PEOPLE'S DEMOCRATIC REPUBLIC OF ALGERIA

MINISTRY OF HIGHER EDUCATION AND SCIENTIFIC RESEARCH

UNIVERSITY OF ECHAHID HAMMA LAKHDAR - EL OUED

FACULTY OF EXACT SCIENCES

DEPARTMENT OF CHEMSTRY

Dissertation Submitted in Partial Fulfillment of the Requirements for Master's Degree in:

Field: Chemistry

Specialty: Organic Chemistry

THEME

Investigation of extracted bioactive compounds from local medicinal plant and theirs applications in preventive therapy from drugs addiction

Discussed on June 26th, 2024

Presented By:

- **Rahil Benamor**
- **Ouissam Abid**

Before the discussion Committee:

Pr. Abdelkrim Rebiai	**President**	**University of El Oued**
Pr. Ahmed Khalifa Chemsa	**Examiner**	**University of El Oued**
Dr. Kamel Layes	**Representative of the economic partner**	**Addiction treatment center**
	Representative of the incubator	**University of El Oued**
Dr. Larbi Haddad	**Supervisor**	**University of El Oued**

Academic year: 2023/2024

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Summary

This dissertation investigates the presence and potential therapeutic applications of alkaloids in Lavender and Ephedra plants. Through a detailed botanical and chemical analysis, various extraction and testing methods were employed to identify the alkaloid presence in these plants. Lavender showed a higher qualitative content of alkaloids compared to Ephedra, confirmed by Mayer's, Wagner's, and Dragendorff's tests. Lavender's alkaloids exhibited significant antioxidant, antimicrobial, and antidiabetic effects more than Ephedra's study. These findings suggest that the alkaloids in these plants may hold potential for developing new treatments for addiction, highlighting the importance of further research into their bioactive compounds.

Keywords: Lavender, Ephedra, alkaloids, phytochemistry, therapeutic potential, extraction methods, biological activities.

ملخص

تبحث هذه الدراسة في وجود القلويدات والتطبيقات العلاجية المحتملة في نبتتي الخزامى و العلندة. ومن خلال تحليل نباتي وكيميائي للمستخلصات ، أظهرت نبتة الخزامى محتوى نوعي أعلى من القلويدات مقارنة بالعلندة ، وهو ما أكدته اختبارات ماير، فاجنر، و دراجندورف. أظهرت قلويدات الخزامى تأثيرات كبيرة مضادة للأكسدة ومضادة للميكروبات ومضادة لمرض السكر أكثر منها مقارنة بنبتة العلندة. وتشير هذه النتائج بالإضافة إلى بعض الدراسات إلى أن القلويدات الموجودة في هذه النباتات قد تحمل إمكانية تطوير علاجات جديدة للإدمان ، مما يسلط الضوء على أهمية إجراء مزيد من البحوث في مركباتها النشطة بيولوجيا.

الكلمات المفتاحية: الخزامى، العلندة، القلويدات، الكيمياء النباتية، الإمكانات العلاجية، طرق الاستخلاص، النشاطيات البيولوجية.

الاهداء

(و آخر دعواهم أن الحمد لله رب العالمين)

"الحمد لله ما تم جهد ولا ختم سعي الا بفضله ، وما تخطيت هذه العقبات والصعوبات الا بتوفيقه "

الى من كلله الله بالهيبة والوقار ..الى من علمني العطاء.. الى من أحمل اسمه بكل افتخار ..

الى من قام بكل التضحيات .. **والدي الغالي** .

الى ملاكي في الحياة .. الى معنى الحب والى معنى الحنان ..الى بسمه الحياة وسر الوجود

الى من كان دعائها سر نجاحي وحنانها بلسم جراحي الى من كانت الداعم الأكبر لي الى ست الحبايب .. **والدتي الحبيبة** .

الى من كانا مصدر قوتي ..سنداااي الغاليين ..**عبد القادر . محمد نافع** دمتما لي سندا لا يميل .

الى من كان له جهدا وتعبا معي في كل المراحل والاوقات ..توأم روحي ..**وجيه** .

الى سندي الصغيير ..منبع سعادتي .. **فيض الله .**

الى النعمة التي طالما انتظرتها ..رفيقات دربي .. اخواتي **.. ولاء .راما** .

الى من انعقد العهد بين قلبي وقلبه .. الى من اصطفاه الله من بين الرجال ان يكون خطيبا لي وشريكا في الحياة .. **صلاح الدين** .

الى من اسكنها الله فسيح جنانه ..**جدتي الغالية** .

الى اخت الدراسة والمواقف الصعبة وشريكتي في المذكرة والتي كان لها دور كبير في اتمامها ودعمها اللامحدود .. **وسام** .

الى من كان معهن لحظات سعيدة ..وذكريات جميلة ..صديقات العمر وانيسات القلب والروح **ريحانه .هنيه . وصال. هديل. اماني**

الى من كان لها دعم ومساعدة ..الى العضوة الجديد في العائلة .. **انفال**

أهدي هذا العمل الذي ما كان ليكتمل بدونكم.

رحيل

إهداء

إلى من كان لهم الفضل بعد الله في كل خطوة خطوتها نحو تحقيق أحلامي وطموحاتي، إلى من تعبوا وسهروا من أجلي

إلى والدتي العزيزة **فيروز**، التي كانت دوماً مصدر الدعم والحنان

ووالدي الحبيب **الحكيم محمد** الذي كان لي القدوة والمثل الأعلى في الصبر والإصرار

الى ابي الثاني الذي كان دوره عظيم في اكمال مسيرتي الدراسية **عمي نصر الله**.

إلى إخوتي الأعزاء **مسعود** و**فراس** و**إياد**، الذين وقفوا بجانبي في كل مراحل دراستي، وحموني ودعموني بكل ما يملكون.

إلى مؤنستي الصغيرة **ميس**، التي كانت بسمة طفولتها تضفي على أيامي سعادة لا توصف.

إلى صديقتي الوفية **لينا** التي كانت دوماً السند والعون في كل الأوقات

وإلى صديقتي المقربة ،رفيقة دربي الجامعي، **رحيل**، التي كانت شمعة تضيء لي دربي، وأختاً لم تلدها أمي، ورفيقةً في كل لحظة من مشواري الدراسي مثالاً للصديقة المثالية بحبها وإخلاصها ودعمها اللامحدود ..

لكم جميعاً أقدم هذا العمل، فهو ثمرة جهودكم ودعمكم المستمر. شكراً من القلب.

Acknowledgments

We would like to express our sincere gratitude to all those who have supported and guided us throughout the completion of this dissertation.

First and foremost, we would like to thank our advisor, Dr. Larbi Haddad, for his invaluable guidance, continuous support, and encouragement throughout our research. His insightful feedback has been instrumental in shaping this work.

We are also deeply grateful to the members of our dissertation committee, Pr. Abdelkrim Rebiai (President) and Pr. Ahmed Khalifa Chemsa (Examiner) for their valuable suggestions and critical insights, which have significantly contributed to the improvement of this dissertation.

Special thanks to the faculty and staff of the Department of [Department Name] at [University Name], for providing the necessary resources and a conducive environment for our research. We also extend our gratitude to the laboratory technicians, especially [Technician's Name], for their assistance with the experimental work.

We are profoundly thankful to Dr. Kamel Layes, An employee at the Addiction Treatment Center in Al-Wadi, for all the information he provided us, as well as the directions and instructions related to the purpose of our research.

Thank you all for believing in us and helping us achieve this milestone.

List of Tables

List of Figures

List of Abbreviations

Symbol or abbreviation	Significance
R-N	**Rhoeadine**
Ac	**Absorbance intensity of DPPH in the absence of the sample**
As	**Absorbance intensity of DPPH in the presence of the sample**
$BiONO_3$	**Bismuth subnitrate**
CN	**Gentamicin**
DPPH	**2,2-diphenyl-1-picrylhydrazyl**
I %	**Inhibition Percentage**
IC50	**Inhibitory concentration 50%**
$KBiI_4$	**Bismuth iodide complex**
RWAL	**Raw extract of nightshade plant**
RWKHZ	**Raw extract of lavender plant**
T0, T15, T30, T60, T120	**Time intervals in minutes**

Tables of Contents

General Introduction

The investigation of plant extracts for their potential therapeutic applications has garnered considerable attention in recent years. Among the diverse classes of phytochemicals, alkaloids have stood out due to their significant pharmacological activities, which include analgesic, antimalarial, anticancer, and antimicrobial properties. This dissertation focuses on the botanical and chemical study of Lavender and Ephedra plants, with a particular interest in detecting and analyzing their alkaloid content. The ultimate aim of this research is to explore the possibility of using specific alkaloid molecules from these plants in drug addiction therapy, thereby proposing a novel approach to addiction treatment [1, 2].

