
Butanol	0.118		0.21	
Chloroform	0.019		0.032	



Acetat	0.043		0.027	
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**Phytochemical examination:**

We performed the initial phytochemical analysis using the aqueous extract of chamomile (crude extract), using a combination of different standard procedures, and the obtained results are translated in the following table.

Table 12: tests phytochimiques

	Detector	the color	
alkaloids	Dragendrov	reddish orange	+++
			
flavonoids	Acetate 10%	yellow	++
			
tannins	FeCl <sub>3</sub>	green	+++
			

saponins	distilled water	 -	-
turbines	Chloroform+ H <sub>2</sub> SO <sub>4</sub>	grayish 	+

+++ : large percentage

++ : average ratio

+ : small percentage

- : not being present

### **Extraction yield :**

The extraction yield is calculated according to the above formula. It is expressed as a function of the dry matter (% MS):

$$R (\%MS) = M_{ex} / M_{mv} * 100$$

R: the yield in %

M<sub>ex</sub>: the mass of the dry extract;

M<sub>mv</sub> : mass of dry plant matter.

Table 13 : Extraction yield

R%	FLOWER	Leaves
chloroform	0.19	0.14
acetat	0.43	0.12
butanol	1.18	0.93

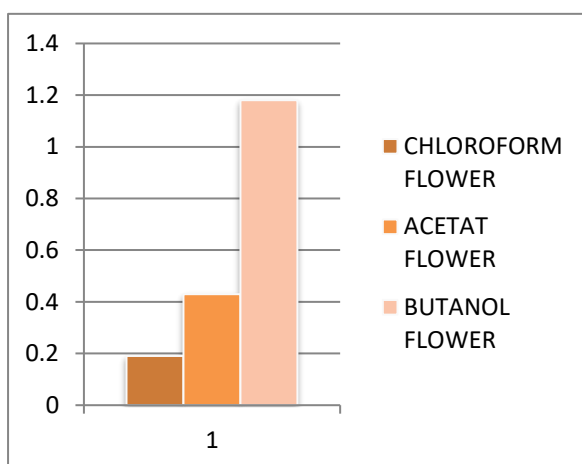


Figure 10: Extraction yield of flowers

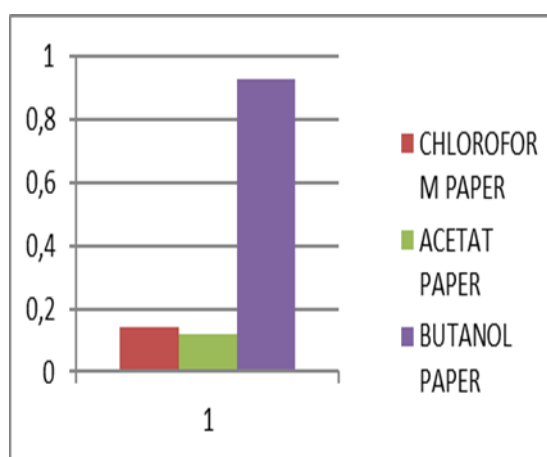


Figure 11: extraction yield of leaves

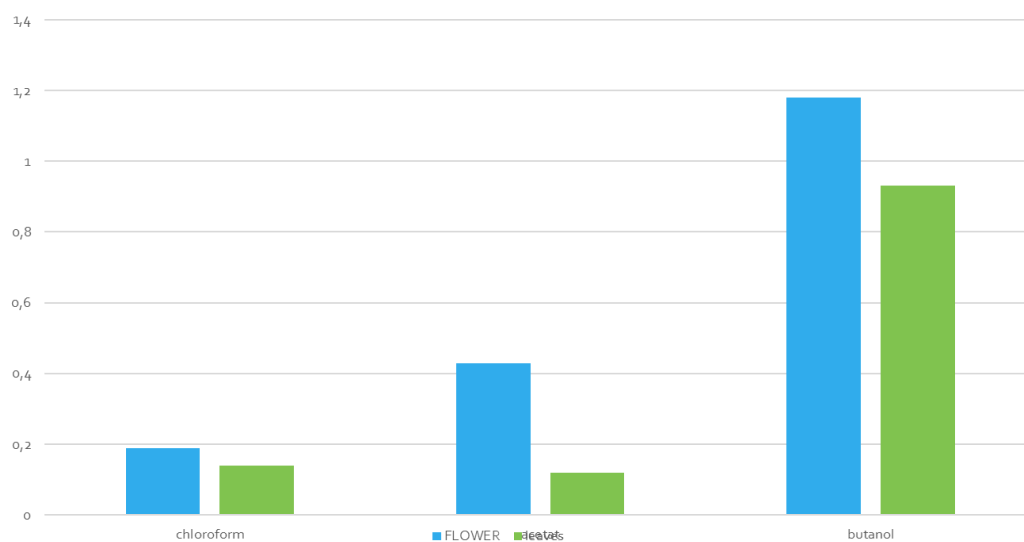


Figure 12 : Extraction yield of leaves and flower

On note que le rendement d'extraction des fleurs est supérieur à celui des feuilles dans les trois extraits

Comme le butanol est le pourcentage le plus élevé dans chacun d'eux, et la différence réside dans les deux autres extraits

Dans les feuilles, le chloroforme est plus élevé que l'acétate

Dans les fleurs, l'acétate est plus élevé que le Chloroforme .

**Essential oil extraction:**

In this method, the Chamomile plant is immersed in water, which will be boiled by the action of the thermal agent via the electronic burner. What distinguishes this method is that there is direct contact between the boiling water and the aromatic substance.

Precautions must be taken to prevent heat from spoiling the aromatic substance. There are aromatic materials from which mucilage gels are produced that may hinder the extraction process, such as cinnamon, so they must be ground well. It is always preferable to carry out experiments in the laboratory on small quantities before starting the production process, by using the Clevenger system shown in the image below. It also extracts a strong-smelling aromatic substance called hydrola.





Figure13: Laboratory image

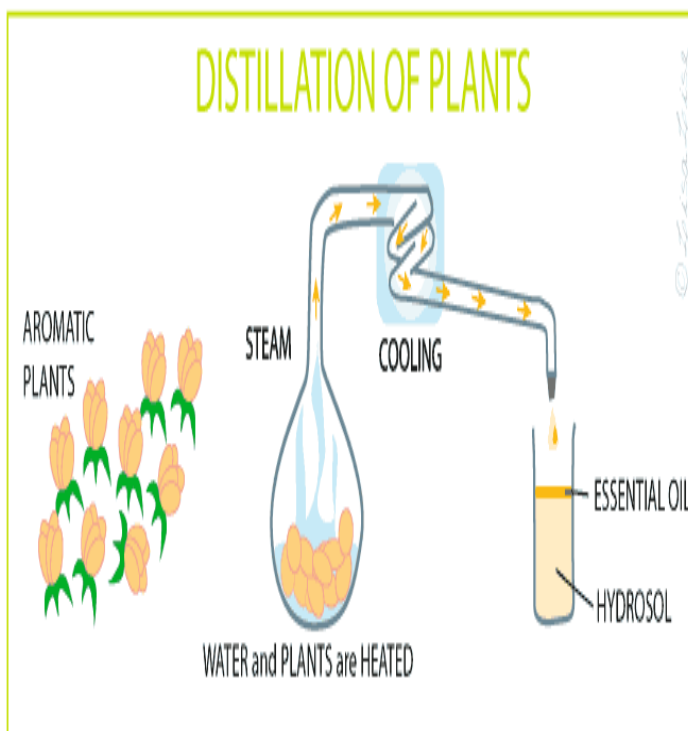


Figure14 : Clevenger diagram  
of the Clevenger apparatus

### Polyphenol Dosage:

The assay of polyphenols was carried out according to the method described by Boizot and Carpenter (2006).