Significance of the Study

The exploration of plant extracts for medicinal purposes is a field of enduring relevance and innovation. By focusing on Lavender and Ephedra plants, this dissertation aims to contribute to the understanding of their chemical composition and potential therapeutic applications. The detection of alkaloids in these plants and the evaluation of their biological activities provide a basis for further research into their possible use in drug addiction therapy. This study not only enhances the knowledge of phytochemical constituents of Lavender and Ephedra but also opens new avenues for their application in modern medicine.

Chapter Overview

The first chapter provides a comprehensive botanical study on Lavender and Ephedra plants. Lavender, widely recognized for its aromatic and therapeutic properties, has been used traditionally for its sedative and calming effects [3]. Ephedra, on the other hand, is known for its stimulant properties and has been used in traditional medicine for the treatment of asthma and bronchitis [4]. This chapter delves into the morphological characteristics, distribution, and traditional uses of these plants, establishing a foundation for the subsequent chemical analyses.

The second chapter addresses the various methods employed for the extraction of plant alkaloids. The extraction techniques discussed include maceration, Soxhlet, and Clevenger methods, each with its own advantages and limitations depending on the nature of the plant material and the desired compounds [5]. Rapid tests such as Mayer, Wagner, and Dragendorff reagents are used to qualitatively detect the presence of alkaloids in the extracts [6]. Additionally, this chapter explores the biological activities of the extracts, including antioxidant activity through DPPH assay, antidiabetic potential, and antimicrobial effects, highlighting the multifaceted applications of these plant-derived compounds.

Chapter three transitions into the experimental realm, detailing the materials and chemicals used, the sampling of plants, and the specific protocols followed for extraction and testing. This section provides a meticulous account of the experimental procedures, ensuring reproducibility and transparency in the research methodology. The protocols for rapid tests and biological activity assays are described in detail, underscoring the rigorous approach taken to validate the presence and efficacy of alkaloids in the plant extracts.

The fourth chapter presents the results and discussion, interpreting the findings of the experimental work. This chapter not only reports the outcomes of the alkaloid detection tests and biological assays but also discusses their implications in the context of existing literature. The potential of total alkaloids – as candidates for drug addiction therapy – is considered, with a cautious interpretation to maintain the scientific rigor of the study. The discussion integrates the botanical, chemical, and biological data, offering a holistic view of the research findings.

In conclusion, this dissertation offers a systematic approach to investigating the alkaloid content of Lavender and Ephedra plants, utilizing both theoretical and experimental methodologies. The integration of botanical studies, extraction techniques, rapid tests, and biological activity assays forms a comprehensive framework for the evaluation of these plants' therapeutic potential. The findings of this research may pave the way for future studies aimed at developing new strategies for addiction treatment, thereby underscoring the importance of plant-derived alkaloids in medical research.

1. Cordell, G. A. (1981). Introduction to Alkaloids: A Biogenetic Approach. Wiley.
2. Dewick, P. M. (2002). Medicinal Natural Products: A Biosynthetic Approach. Wiley.
3. Cavanagh, H. M. A., & Wilkinson, J. M. (2002). Biological activities of Lavender essential oil. Phytotherapy Research, 16(4), 301-308.
4. Cooper, C. E., & Johnson, J. E. (1984). The Use of Ephedra in Traditional Medicine. Journal of Ethnopharmacology, 11(3), 337-342.
5. Handa, S. S., Khanuja, S. P. S., Longo, G., & Rakesh, D. D. (2008). Extraction Technologies for Medicinal and Aromatic Plants. United Nations Industrial Development Organization and the International Centre for Science and High Technology.
6. Wagner, H., Bladt, S., & Zgainski, E. M. (1984). Plant Drug Analysis: A Thin Layer Chromatography Atlas. Springer.

Theoretical Section

Chapter I

Studied Plants

Lavandula

1. Botanical Study on Lavender Plant:

1.1. A Historical Overview of Lavender Plant:

Among the plants of the Lamiaceae family, which have been known since the time of the ancient Egyptians and used in mummification, the lavender plant is also known as "lavender", derived from the Latin word "lavare", meaning "to wash". Some suggest that the name "lavender" may derive from the Arabic word "khazam", meaning "to gather", due to its ability to attract bees and butterflies. Lavender is considered a perennial plant that can live up to 20 years under suitable conditions, and it is classified along with rosemary and basil in the mint family [1] .

1.2. The classification of the lavender plant:

The botanist Philip Miller classified the lavender plant in 1768, as illustrated in the table 01.

Table 01: Classification of the lavender plant [2]

Kingdom	Plantae
Division	Magnoliophyta
Class	Magnoliopsida
Order	Lamiales
Family	Lamiaceae
Genus	Lavandula

1.3. Common Names for Lavender Plant:

Scientific Name: Lavandula

English Name: Lavender

French Name: La lavande.

1.4. Botanical Description of Lavender Plant:

Figure 01: Lavender flowers with bracts

1.4.1. Leaf:

Spear-shaped, with entire margins, grayish-green in color [3] .

Figure 02: Lavender Plant Leaf

1.4.2. Flower:

The lavender flower consists of five delicate petals that elegantly interlace with each other, resembling the shape of a butterfly. Their colors vary between light purple and dark blue, with some species being white or pink [3] .

Figure 03: Lavender Plant Flower

1.4.3. Seeds:

Lavender seeds are very small, not exceeding a diameter of 2 millimeters. They are dark brown or black in color and have a rough, uneven surface. They resemble poppy seeds in shape and size[3] .

1.5. Geographical Distribution of Lavender Plant [4] :

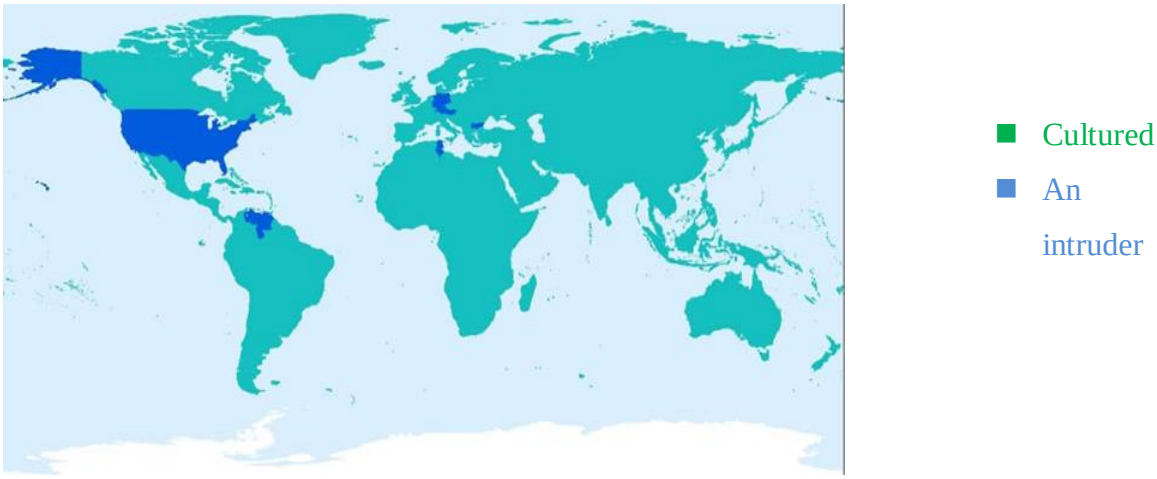


Figure 04: Geographical Distribution Map of the Lavender Plant

1.6. The Economic Significance of the Lavender Plant:

Recent statistics indicate that the global market value of lavender is estimated at around $115 billion, underscoring the significance of this crop on a worldwide scale. While France was previously the leader in lavender production, Bulgaria has surpassed it in recent years.

Currently, Bulgaria holds the lion's share of lavender production, accounting for 52% of the total global output. France follows with 26%, and then China with 12%. The remaining countries contribute only 10% of the global production, with most of them located in Ukraine, Spain, and the Arab Maghreb [5] .

These numbers illustrate the recent developments in the lavender industry on a global scale, with Bulgaria experiencing tremendous growth in its production. This reflects shifts and changes in global demand and economic dynamics in this vital sector [6] .

1.7. The Chemical Composition of the Lavender Plant:

The chemical composition of lavender plant includes several compounds, among the most prominent are:

Linalool: This major aromatic compound constitutes a significant portion of lavender oil and is responsible for the plant's pleasant scent.

Linalyl acetate: This compound is considered a key component in lavender oil, contributing to its distinctive aroma.

Benzene: An organic compound found in small amounts in lavender plants.

Camphor: This compound represents a small part of the lavender's composition and possesses anti-inflammatory and soothing properties.

Cineole: Another compound found in the essential oils of lavender plants, credited with some important medicinal benefits [7].

Ephedra

2. Botanical Study on Ephedra Plant:

2.1. Historical Overview of Ephedra Plant:

The Ephedra plant belongs to the genus Ephedra, which falls under the Ephedraceae family of the Gymnosperm phylum. Ephedra, along with its varieties such as Ephedra distachya, Ephedra fragilis, Ephedra alata, and Ephedra helvetica, constitutes a type of plant known as the Ephedraceae family. This family comprises only one genus, Ephedra, which includes approximately 51 different species. These plants have been historically utilized in ancient

civilizations such as the Greeks and Romans for culinary purposes and traditional medicine. The origin of the word "Ephedra" is derived from the Arabic language, with its etymological root "Aln" meaning "resembling" or "entering into something". Ephedra plants are distributed across various regions worldwide, including temperate and tropical zones, and have played significant roles in cultural and medicinal practices [8].

2.2. Classification of Ephedra Plant:

The botanical classification of the Ephedra plant is as follows table 2 [9].

Table 02: Classification of Ephedra Plant

Kingdom	Plantae
Division	Gnetophyta
Class	Gnetopsida
Order	Ephedrales
Family	Ephedraceae
Genus	Ephedra

2.3. Common Names of the Ephedra Plant [10] :

Scientific Name: Ephedra alata

French Name: Éphédrine

English Name: Ephedra

2.4. Botanical Description of the Ephedra Plant:

Ephedra is a perennial shrub characterized by short stems, often appearing as bushes, creeping, or climbing plants. It typically grows near prickly pear and olive trees, featuring numerous branches and dense foliage. Branch lengths can reach approximately one meter. The plant is distinguished by its greenish-yellowish hues. Ephedra branches are primarily used in pharmaceutical manufacturing, in addition to the potential utilization of its thick tubular roots and leaves. Ephedra fruits are cone-shaped, and its extracts have an acceptable taste [11].

Figure 05: Ephedra Plant

2.5. Geographic Distribution of Ephedra Plant:

The geographical distribution of the Ephedra plant spans across Asia, including the Kingdom of Saudi Arabia. It is common in desert regions from Morocco to Algeria, Libya, Egypt, and the Arabian Peninsula [4] .

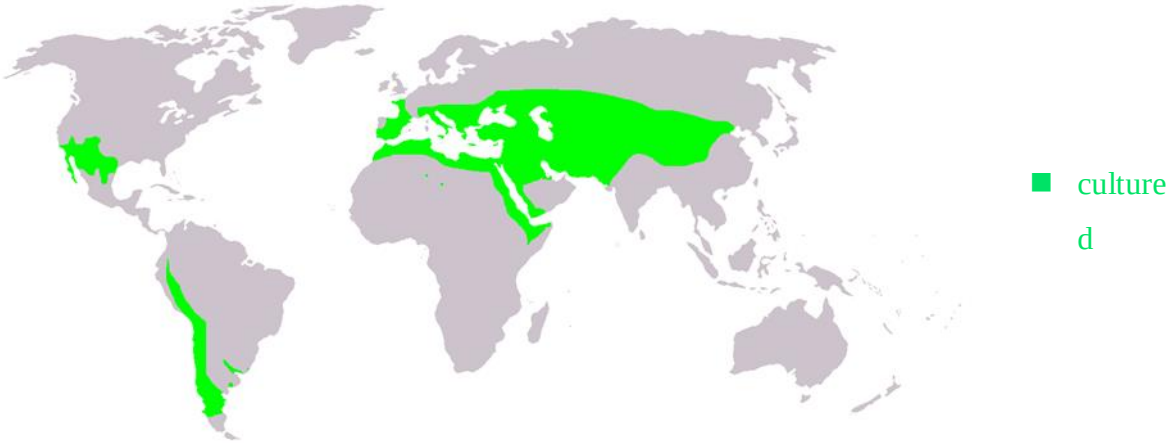


Figure 06: Geographical Distribution Map of the Ephedra Plant

2.6. The Economic Significance of the Ephedra Plant:

The Indian subcontinent leads the list of major countries in cashew production globally, possessing vast expanses of land suitable for cultivating this plant. Following closely is Brazil, which benefits from a favourable climate and natural resources conducive to large-scale cashew cultivation. Indonesia ranks third in cashew production, with regions such as Sumatra and Java renowned for their production of this type of nut [12].

2.7. The Chemical Composition of the Ephedra Plant:

Ephedra, also known as Ma Huang, possesses a complex chemical composition comprising a diverse array of active compounds. Among these compounds, ephedrine constitutes a significant and vital proportion of the alkaloids present in the plant. Several regions where this plant grows have been designated "Ephedra" due to its varying content of ephedrine, typically ranging from 0.5% to 2.5% of the plant's total mass.

Studies have indicated that the proportion of ephedrine in Ephedra varies among regions, influenced by climatic conditions and harvesting time. Ephedrine content has been identified to represent between **30%** to **90%** of the total alkaloids found in the plant [13].

In addition to ephedrine, Ephedra contains a diverse range of other active compounds such as flavonoids, phenols, amino acids, carboxylic acids, and terpenes. Flavonoids are considered one of the most important components of Ephedra due to their antioxidant and anticancer properties.

3. Chemical Study of Plants Used in Novel Approaches:

The field of medicinal plant studies has witnessed tremendous growth due to their potential in treating various diseases. These plants are rich sources of bioactive compounds with significant therapeutic properties, making them promising candidates for the development of new drugs. Some studies have shown that certain plant-derived drugs exhibit therapeutic efficacy surpassing that of manufactured medications in treating certain diseases. Consequently [14], considerable efforts have been made in research to study the properties of medicinal plants and to develop more effective and safe drugs from natural sources [15].

3.1. Definition of Active Natural Products:

Active natural products are organic compounds of natural origin produced by living organisms. These products derive their importance from their role in metabolic reactions and are characterized by their ability to positively or negatively affect human health [16]. These products are classified into two main types:

3.1.1. Primary Metabolism:

Primary metabolism refers to a set of fundamental chemical reactions that occur within living cells. These reactions are responsible for generating energy and building cellular components [17].

3.1.2. Secondary Metabolism:

Secondary metabolism refers to an additional set of chemical reactions that occur within living cells. These reactions produce compounds with specific properties such as colors and antioxidant substances [17].

4. Definition of Alkaloids

Alkaloids are complex natural compounds with unique activity properties. The term "alkaloid" was coined in 1818 by the researcher Meissner to describe a distinct class of natural compounds produced by plants [18].

Alkaloids are distinguished by their unique chemical properties, typically consisting of complex nitrogen-containing bases that impart alkaline characteristics. Their structures often include functional groups such as hydroxyl or ketone groups, along with the presence of non-homogeneous rings in many cases.

A single plant may contain over 100 different types of alkaloids, and these compounds are distributed throughout various plant families in concentrations that sometimes exceed 10% of the plant's dry weight.

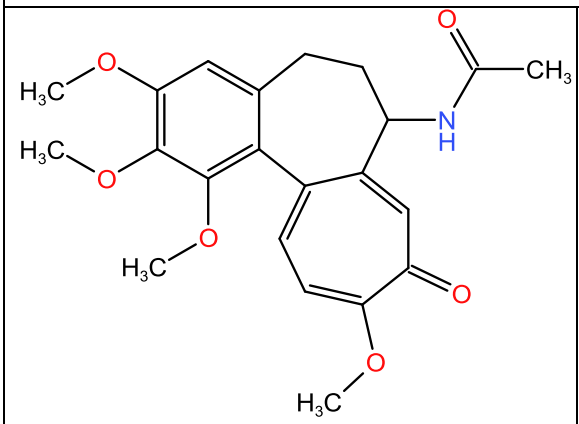
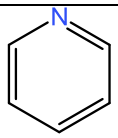
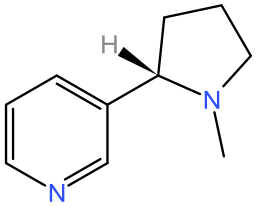
Alkaloids are typically associated with organic acids or tannins in plants, and their content varies between 1% and 3% of the dry weight. Their molecular mass ranges from 100 to 900 grams/mol, and they often exist in a crystalline solid form. Alkaloids exhibit basic properties, reacting with acids to form salts. Their solubility varies with pH and the basic state; they dissolve in non-polar organic solvents but are insoluble in water and polar organic solvents.