#### Principle :

The determination of total polyphenols using the Folin-Ciocalteu reagent was described in 1965 by Singleton and Rossi. The reagent consists of a mixture of phosphotungstic acid ( $H_3PW_{12}O_{40}$ ) and phosphomolybdic acid ( $H_3PMO_{12}O_{40}$ ). It is reduced during the oxidation of phenols, into a mixture of blue oxides of tungsten ( $W_8, O_{23}$ ) and molybdenum ( $Mo_8, O_{23}$ ). The color produced, whose maximum absorption is between 725 and 760 nm, is proportional to the quantity of polyphenols present in the plant extracts .

It is calculated according to the following law

$$C(\text{mg/g}) = (A/K * F * V/P)$$

C : amount of total phenolic compounds (mg/g)

A: absorbance at 760 nm

K: the slope of the standard curve for Gallic acid

F: expansion coefficient for extracts

V: the dissolved volume of the crude phenolic extract

P: The initial mass of the dry sample

**We Avon prepare:**

Gallic acid (0.3 g in 100 mL distilled water)

Folan (2ml de Volan Mer Dance 25ml distilled water)

Na<sub>2</sub>CO<sub>3</sub>



after to prepare a series of concentrations of gallic acid, we take from each concentration a volume of 0.1 ml and add 0.5 ml of folin and wait 5 minutes to add 2 ml Na<sub>2</sub>CO<sub>3</sub> , then read the absorbance after half an hour in UV spectrophotometer (760 nm)

Table 14: Series volumes taken in the quantification of flavonoids

Concentration (g/ml) of gallic acid	0,1	0,2	0,3	0,4	0,5	0,6	0,7	0,8	0,9	1
Distilled water	0,9	0,8	0,7	0,6	0,5	0,4	0,3	0,2	0,1	0
Extract volume (ml)	0,1	0,1	0,1	0,1	0,1	0,1	0,1	0,1	0,1	0,1

Volume of Folin-Ciocalteu (ml)	0,5	0,5	0,5	0,5	0,5	0,5	0,5	0,5	0,5	0,5	0,5
Volume of Na <sub>2</sub> CO <sub>3</sub> (ml)	2	2	2	2	2	2	2	2	2	2	2

### Estimation of total polyphenols intake TPC :

The amount of phenolic compounds was estimated using the standard curve of Gallic acid as shown in the figure, as the amount of TPC (mg) was calculated on the basis of Gallic acid

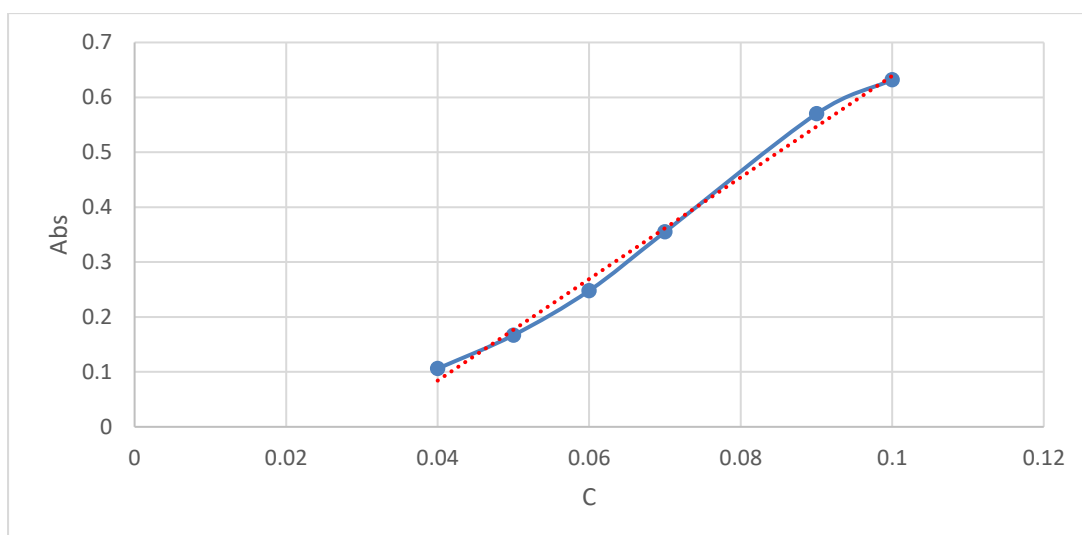


Figure 15 :Standard curve for Galic acid

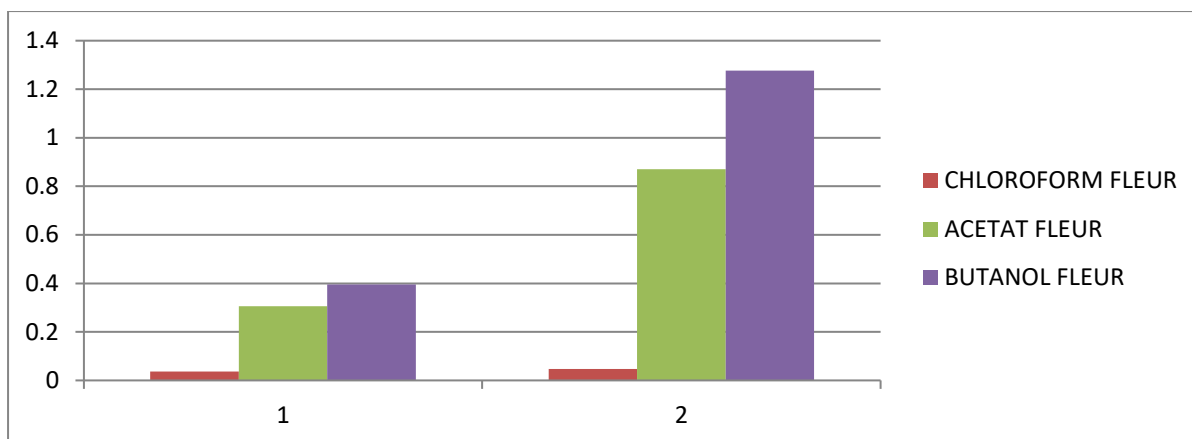


Figure 16: dosage of polyphenol ( flowers) (c mg /g)

Table 15: dosage of polyphenol ( flowers) (c mg /g)

STDev	DOSAGE of POLYPHENOL (c mg /g)
Chloroform ± 0.014	0.031
ACETAT ± 0.14	0.34
BUTANOL ± 0.64	1.65