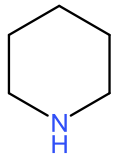
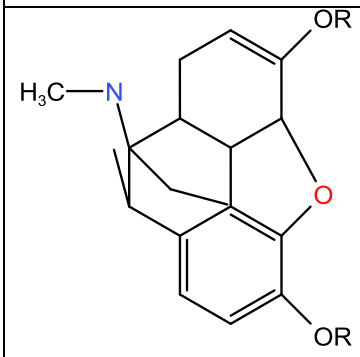
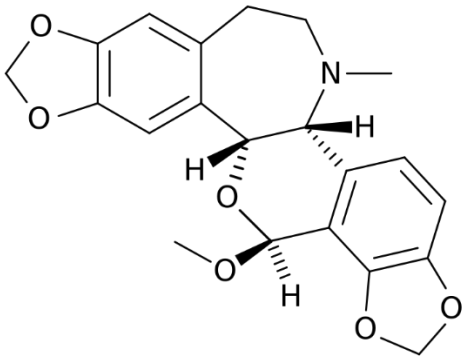
Alkaloids are a significant source of medicines and treatments and find various applications in fields such as cosmetics, food products, and agriculture. Considerable research efforts are dedicated to studying alkaloid properties and developing new applications for them [19].

4.1. Classification of Alkaloids:

Alkaloids are typically classified into various groups based on the type of basic ring present in the alkaloid portion [20]. Common alkaloids are listed in table 03.

Table 03: Some Categories of Alkaloids

Example	Type	Characteristic	Section
Artificial alkaloids			
	Amino **Alkaloids**	A ring group Two nitrogen atoms Compounds derived from phenyl ethyl amine	Homocyclic ring alkaloid group
	**pyridine**	Ring compounds containing one or more nitrogen atoms depending on the type of alkaloid Their alkaloids contain multiple ring groups	Heterocyclic ring alkaloid group
	**nicotine**		

	**Pipéridine**	Non-homogeneous fundamental	
	**Morphine**		
Natural alkaloids			
	**Rhœadine**	It consists of a xanthene ring connected to a phenyl group and an amine group. It contains one chlorine (Cl) atom	Homocyclic ring alkaloid group

4.2. The Role and Importance of Biological Alkaloids

Alkaloids are secondary metabolites that play a vital role in plant defense against herbivores and contribute to their biological system. Moreover, they are utilized in the field of pharmacy. Their significance lies in the following[21] .

4.2.1. Role of Alkaloids in Plants:

-Plant alkaloids play a crucial biological role in the plant's life system by producing compounds with distinct biological effects and physiological activities that contribute to growth regulation.

-They serve as a reservoir for nitrogen and other essential substances for plant growth in various stages of its life cycle.

-They constitute the results of protein breakdown or end products of nitrogen metabolism and are stored in non-harmful forms for the plant [21] .

4.2.2. The Importance of Alkaloids:

-They contribute to plant defense against herbivores and protect them from harmful effects.

-Anti-inflammatory agents: Colchicine.

-Analgesics: Morphine and codeine.

-Antitussives: Codeine.

-Alzheimer's treatment: Includes improving cognitive functions and slowing disease progression.

-Diuretic and antipyretic, such as the compound hordenine found in barley.

-It is characterized by its antibacterial, antiviral, and anti-allergic roles.

-Increasing hydroxyl groups enhances its anti-tumor activity.

-Increasing the number of methoxy groups enhances its anticancer activity [22] .

1- Khan, S. U., Hamza, B., Mir, R. H., Fatima, K., & Malik, F. (2024). Lavender Plant: Farming and Health Benefits. Current Molecular Medicine.

2- Lis-Balchin, M. T. (2012). Lavender. In Handbook of herbs and spices (pp. 329-347). Woodhead Publishing.

3- Coelho JP, Cristino AF, Matos PG, Rauter AP, Nobre BP, Mendes RL, Barroso JG, Mainar A, Urieta JS, Fareleira JM, Sovová H, Palavra AF. Extraction of volatile oil from aromatic plants with supercritical carbon dioxide: experiments and modeling. Molecules. 2012 Sep 5;17(9):10550-73. doi: 10.3390/molecules170910550. PMID: 22951395; PMCID: PMC6268743.

4- Application picturethis

5- Oltean H, Robbins C, van Tulder MW, Berman BM, Bombardier C, Gagnier JJ. Herbal medicine for low-back pain. Cochrane Database Syst Rev. 2014 Dec 23;2014(12):CD004504. doi: 10.1002/14651858.CD004504.pub4. PMID: 25536022; PMCID: PMC7197042.

6- Ruffoni B, Pistelli L, Bertoli A, Pistelli L. Plant cell cultures: bioreactors for industrial production. Adv Exp Med Biol. 2010;698:203-21. doi: 10.1007/978-1-4419-7347-4_15. PMID: 21520713.

7- [19] Samuelson R, Lobl M, Higgins S, Clarey D, Wysong A. The Effects of Lavender Essential Oil on Wound Healing: A Review of the Current Evidence. J Altern Complement Med. 2020 Aug;26(8):680-690. doi: 10.1089/acm.2019.0286. Epub 2020 Jun 24. PMID: 32589447.

8- González-Juárez DE, Escobedo-Moratilla A, Flores J, Hidalgo-Figueroa S, Martínez-Tagüeña N, Morales-Jiménez J, Muñiz-Ramírez A, Pastor-Palacios G, Pérez-Miranda S, Ramírez-Hernández A, Trujillo J, Bautista E. A Review of the Ephedra genus: Distribution, Ecology, Ethnobotany, Phytochemistry and Pharmacological Properties. Molecules. 2020 Jul 20;25(14):3283. doi: 10.3390/molecules25143283. PMID: 32698308; PMCID: PMC7397145.

9- Khalil AMA, Hassan SE, Alsharif SM, Eid AM, Ewais EE, Azab E, Gobouri AA, Elkelish A, Fouda A. Isolation and Characterization of Fungal Endophytes Isolated from Medicinal Plant Ephedra pachyclada as Plant Growth-Promoting. Biomolecules. 2021 Jan 22;11(2):140. doi: 10.3390/biom11020140. PMID: 33499067; PMCID: PMC7911138

10- Yao T, Wang Q, Han S, Lu Y, Xu Y, Wang Y. Potential Molecular Mechanisms of Ephedra Herb in the Treatment of Nephrotic Syndrome Based on Network

Pharmacology and Molecular Docking. Biomed Res Int. 2022 Jul 5;2022:9214589. doi: 10.1155/2022/9214589. PMID: 35837376; PMCID: PMC9276517.

11- Miao SM, Zhang Q, Bi XB, Cui JL, Wang ML. A review of the phytochemistry and pharmacological activities of Ephedra herb. Chin J Nat Med. 2020 May;18(5):321-344. doi: 10.1016/S1875-5364(20)30040-6. PMID: 32451091.

12- Guo L, Gao Y, He P, He Y, Meng F. Modeling for Predicting the Potential Geographical Distribution of Three Ephedra Herbs in China. Plants (Basel). 2023 Feb 9;12(4):787. doi: 10.3390/plants12040787. PMID: 36840134; PMCID: PMC9963152.

13- Ibragic S, Sofić E. Chemical composition of various Ephedra species. Bosn J Basic Med Sci. 2015 Jul 18;15(3):21-7. doi: 10.17305/bjbms.2015.539. PMID: 26295290; PMCID: PMC4594322.

14- BROWN D., RASHOTTE A., MURPHY A., NORMANLY B., TAGUE W., PEER L., TAIZ G., 2001. Flavonoids act as negative regulators of auxine pharmacie. Botanique – Pharmacognosie – Phytothérapie – Homéopathie. Ed. Pharmacol, 32.P :1995.

15- ZHOU J., WANG L., WANG J., TANG N., 2001. Antioxidative and antitumour activities of solid quercetin metal(II) complexes. Transition Met. Chem ,26(1-2), 57-63.

16- Zhao Y, Liu Z, Liu G, Zhang Y, Liu S, Gan D, Chang W, Peng X, Sung ES, Gilbert K, Zhu Y, Wang X, Zeng Z, Baldwin H, Ren G, Weaver J, Huron A, Mayberry T, Wang Q, Wang Y, Diaz-Rubio ME, Su X, Stack MS, Zhang S, Lu X, Sheldon RD, Li J, Zhang C, Wan J, Lu X. Neutrophils resist ferroptosis and promote breast cancer metastasis through aconitate decarboxylase 1. Cell Metab. 2023 Oct 3;35(10):1688-1703.e10. doi: 10.1016/j.cmet.2023.09.004. PMID: 37793345; PMCID: PMC10558089.

17- Ibragic S, Sofić E. Chemical composition of various Ephedra species. Bosn J Basic Med Sci. 2015 Jul 18;15(3):21-7. doi: 10.17305/bjbms.2015.539. PMID: 26295290; PMCID: PMC4594322..

18- Bhambhani S, Kondhare KR, Giri AP. Diversity in Chemical Structures and Biological Properties of Plant Alkaloids. Molecules. 2021 Jun 3;26(11):3374. doi: 10.3390/molecules26113374. PMID: 34204857; PMCID: PMC8199754.

19- Turnbull D, Rodricks JV, Mariano GF, Chowdhury F. Caffeine and cardiovascular health. Regul Toxicol Pharmacol. 2017 Oct;89:165-185. doi: 10.1016/j.yrtph.2017.07.025. Epub 2017 Jul 26. PMID: 28756014.

20- [32] Wei WJ, Chen XH, Guo T, Liu XQ, Zhao Y, Wang LL, Lan JX, Li HW, Si YP, Wang ZM. A Review on Classification and Biological Activities of Alkaloids from the

Genus Zanthoxylum Species. Mini Rev Med Chem. 2021;21(3):336-361. doi: 10.2174/1389557520666200910091905. PMID: 32912124.

21- [33] Cui X, Wang S, Cao H, Guo H, Li Y, Xu F, Zheng M, Xi X, Han C. A Review: The Bioactivities and Pharmacological Applications of Polygonatum sibiricum polysaccharides. Molecules. 2018 May 14;23(5):1170. doi: 10.3390/molecules23051170. PMID: 29757991; PMCID: PMC6099637.

22- [34] Dai P, Zhu L, Yang X, Zhao M, Shi J, Wang Y, Lu L, Liu Z. Multidrug resistance-associated protein 2 is involved in the efflux of Aconitum alkaloids determined by MRP2-MDCKII cells. Life Sci. 2015 Apr 15;127:66-72. doi: 10.1016/j.lfs.2015.02.011. Epub 2015 Mar 2. PMID: 25744397.

Chapter II

Extraction Methods & Biological Activities

1. Gupta R, Gupta N, Weber, DDS KK. Dental Implants. 2023 Aug 8. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2024 Jan–. PMID: 29262027.
2. [2] Armanian AM, Barekatain B, Hoseinzadeh M, Salehimehr N. Prebiotics for the management of hyperbilirubinemia in preterm neonates. J Matern Fetal Neonatal Med. 2016 Sep;29(18):3009-13. doi: 10.3109/14767058.2015.1113520. Epub 2015 Nov 30. PMID: 26513278.
3. Ling JKU, Hadinoto K. Deep Eutectic Solvent as Green Solvent in Extraction of Biological Macromolecules: A Review. Int J Mol Sci. 2022 Mar 21;23(6):3381. doi: 10.3390/ijms23063381. PMID: 35328803; PMCID: PMC8949459.
4. Fagbemi KO, Aina DA, Olajuyigbe OO. Soxhlet Extraction versus Hydrodistillation Using the Clevenger Apparatus: A Comparative Study on the Extraction of a Volatile Compound from Tamarindus indica Seeds. ScientificWorldJournal. 2021 Dec 2;2021:5961586. doi: 10.1155/2021/5961586. PMID: 34899085; PMCID: PMC8660188.
5. Morsy, N. (2014). Phytochemical analysis of biologically active constituents of medicinal plants. Main Group Chemistry, 13(1), 7-21.
6. Evans, W. C. (2009). Trease and Evans' pharmacognosy. Elsevier Health Sciences.
7. Harborne, A. J. (1998). Phytochemical methods a guide to modern techniques of plant analysis. springer science & business media.
8. Balouiri, M., Sadiki, M., & Ibnsouda, S. K. (2016). Methods for in vitro evaluating antimicrobial activity: A review. Journal of pharmaceutical analysis, 6(2), 71-79.
9. Sabu, M. C., & Kuttan, R. (2002). Anti-diabetic activity of medicinal plants and its relationship with their antioxidant property. Journal of ethnopharmacology, 81(2), 155-160.
10. Sirivibulkovit, K., Nouanthavong, S., & Sameenoi, Y. (2018). based DPPH assay for antioxidant activity analysis. Analytical sciences, 34(7), 795-800.

I. Extraction:

1.1. Definition of extraction:

Extraction is the process of separating a specific substance from a mixture using a suitable solvent, based on the differing solubility of substances. Common types include solvent extraction, liquid-liquid extraction, and solid-liquid extraction. Applications range from the pharmaceutical industry, where active ingredients are extracted from plants, to analytical chemistry for isolating mixture components, and environmental remediation for removing contaminants. This process is essential for obtaining pure, high-quality materials, improving the effectiveness of chemical analyses and procedures [1].

1.2. Extraction Methods:

1.2.1. Water Distillation:

The principle of this method involves mixing the reacting materials, namely water and the plant from which the oil is to be extracted. It is then heated to the boiling point, causing steam to carry essential oil particles. These particles are then condensed using a special condenser and separated due to their difference in density to obtain the oil [2].

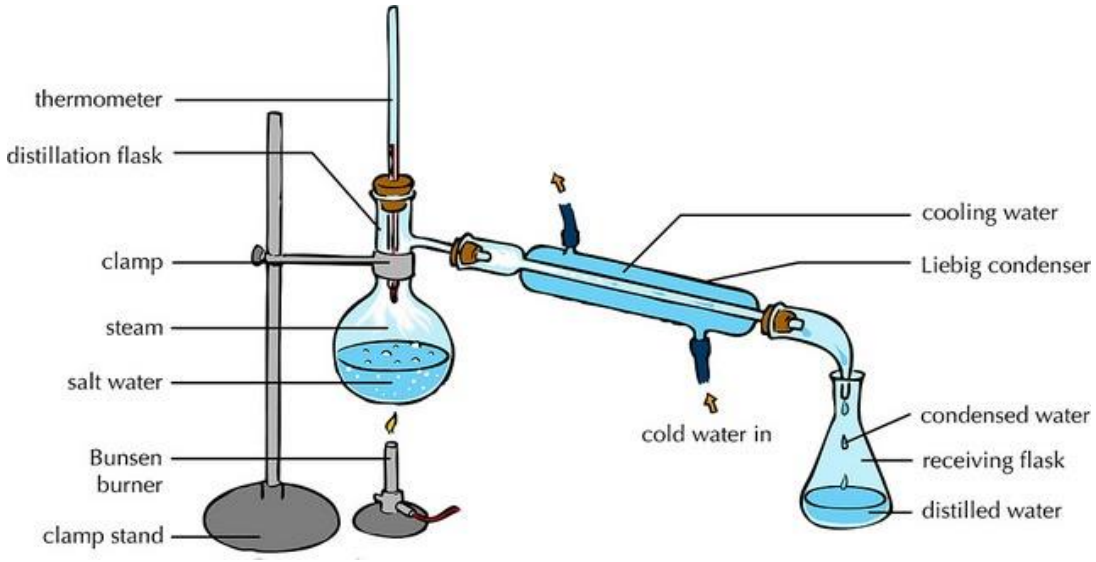


Figure 01: Water Distillation apparatus

1.2.2. Water Distillation Benefits:

High Efficiency: Extracts essential oils and aromatics efficiently, ensuring purity

Preservation: Maintains chemical composition without decomposition

Ease: Simple implementation, minimal equipment needed, suitable for small-scale projects

Control: Allows precise temperature and pressure control for accurate extraction

Eco-friendly: Relies on water and steam, avoids harmful chemical solvents

1.2.3. Water Distillation Disadvantages:

Time, energy-intensive process

Risk of losing heat-sensitive compounds

Limited efficiency in extracting all aromatic components

High water consumption

Potential contamination of oils with water

Requires specialized equipment

1.3. Solvent Extraction:

This method is used for plants containing very small quantities of oils and those sensitive to temperature. The plant material, intended for oil extraction, is dissolved in an organic solvent, forming a mixture of oil and solvent. The separation between them is then achieved through distillation under low pressure [3].

1.3.1. The Soxhlet apparatus:

extracts organic compounds efficiently from solid samples using continuous solvent cycling. It consists of a flask, condenser, and extractor. Solvent vaporizes rises, condenses, and drips back through the sample, repeating until extraction completes. Despite long extraction times, it's valuable for its automation and ability to process multiple samples.

Benefits:

1- Efficient extraction through continuous solvent cycling.
2- Automated operation minimizes supervision requirements.
3- Versatile, enabling batch processing of multiple samples simultaneously.