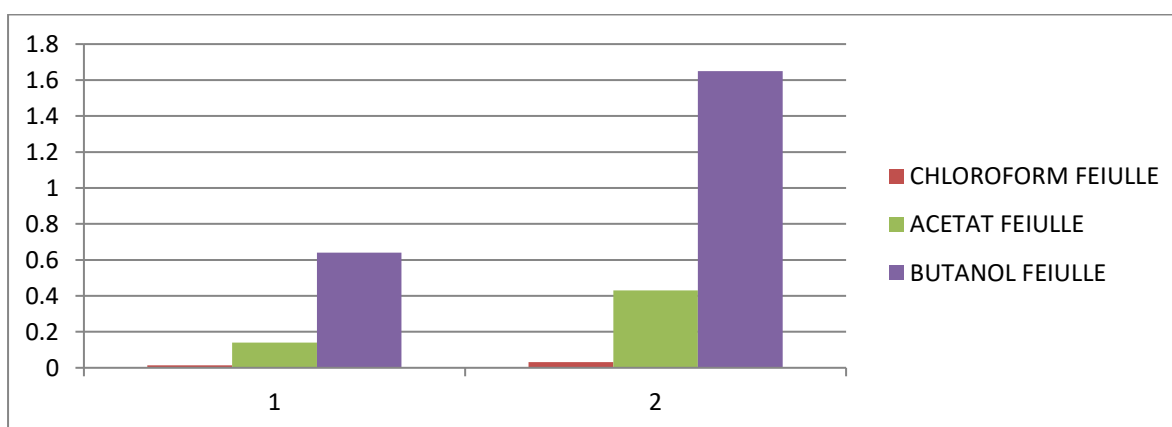


Figure 17: dosage of polyphenol ( leaves) (c mg /g)

Table16: dosage of polyphenol ( leaves) (c mg /g)

stdev	dosage of polyphenol (c mg /g)
chlorform ± 0.036	0.047
ACETAT ± 0.306	0.871
BUTANOL ± 0.395	1.277

Through the results mentioned above, we notice a difference in the amount of TPC from one extract to another, due to the type of solvent and the method of extraction.

The percentage in flowers ranged between  $0.031 \pm 0.014$  to  $1.65 \pm 0.64$  (mg/g).

The percentage in papers ranged between  $0.047 \pm 0.036$  to  $1.277 \pm 0.395$  (mg/g).

Where we recorded the highest value of butanol extract for flowers as well as leaves, which are arranged as follow :

Chloroform(p/f) < acetat (p/f) < butanol (p/f)

### **Flavonoids Dosage:**

The determination of total flavonoids was carried out by the Aluminum trichloride (AlCl<sub>3</sub>) method described by Bahorum et al. (1996) using quercetin as a standard.

### **Principle:**

Aluminum chloride forms stable acid complexes with the C 4 ketone group and with the C-3 or C-5 carbon hydroxyl group of flavones and flavonols. In addition,

aluminum chloride forms acid-labile complexes with orthodihydroxyl groups in ring A or B of flavonoids (Chang et al., 2002)

**We have prepared:**

Queristine ( 20 g in 100 ml distilled water)

AlCl<sub>3</sub> (3.6 g in 100 ml distilled water)

After preparing the series of concentrations, we take 1.5 ml of it, then add AlCl<sub>3</sub> it and wait half an hour and read the absorbance at 430 nm.

**Estimation of flavonoid intake TFC:**

The amount of flavonoids was estimated using the standard curve of Quercetin as shown in the figure, as the amount of TFC (mg) was calculated

It is calculated according to the following law

$$C(\text{mg/g}) = (A/K * F * V/P)$$

C : amount of total flavonoids (mg/g)

A: absorbance at 430 nm

K: the slope of the standard curve for Quercitine acid

F: expansion coefficient for extracts

V: the dissolved volume of the crude phenolic extract

P: The initial mass of the dry sample

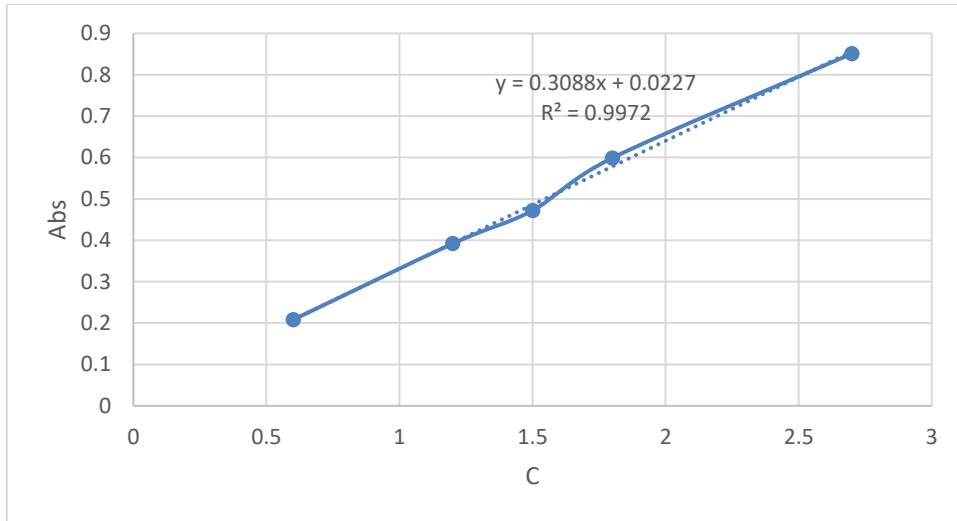


Figure 18: Standard curve for Quercitine

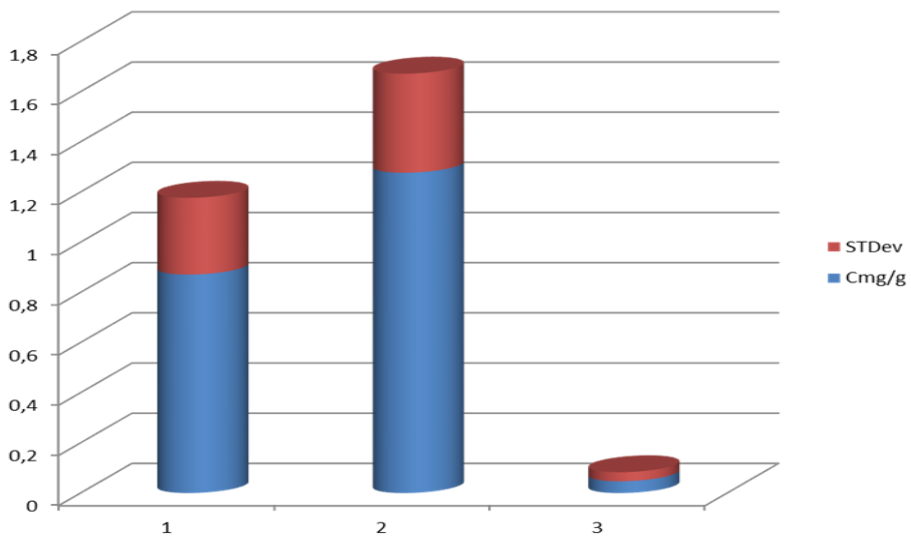


Figure 19: dosage of flavonoids (flowers) (c mg /g)

Table 17: dosage of flavonoids (flowers) (c mg /g)

STDev	DOSAGE of FLAVONOIDS (c mg /g)
Chloroform ± 0.064	0.102
ACETAT ± 0.053	0.239
BUTANOL ± 0.49	1.197

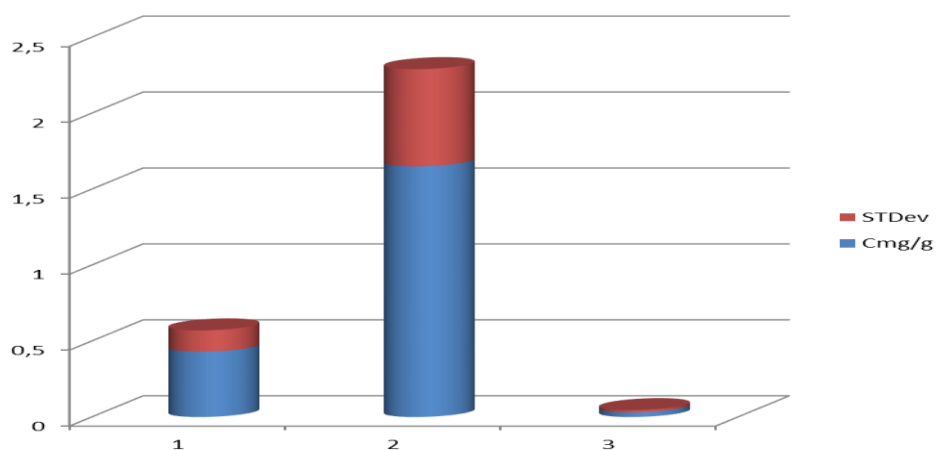


Figure 20: Dosage Of flavonoid(leaves) (c mg /g)

Table 18: Dosage Of flavonoid(leaves) (c mg /g)

StDev	Dosage Of flavonoid (c mg /g)
Chloroform ± 0.017	0.032
ACETAT ± 0.066	0.263
BUTANOL ± 0.248	1.082



Through the results mentioned above, we notice a difference in the amount of TFC from one extract to another, due to the type of solvent and the method of extraction.