Disadvantages:

1- Long extraction times.
2- Risk of solvent loss.
3- Limited solvent options .

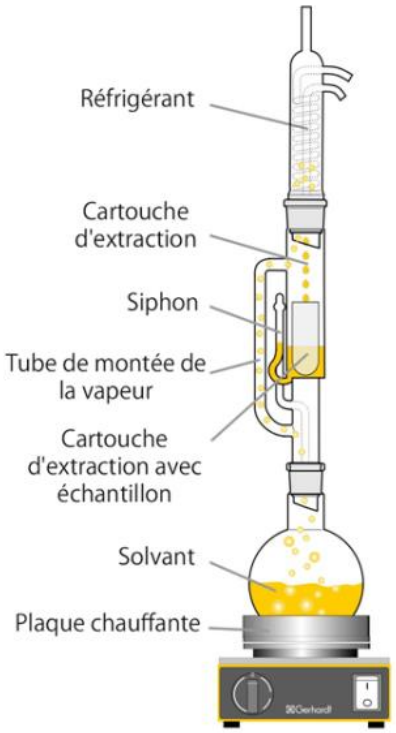


Figure 02: Soxhlet Apparatus

1.4.Maceration Extraction:

The principle of soaking extraction involves immersing the plants in large containers filled with solvent for a sufficient period to dissolve the essential oil present in them[4] .

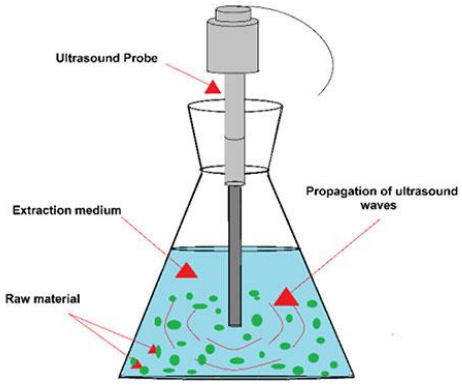


Figure 03: Maceration extraction

1.4.1. Steps:

1- Soaking: The plants are washed and soaked in water for a period ranging from 24 to 48 hours, with occasional stirring to ensure even distribution of water

2- Filtration: After the soaking period, the plants are separated from the water using a strainer or sieve.

3- Packaging: The filtered plants are packed into suitable bottles or containers.

4- Storage: The bottles are stored in a cool, dry place away from direct sunlight.

1.4.2. Factors Influencing:

1-Water Temperature: Control of the water temperature during the soaking process is crucial to prevent plant damage or any adverse effects on the quality of the extracted oil.

2-Soaking Duration: The duration of soaking directly affects the quantity of the extracted oil.

3-Plant Type: The quantity and quality of the oil vary depending on the type of plants used

1.4.3. Benefits of Soaking Extraction:

1- Ease of Application: Soaking extraction is one of the simplest methods for extracting essential oils.

2-Cost-Effectiveness: This method does not require complex or expensive equipment.

3-High Quality: High-quality oil can be obtained through this method when applied correctly.

1.4.4. Disadvantages of Soaking Extraction:

1- Time Consuming: This method takes longer compared to some other methods such as distillation.

2- Loss of Some Oil: Some quantity of oil may be lost during the soaking process.

3- Susceptibility to Damage: Plants may be susceptible to damage if storage conditions are not properly controlled.

Adding some salt to the solvent during the soaking process can help preserve the plant material and prevent bacterial growth. It is recommended to change the solvent every 24 hours, or every 12 hours if distilled water is used, during the soaking process to achieve the best results [4].

II. Biological activities

1. Rapid Tests Screening

1.1 Mayer's Test

Mayer's test is a qualitative assay used to detect alkaloids in plant extracts. Alkaloids are a group of naturally occurring organic compounds that mostly contain basic nitrogen atoms. They have significant pharmacological effects on humans and animals. The procedure for Mayer's test involves adding Mayer's reagent, which is a solution of mercuric chloride in potassium iodide, to a sample of the plant extract. The presence of alkaloids is indicated by the formation of a cream-colored or white precipitate, which results from the reaction of the alkaloids with the reagent. This test is highly sensitive and is commonly used in phytochemical screening to confirm the presence of alkaloid compounds. It is an essential initial step in the identification and characterization of medicinal plants [5].

Example: Proposed Reaction for Mayer Test with Rhoeadine

1. Reaction between $HgCl_2$ and KI:

$HgCl_2 + 2\ KI \rightarrow HgI_2 + 2\ KCl$

2. Formation of the Tetraiodomercurate (II) Complex:

$HgI_2 + 2\ KI \rightarrow K_2[HgI_4]$

3. Reaction with Rhoeadine (R-N):

$R\text{-}N + K_2[HgI_4] \rightarrow [R\text{-}NH^+][HgI_4^{2-}] + K[HgI_4]$

Mechanism Explanation

The tetraiodomercurate(II) complex ($K_2[HgI_4]$) reacts with the alkaloid rhoeadine. The lone pair of electrons on the nitrogen atom of rhoeadine coordinates with the Hg^{2+} ion, forming a complex. This results in the formation of a cream-colored precipitate, indicating the presence of the alkaloid.

1.2 Wagner's Test

Wagner's test is another qualitative assay used to detect the presence of alkaloids in plant extracts. This test employs Wagner's reagent, which consists of iodine in potassium iodide. When this reagent is added to the plant extract, the formation of a reddish-brown precipitate indicates a positive result for alkaloids. This test is widely used due to its simplicity and reliability in detecting alkaloids. Alkaloids have diverse and significant biological activities, including analgesic, antimalarial, and anticancer properties. Therefore, the detection of

alkaloids is crucial in pharmacognosy and natural product research. Wagner's test, along with Mayer's test, provides a preliminary confirmation of alkaloid presence before further detailed studies are conducted [6].

Example: Proposed Reaction for Wagner Test with Rhoeadine

1. Reaction between Iodine and Potassium Iodide:

$I_2 + KI \rightarrow KI_3$

2. Formation of the Iodine-Potassium Iodide Complex:

$KI + I_2 \rightarrow KI_3$

3. Reaction with Rhoeadine (R-N):

$R\text{-}N + KI_3 \rightarrow [R\text{-}NH^+][I_3^-]$

Mechanism Explanation

The iodine-potassium iodide complex (KI_3) reacts with the alkaloid rhoeadine. The lone pair of electrons on the nitrogen atom of rhoeadine coordinates with the I_2, forming a complex. This results in the formation of a precipitate, indicating the presence of the alkaloid.

1.3 Dragendorff's Test

Dragendorff's test is another qualitative method for detecting alkaloids. It uses Dragendorff's reagent, a solution composed of bismuth subnitrate and potassium iodide. The procedure involves adding this reagent to the plant extract, where the presence of alkaloids is indicated by the formation of an orange or orange-red precipitate. Dragendorff's test is frequently used alongside Mayer's and Wagner's tests to provide a comprehensive screening for alkaloid presence. Alkaloids identified by these tests can be further isolated and studied for their pharmacological properties. The test is an important tool in the field of pharmacognosy, aiding in the discovery of new therapeutic agents from natural sources [7].

Example: Proposed Reaction for Dragendorff Test with Rhoeadine

1. Formation of Bismuth Subnitrate:

$Bi(NO_3)_3 + H_2O \rightarrow BiONO_3 + 2\ HNO_3$

2. Reaction with Potassium Iodide:

$BiONO_3 + 4\ KI + H_2O \rightarrow KBiI_4 + KNO_3 + 2\ HNO_3$

3. Reaction with Rhoeadine (R-N):

$R\text{-}N + KBiI_4 \rightarrow [R\text{-}NH^+][BiI_4^-] + KNO_3$

Mechanism Explanation

The bismuth subnitrate reacts with potassium iodide to form the bismuth iodide complex ($KBiI_4$). This complex reacts with the alkaloid rhoeadine, where the lone pair of electrons on the nitrogen atom of rhoeadine coordinates with the Bi^{3+} ion. This results in the formation of an orange precipitate, indicating the presence of the alkaloid.

2. Biological Activities

2.1 Anti-Microbial Activity: Agar Diffusion Method

The agar diffusion method, specifically the well diffusion technique, is a fundamental procedure used to evaluate the antimicrobial properties of substances. This method involves the use of Petri dishes filled with agar medium, which is inoculated with a standardized microbial suspension. Wells are then created in the agar and filled with the test substance. The plates are incubated, and the antimicrobial activity is determined by measuring the clear zones around the wells where microbial growth has been inhibited. This technique is simple, cost-effective, and widely used in microbiology to screen for potential antimicrobial agents [8].

2.2 Anti-Diabetic Activity

Anti-diabetic activity assays involve evaluating the ability of substances to manage blood glucose levels, often through in vitro or in vivo models. These assays typically assess the inhibition of carbohydrate-hydrolyzing enzymes, such as alpha-amylase and alpha-glucosidase, or the enhancement of insulin secretion and sensitivity. In vitro studies might involve using enzyme assays, while in vivo studies often use animal models to observe the effect on blood

glucose levels over time. Such studies are essential in identifying potential treatments for diabetes, a prevalent metabolic disorder characterized by chronic hyperglycemia [9].

2.3 Antioxidant Activity: DPPH Assay

The DPPH (2,2-diphenyl-1-picrylhydrazyl) assay is a widely used method to evaluate the antioxidant activity of substances. It measures the ability of antioxidants to scavenge free radicals, which is indicated by a color change from purple to yellow as the DPPH radical is reduced. The degree of discoloration is quantitatively measured using a spectrophotometer, and it reflects the scavenging ability of the test compound. The results are often expressed as IC50, the concentration required to inhibit 50% of the DPPH radicals. This assay is essential in the screening of natural products and synthetic compounds for antioxidant properties, which are crucial in preventing oxidative stress-related diseases [10].

Experimental Section

Chapter III

Materials & Methods

1. Materials and chemicals:

In the study of plant-based extracts and their applications, the selection and preparation of materials, glassware, and chemicals play a critical role in ensuring accurate and reproducible results. This chapter outlines the essential components and methodologies utilized in our laboratory work, focusing on the extraction and analysis of compounds from lavender and ephedra plants. By systematically listing the materials, glassware, and chemicals in structured tables, we provide a clear overview of the tools and substances employed in our experiments. These foundational elements are crucial for maintaining consistency and precision throughout the experimental procedures, ultimately contributing to the reliability of our findings.

Table 01: Equipment used in laboratory work

Apparatus or device	Reference	Working principle
Magnetic agitator	MS7-H550-PRO	The magnetic stirrer uses magnetic interactions to effectively and precisely stir liquids, and is utilized in chemical laboratories, the food and pharmaceutical industries, and water treatment.
Magnetic bar	/	The magnetic stirrer generates a magnetic field to rotate a magnetic stir bar within a vessel, achieving efficient and precise liquid mixing.
Sensitive balance	EX125D	It measures the weight of samples with complete accuracy
Soxhlet extractor	/	The Soxhlet extraction method continuously extracts compounds from a solid with a heated solvent, which vaporizes, condenses, and repeats, efficiently used in food science, pharmaceuticals, and chemical analysis.
Clevenger apparatus	/	The Clevenger apparatus extracts essential oils from plant materials via steam distillation, where steam carries the oil vapors to a condenser. The oil then separates from water and collects in a graduated tube, efficiently used in fragrance, cosmetics, and herbal medicine.
Balloon heater	Wise therm	It works to heat and reach the required temperature
Water pump	Nubios MY-10L	A water pump in chemical experiments continuously circulates water through a heat exchanger to absorb and

		remove heat, cooling equipment or processes before recirculating the cooled water.
Rotary evaporator	Heidolph	A rotary evaporator removes solvents by rotating a sample in a heated water bath under vacuum, evaporating and condensing the solvent, and leaving a concentrated sample.
Ultrasonic apparatus	Iso LAB	The ultrasonic device (Ultrason) in chemical labs generates high-frequency vibrations in liquids, inducing cavitation, which enhances reactions, extracts compounds, breaks down molecules, prepares emulsions, and cleans equipment, thus accelerating experimental processes.
The ultraviolet (UV) device	Prove 300	The UV device in chemical labs generates ultraviolet light at specific wavelengths using lamps like mercury or deuterium, controlled by filters or monochromators. It interacts with substances to excite electrons, break bonds, induce fluorescence, enabling spectrophotometric analysis for substance concentrations, purity verification, fluorescence analysis, sterilization, and studying biological structures, enhancing chemical analysis precision and quality.
Laboratory Centrifuge	Cenlee 300ml	A centrifuge spins samples at high speeds, causing heavier particles to settle at the bottom while lighter components remain suspended, allowing for efficient separation based on density or size.

Table 02: Glassware used in the practical part

Glassware	*Capacity (mL)*
Glass Separatory Funnel	250
Becher	100, 300, 500, 1000
Titration burette	1, 5, 10
Filter paper	/
Flask	250
G	25, 50, 100
spatula	/
Flask	250, 1000
Erlenmeyer	500

Table 03: Chemicals and plant materials

Designation	Quantity
Lavender plant	300 g
Ephedra plant	300 g
Ethanol (96%)	1.5 L
Methanol (96%)	2 L
Distilled water	/
H_2SO_4 (%2)	10 mL
NH_4^+(33%)	250 mL(Diluted)
$(C_2H_5)_2O$	300 ml
Yeast	125 g
Glucose Solution	/
$C_4H_8O_2$	300 ml
Alkaloid indicators (Mayer, Wagner, Dragendorff)	drops

2. Sampling:

The plants were purchased from a very well-known herbal seller or herbalist in the city market, whose name is Obaidi spice dealer. The information about the place where the samples were taken was transmitted by the financier from whom the spice dealer buys the plants.

2.1. Lavender plant:

2.1.1. Plant location:

Lavender, *Lavandula officinalis* L., traditionally known for its growth in India, has recently been successfully cultivated in Oran. Lavender harvested specifically from the rural area of **Arzew** in Oran city. This plant thrives in sunny locations with well-drained soil, making Arzew an ideal region for its cultivation.

2.1.2. Geographical location :

Aïn El Kerma or Aïn Kerma, is an Algerian commune in the city of Oran, located 45 km west of the city of Oran. Its geographic coordinates are as follows: 35° 38′ 56″ north, 0° 58′ 33″ west.

Figure 01: Location of Ain El Kerma according to google maps

2.2. Ephedra plant:

2.2.1. Plant location:

Ephedra, *Ephedra alata*, also has recently been successfully cultivated in El Oued. Ephedra harvested specifically from Oued El Alanda in El Oued city. This plant thrives in sunny locations with well-drained soil, making Oued El Alanda an ideal region for its cultivation.

2.2.2. Geographical location :

Oued El Alenda is a commune in the city of El Oued in Algeria. The territory of the commune of Oued El Alenda is located in the southwest of El Oued city. Its geographic coordinates are as follows: 33° 13′ 44″ north, 6° 45′ 26″ east.

Figure 02: Location of Oued El Alanda according to google maps

3. Preparation of materials:

3.1. Ephedra plant

The Ephedra plant sample was bought in winter, on February 22, 2024. The optimal time for sampling was determined, varying depending on the substance to be extracted. A fresh sample was selected to ensure accurate results. The plant sample was rinsed three times with lukewarm water and then washed three times with distilled water to remove any adhering substances and dirt.

Figure 03: Sticks of the Ephedra alata plant

After rinsing, the sample was dried at room temperature for 7 days, from February 23 to February 29, 2024. Half of the sample was then ground smoothly, while the other half was left in the form of green twigs.

Figure 04: Alanda plant powder

3.2. Lavender plant:

The lavender plant sample was collected in early spring on March 26, 2024. The optimal time for sampling was determined, varying depending on the substance to be extracted. A fresh sample was selected to ensure accurate results. The plant sample was rinsed three times with tap water and then washed three times with distilled water to remove any adhering substances and dirt.

Figure 05: Lavender flowers

After rinsing, the sample was dried at room temperature for 8 days, from March 26 to April 4, 2024. Half of the sample was then ground smoothly, while the other half was left in the form of small purple roses.