The percentage in flowers ranged between  $0.102 \pm 0.064$  to  $1.197 \pm 0.49$  (mg/g).

The percentage in papers ranged between  $0.032 \pm 0.017$  to  $1.082 \pm 0.248$  (mg/g).

Where we recorded the highest value of butanol extract for flowers as well as leaves, which are arranged as follow :

Chloroform(p/f) < acetat (p/f) < butanol (p/f)

### **Determination of total antioxidant capacity:**

**We prepared:**

**Solution1 :** Ammonium molybdate tetrahydrate 2.48 g in 100 ml distilled water

**Solution2 :** Sodium phosphate monobasic dehydrate analytical reagent 17.2 g in 100 ml distilled water

**Solution3 :** Sulfuric acid 16,4 mL in 100 mL of distilled diamond

The above three solutions are placed in a measuring flask in 500 ml and the volume is completed to the standard line with distilled water

after than we shook it for an hour at a moderate temperature

The total antioxidant capacity of plant was evaluated by phosphomolybdenum method.

To 1ml of the extract , added 1ml of reagent solution(0.6Msulfiric acid,28mMsodiumphosphate and 4Mm ammonium molybdate). The tubes were capped and incubated in boiling water bath at  $95^{\circ}\text{C}$  for 90 min. after the samples



Figure 21 : Molybdate preparation

cooled to room temperature , the absorbance of each solution was measured at 695nm against reagent blank using.



Figure22: A picture of the prepared series of molybdate after reading it

### Alternator return test :

It is a rapid test that allows us to measure the antioxidant capacity of the extracts to be studied, as the basic principle of it depends on referring Mo(IV) to Mo (V) by plant extracts that contain antioxidant compounds to form a green complex.

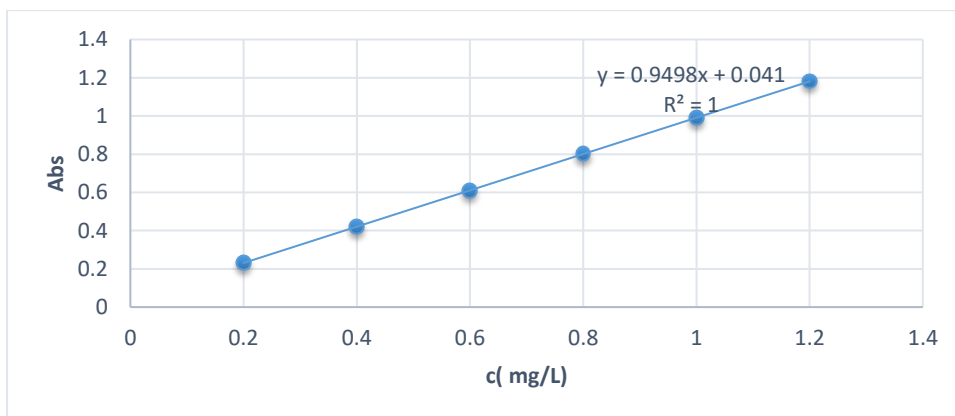


Figure 23: Standard curve for ascorbic acid

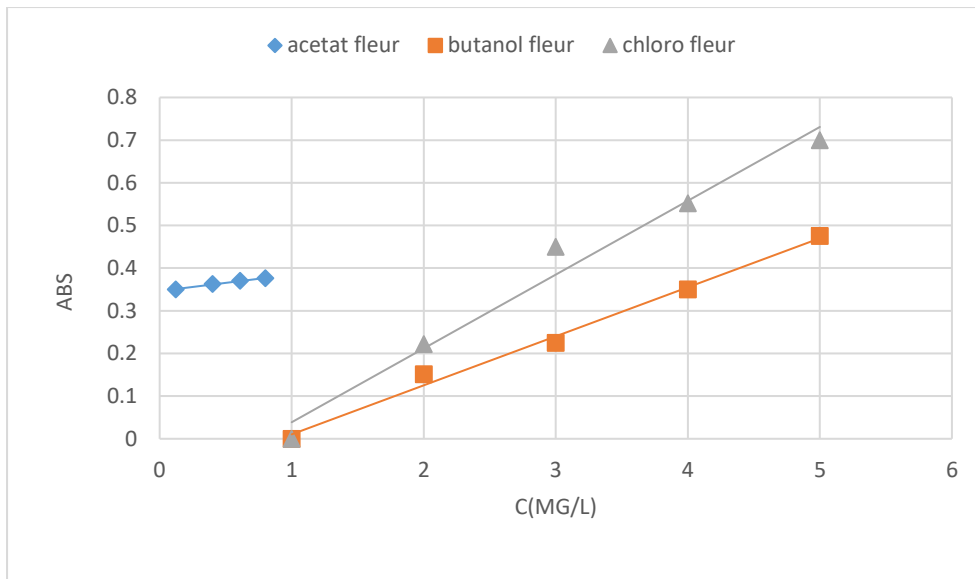


Figure24 : Molybden flower

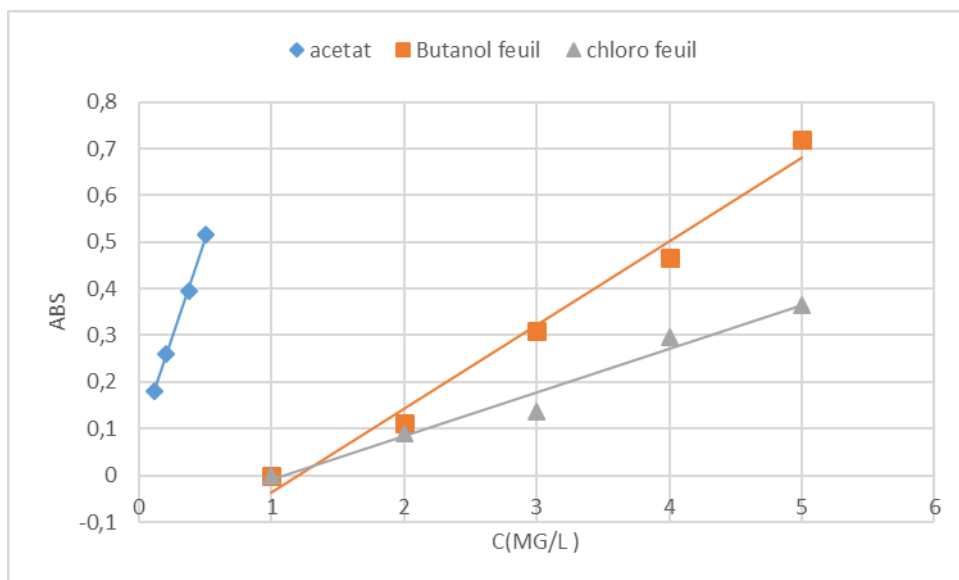


Figure 25 : Molybden leaves

For the flowering and leaf curves, there is a direct relationship between the absorbance and the concentration, as we note that the higher the concentration, the greater the absorbance.

As for the flowering curve, there is a relative increase in the acetate extract compared to the acetate extract of the leaves (significant increase).

The absorbance of flowers with chloroform is higher than that of butanol, in contrast to the leaves, where we find that the absorbance of butanol is higher than that of chloroform.

$$TAC = K/K'$$

TAC : total antioxidant capacity

K: the slope of the extracts curve

K': the slope of the standard curve for ascorbic acid

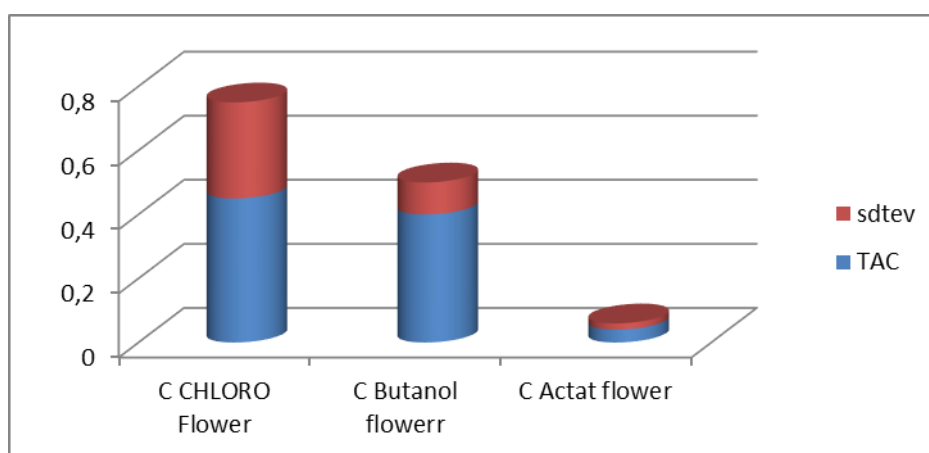


Figure 26 : molybdate flower TAC

Table19: molybdate flower TAC

Extract FLOWER	TAC
Chloroform	0.45 ± 0.3
Acetat	0.04 ± 0.02
Butanol	0.4 ± 0.1

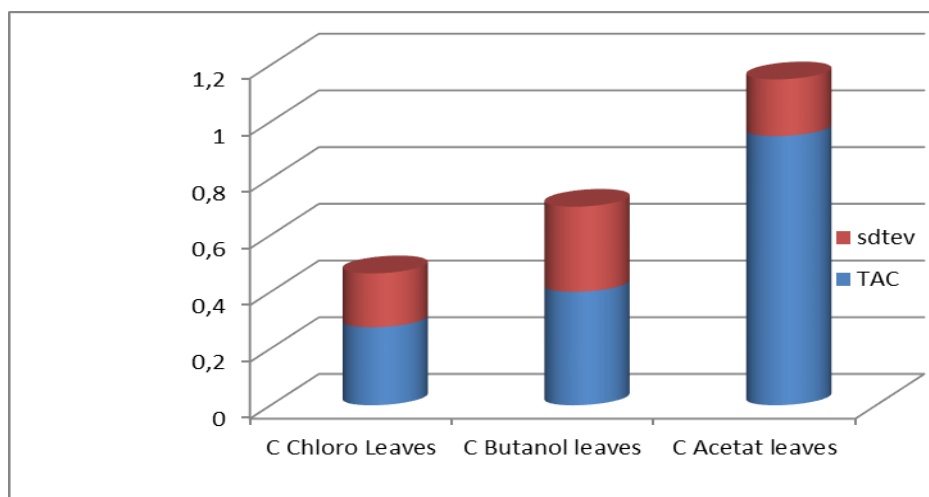


Figure 27: molybdate leaves TAC

Table 20: molybdate leaves TAC

Extract leaves	TAC
Chloroform	0.275 ± 0.1
acetat	0.949 ± 0.2
butanol	0.4 ± 0.3

We note that all extracts have antioxidant capacity, which varies from one extract to another

For flowers, the highest value was in butanol, followed by chloroform and acetate, respectively

For leaves, the highest value is taken by acetate, followed by croform, then butanol

**Standard curve for DPPH:**

DPPH's free radical scavenging activity depends on the ability of 1,1-Diphenyl-2-picryl-hydrazyl (DPPH), a stable free radical, to decolorize in the presence of antioxidants.

Briefly, a DPPH solution was prepared by dissolving 0.01 g in 260 ml MeOH.

Then we prepared a series of solutions of different concentrations of ascorbic acid (0.002g in 100ml distilled water) that were treated with DPPH and left in the dark for 30 min at room temperature, then the absorbance was read at 517 nm.

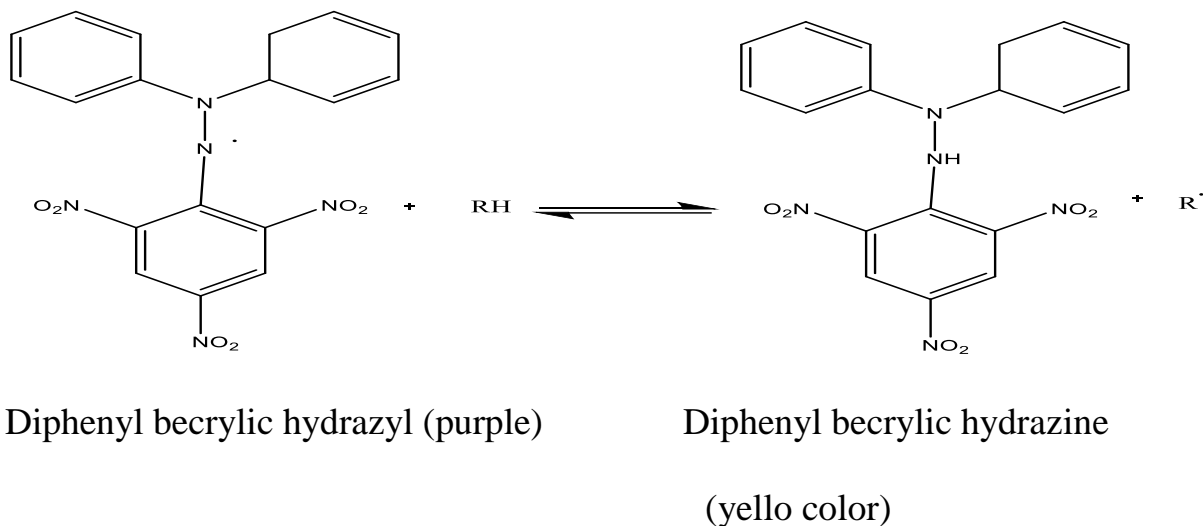


Figure 28: Root oxidation equation

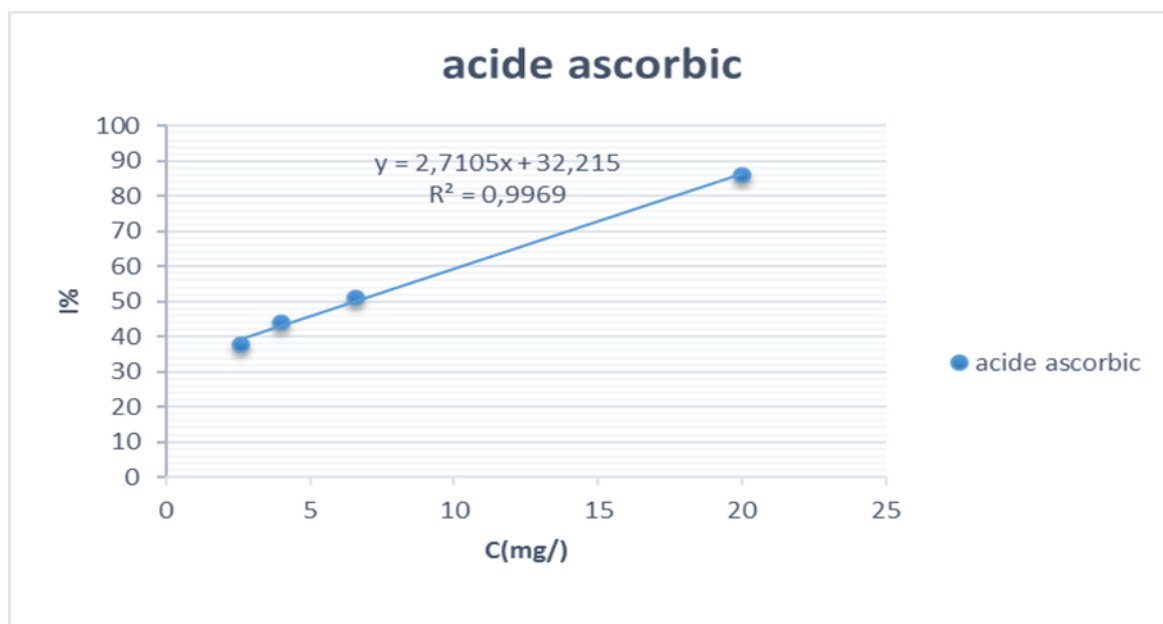


Figure 29 : curve standard of DPPH

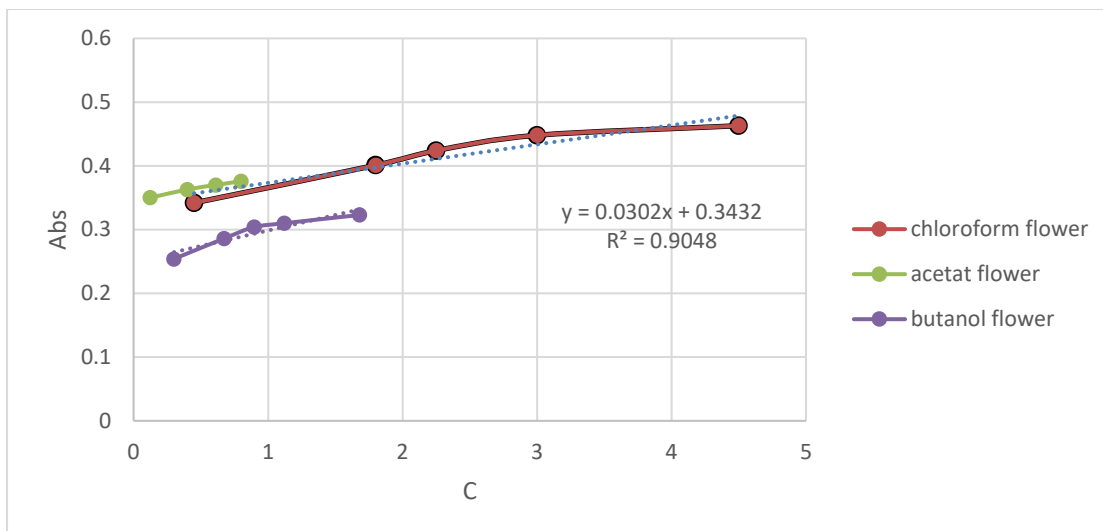


Figure 30 :DPPH OF flower

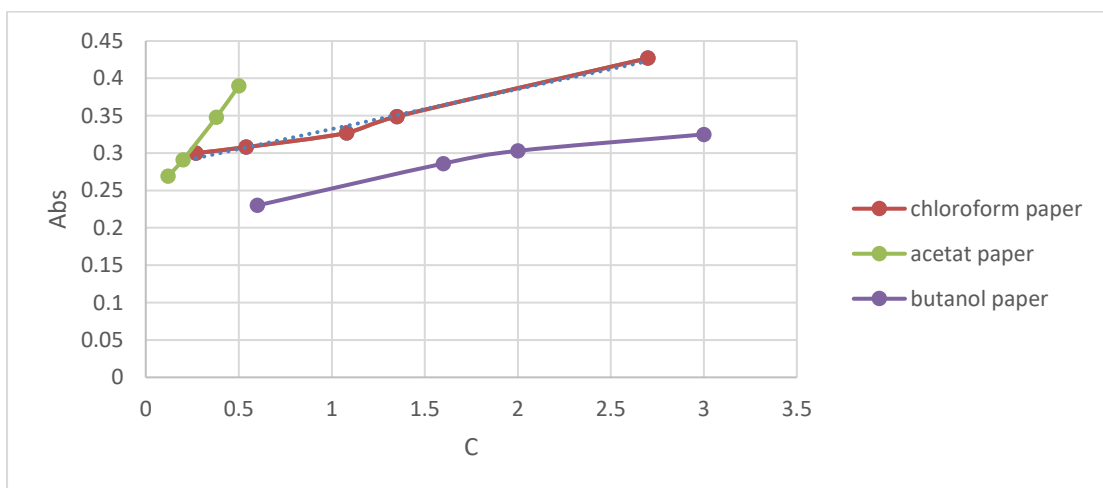
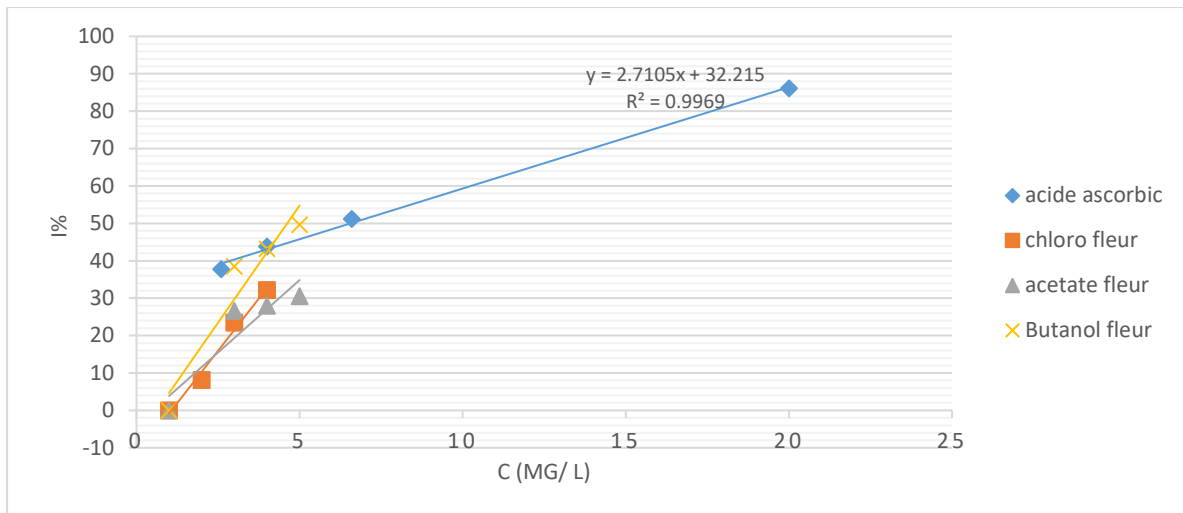


Figure 31 : DPPH of leaves

Through the absorption curves for each of the flowers and papers , we notice that there is a direct relationship between the concentration and absorption of the extracts, arranged as follows :

Abs butanol < Abs chloroform < Abs acetat



FIGUREE 32 : DPPH flowers

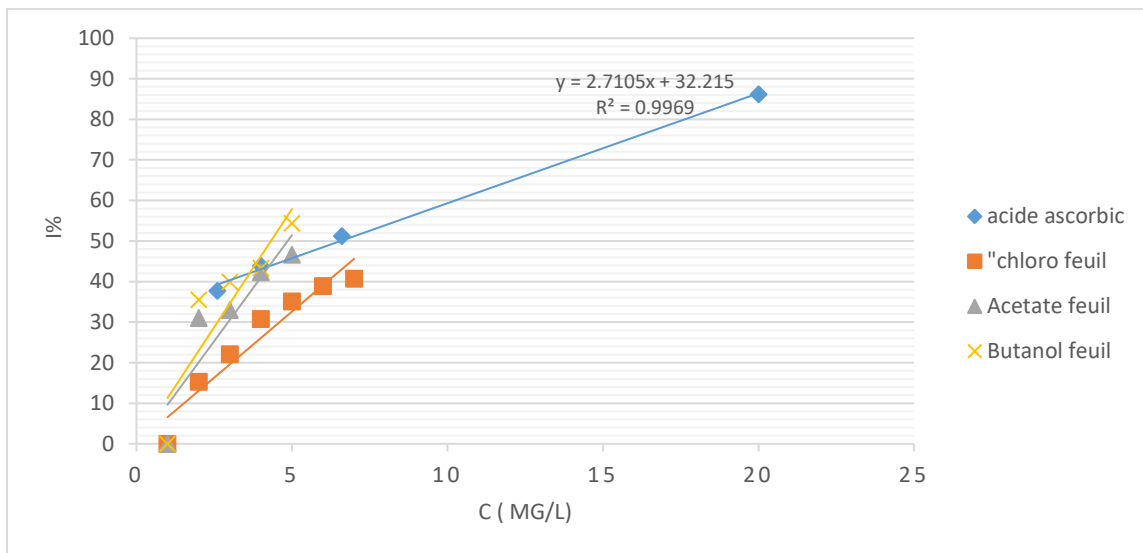


Figure 33: DPPH of leaves

Through the absorption curves for each of the flowers and leaves...we notice that there is a direct relationship between the concentration and absorption of the extracts, arranged as follow



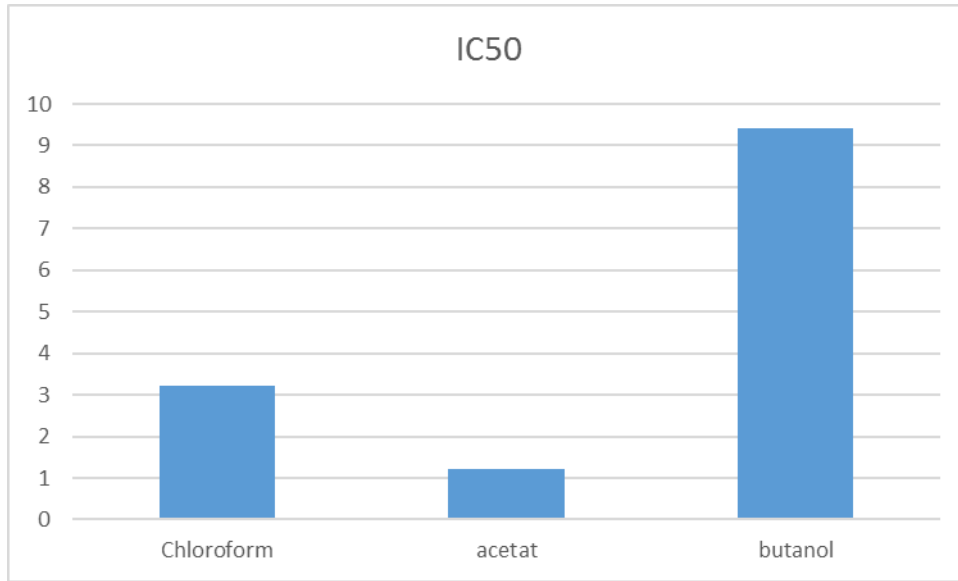


Figure34 : IC50 of leaves

Table 21: IC50 of leaves

EXTRACT LEAVES	IC50
Chloroform	3.22
acetat	1.22
butanol	9.42

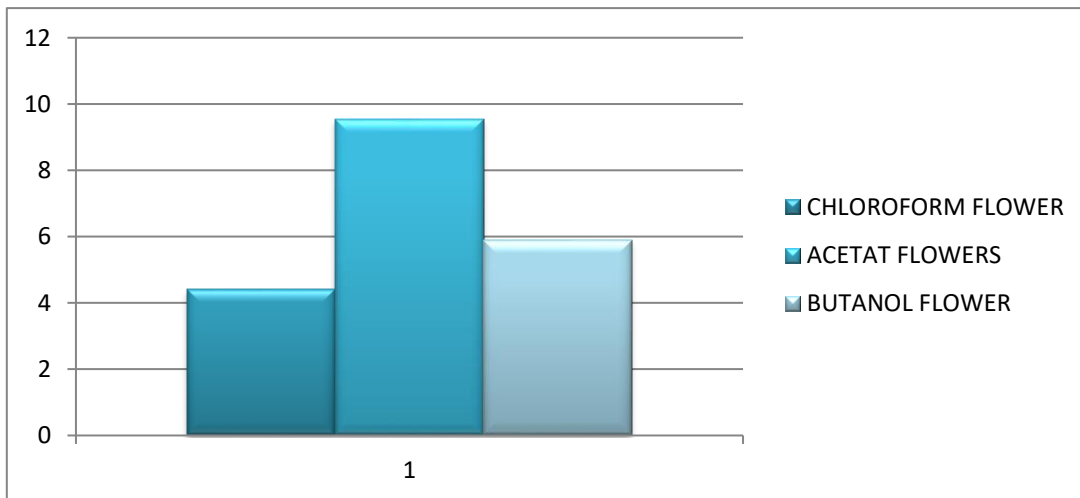


Figure 35 : IC50 OF FLOWERS

Table 22: IC50 OF FLOWERS

EXTRACT Flowers	IC50
Chloroform	4.41
Acetat	9.55
Butanol	5.90

In this study, the antioxidant capacity was evaluated on the basis of the value of IC50 which was recorded in the above tables

For papers, the values ranged between 1.22 – 9.42

The order is as follows

Acetat < chloroform < butanol

For flowers, the values ranged between 4.41 – 9.55

The order is as follows

butanol < chloroform < acetat

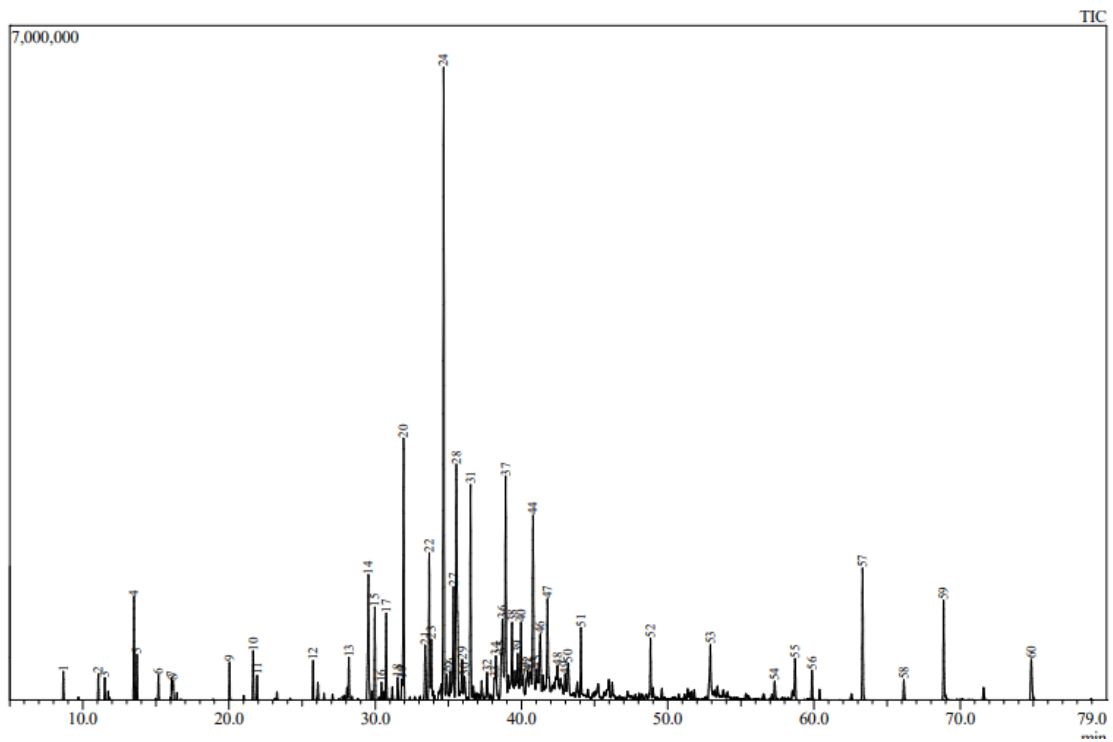
### **Oil analysis by GC/MS :**

For the analysis of volatile compounds (oils) a system is used in which there is a trap so that the sample can be impounded and P&T can be used to enter the samples

The lysed targets are extracted, mixed with water, and entered into a sealed chamber in the presence of an inert gas such as nitrogen (N<sub>2</sub>) . A bubbling product is pumped through the water.



Figure 36 : device of GC /MS



Tabel 23 : riselt of GC /MS

Number	Compounds	Area %	Heigh%
1	Bicyclo(1.3.0)hexane,4-methylene-1-(1-methyl)	0,66	0,80
2	1,3cyclohexane, 1-methyl-4-(1-methylethyle)	0,60	0,73

3	o-Cymene	0,53	0,65
4	gamma,-Terpinene	2,11	2,52
5	1,5Heptadien-4-one, 3 ,3,6-trimethyl	0,95	1,18
6	4-isopropyldiene-1-cyclohexene	0,57	0,69
7	Thujone	0,41	0,53
8	Butanoic acid,2-methyl,2-methylbutyl ester	0,49	0,49
9	3-cyclohexene-1-ol,4-methyl-1-(1-methylethyl)	0,82	0,99
10	Hexanal,3,3-dimethyl-	1,21	1,24
11	2-methylpentyl isovalorate	0,45	0,63
12	5-(1-iodo-1-methyl-ethyl)-3,3-dimethyl-dihydroxy	0,79	1,04
13	Cyclohexene,4-ethyl-4-methyl-3-(1-methylene)	0,90	2,98
14	2,6,10-dodecatrien-1-ol, 3,7,11-trimethyl,-acetate	3,62	2,24
15	Copaene	1,97	0,44
16	2,6-octadien-1-ol,3,7-dimethyl-acetate,(E)-	0,36	2,01
17	1-methyl-1-ethenyl-2,4-bis(I –methylethenyl)c	1,69	0,59
18	1-H-cycloprop(e)azulene 1a, 2,3,4,4a,5,6,7b octene	0,59	0,52
19	Sabinyyl isobutanoate	0,44	6,12
20	Caryophyllene	5,54	1,26
21	Isoledene	1,51	3,36
22	(E) -beta,-famecene	2,82	1,31
23	Chrysantenyl 2-methylbutanoate	1,17	14,57
24	(-)-Germacrene D	13,79	0,48
25	Beta-oplopenone	0,51	0,58
26	1-Naphthalenol,1, 2,3,4,4a,5,6,7,8a,-octahydro-1	0,48	2,55
27	(1S,2 <sup>E</sup> ,6 <sup>e</sup> ,10 R)-3 ,7,11,11,-tetramethylbicyclo	2,57	5,35
28	,beta,-cadinene	7,56	0,77
29	5-oxatricyclo(,8,2,0,04, 6)dodicane,4 ,12,12-tr	0,74	0,41
30	Bicyclo (2,2,1)heptane,7,7,-dimethyl-2-methylene	0,42	4,97

31	Murolan-3,9(11)-diene-10-peroxy	4,57	0,47
32	5,9-Undecadien-1-yne,6,10-dimethyl-	0,39	0,41
33	1H-cycloprop(e)azulen-7-ol,decahydro-1,1,7	0,42	0,92
34	5-oxatricyclo(8,2,0,04,6)dodecane,4,12,12-tr	0,97	0,97
35	Salvial-4(14)-en-1-one	0,90	1,75
36	Ledol	2,07	5,02
37	Spirogatamol	5,10	1,41
38	Pentadecanal	1,19	0,85
39	Beta,-oplopenone	0,98	1,61
40	Ethane-1,2-diyl bis(2-methylbetanoate)	1,74	0,42
41	Isospathulenol	0,43	0,48
42	10,10-dimethyl-2,6-dimethylenebicyclo(7,2,0)	0,76	0,45
43	,tau,-Murolol	0,45	4,06
44	,alpha,-cadinol	4,74	0,52
45	1(2H)-naphtalenone,octahydro-4a,8a-dimetl	0,65	1,33
46	(1R,7S,E)-7-isopropyl-4,10-dimethylenecyclo	2,05	2,06
47	Cyclohexanol,3-ethenyl-3-methyl-2-(1-methyl	2,27	0,38
48	Pentadecanal-	0,38	0,41
49	2-pentadecanone,6,10,14-trimethyl-	0,38	0,70
50	n-hexadecanoic acid	0,99	1,57
51	2-methyltetracosane	1,31	1,33
52	Linolyl acetate	1,07	1,15
53	Androst-5,16-diene-3-beta,-ol	1,52	0,39
54	Tetracosane	0,38	0,90
55	Tetracosane	0,88	0,68
56	Hexatracontane	0,60	3,04
57	Hexatriacontane	2,70	
58			

59			
60			

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# **Conclusion**

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In our study we grew chamomile and it worked

This is contrary to expectations given the harsh nature of the desert

As the tests were carried out on them, we obtained the compounds (flavonoids, alkaloids, terpenes...etc).

We also performed antioxidant tests of the extracts, and n-butanol and acetate showed greater efficacy on dpph, TPC, and TFC. As for TAC, the potency differed between flowers and leaves in each extract.

Given these results and the economic aspect of the studied plant, we hope to expand the cultivation of it and various other medicinal plants in the Algerian desert.