Figure 06: Lavender powder

4. Extraction:

4.1. Maceration:

4.1.1. Ethanoic medium:

Preparation of Equipment:

Take two 500 ml beakers and two magnetic bars. Sterilize them with distilled water and a chemical solution drying oven

Procedure:

1. Weigh 50 grams of each of the studied dried plants (Ephedra, Lavender).

2. Measure 400 ml of ethanol (96%) in each beaker.

3. Add the plant powder to the beaker and stir slightly until it is well mixed and settles a bit.

4. Place a magnetic bar in each beaker and then place them on a magnetic stirrer.

5. Cover the mixture with aluminum foil to avoid contamination with impurities during the stirring period (24 hours).

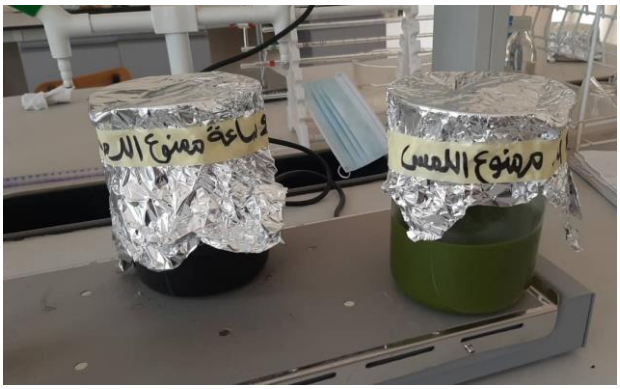


Figure 07: Maceration extraction

After 24 hours:

1. Filter the mixture using three layers of cheesecloth to remove large debris and label the filtrate as (M).

2. Perform the final filtration through three layers of filter paper to obtain a highly pure filtrate.

Figure 08: Step of filtration

3. Place the filtrate in a balloon of a rotary evaporator and evaporate a large amount of ethanol at 76°C to reduce drying time

Figure 09: Drying step by rotavapor

4 .Place the remaining filtrate in drying containers and put them in a chemical solution drying oven at 45°C (to preserve the components and avoid breaking down the fine molecules, especially alkaloids) until completely dry

Figure 10: Drying step in drying oven

5. After drying, place the resulting powder in small glass vials

6. Take (M) and repeat the previous steps of stirring, filtration, and drying to increase the yield

4.1.2. Methanol medium:

Two 250 ml beakers and two magnetic bars are cleaned using distilled water and a chemical solution drying oven. After that, weigh 20 grams of each of the studied dried plants (Ephedra, Lavender). Then, measure 100 ml of methanol (96%) into each beaker. Additionally, place a magnetic bar in each beaker and then place them on a magnetic stirrer. At last, cover the mixture with aluminum foil to prevent contamination during the stirring process (24 hours).

. After 24 hours, we filter the mixture using three layers of gauze to remove large debris, labeling the filtrate as (S)

. We perform a final filtration using three layers of filter paper to obtain a filtrate with a high degree of purity

. We take (S) and repeat the previous steps of stirring, filtering, and drying three times to increase the yield

. We place the filtrate in an Erlenmeyer flask, suspend it on a holder, and subject it to an ultrasonic bath for 20 minutes, repeating this process eight times. This step aids in breaking down the particles to obtain a higher yield of alkaloids.

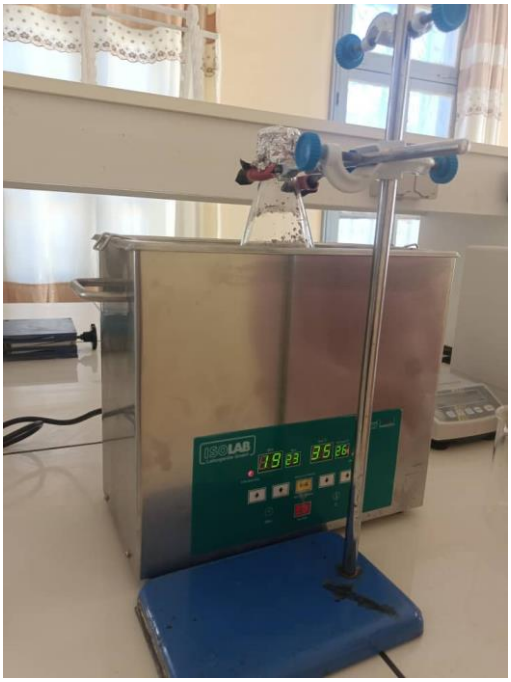


Figure 11: The extract sonification step

. The filtrate was placed in the flask of a rotary evaporator and evaporate a large quantity of ethanol at 40°C to shorten the drying time

. The remaining filtrate is then placed in drying dishes and transferred to a chemical solution drying oven set at 45°C (to preserve its components and prevent the breakdown of its fine particles, especially alkaloids) until completely dry. Once dried, the resulting powder is stored in small glass vials.

- After obtaining the methanolic extract from a three-day maceration, the raw extracts from the onions were acidified using an aqueous solution of sulfuric acid (2% v/v) to achieve a pH of two. The extracts were then cleaned using diethyl ether to remove neutral substances. The aqueous solution was then basified using 25% ammonium hydroxide (v/v) to reach a pH between 9 and 10. Finally, the alkaline extract was obtained from the basified aqueous solution using ethyl acetate, resulting in the alkaloid extract (AE) for each sample.

4.2. Soxhlet :

Starting with the preparation of the apparatus using a Soxhlet extractor, we placed 150 ml of methanol in the bottom flask and placed 20 grams of each raw material in filter bags made of paper. These bags were then securely wrapped and placed in the middle basket of the Soxhlet apparatus.

We operated the apparatus and connected it to a heat source, with continuous cooling water flowing through the condenser coil. During the operation, each material underwent a specific number of cycles, with each cycle lasting for a predetermined time. For lavender, there were 15 cycles, each lasting 40 minutes, and finally, for safflower, there were 5 cycles, each lasting 60 minutes.

After each cycle, we collected the resulting liquids from the extraction process in the bottom flask, in order to separate methanol from the aromatic compounds. Upon completing all cycles, we collected the liquid obtained from the extraction and determined the percentage of aromatic compounds extracted in methanol.

Figure 12: Soxhlet extractor

4.3. Clevenger:

The process of extracting essential oils using a Clevenger apparatus begins with preparing 50 grams of each type of plant (Ephedra, Lavender) separately and adding 600 ml of distilled water to each sample. The Clevenger apparatus, which consists of a heating flask, a distillation tube, and a condenser, is then set up. The plant sample and

distilled water are placed in the heating flask, which is heated until the water boils. The steam carrying the essential oils rises along with the water vapor through the distillation tube to the condenser. The steam condenses in the condenser and returns to a liquid state, flowing into the separation unit of the Clevenger apparatus where the essential oils accumulate on top of the water due to their immiscibility and difference in density. After the distillation process is complete, the essential oils are collected from the separation unit and separated from the remaining distilled water using a pipette or liquid-liquid separator. The essential oil is then purified if necessary to remove any impurities and stored in dark bottles to preserve its properties .

Figure 13: Clevenger apparatus

5. Biological activities:

5.1. Antimicrobial activity:

The agar diffusion method, specifically the well method, is a fundamental technique used to evaluate the antimicrobial activity of a substance. To begin, Petri dishes are prepared with Sabouraud dextrose agar supplemented with 2% glucose for yeasts, and Mueller-Hinton agar for bacteria. These dishes are then aseptically inoculated with a suspension containing 1.5×108 CFU/mL, standardized using McFarland Standard No. 0.5. The suspension is prepared using a VITEK® DENSICHEK® device and a mixture of sterile physiological saline solution (0.9% NaCl) and sterile distilled water

Once the agar surfaces have dried, wells are created at the center of each dish using the upper part of a Pasteur pipette. These wells are subsequently filled with 50 µL of an aqueous solution of the test compounds (RWAL, RWKH, and RWKZ) at concentrations of 120, 60, 30, and 15 mg/mL, prepared with 5% DMSO to ensure proper dissolution of the compounds.

The inoculated plates are incubated at 37°C for 48 hours for yeasts and 24 hours for bacteria. Antimicrobial activity is indicated by forming a clear zone of inhibition around the wells, where microbial growth has been prevented. The diameter of these inhibition zones is measured, and a product is considered to exhibit antimicrobial activity if the zone of inhibition is greater than 6 mm.

Figure 14: Anti microbial test

5.2. Antidiabetic activity:

Protocol of Glucose uptake in yeast cells:

Commercial baker's yeast (Saccharomyces cerevisiae) Saf-Instant® 125g was washed by repeated centrifugation at 3,000 × g for 5 minutes in distilled water until the supernatant fluids were clear. A 10% (v/v) suspension of the yeast was then prepared in distilled water. Various concentrations (80 mg/mL – 10 mg/mL) of the extracts RWKL, RWKH, and RWKZ were prepared. These extracts were added to 2 mL of a 25 mM glucose solution and incubated together for 10 minutes at 37 °C. The reaction was initiated by adding 200 µL of the yeast suspension, vortexing, and further incubation at 37 °C for 15, 30, 60, and 120 minutes. After the incubation periods, the tubes were centrifuged at 2,500 × g for 5 minutes, and the glucose concentration in the supernatant was estimated. The percent increase in glucose uptake by the yeast cells was calculated using the formula:

$$I\ (\%) = \frac{(A_C - A_S)}{A_C} \times 100$$

where I(%) is the increase of the uptake, Abs control is the absorbance of the control reaction (containing all reagents except the test sample) and Abs sample is the

absorbance of the test sample. Following the manufacturer's instructions, absorption was measured at 540 nm using a Mindray® BA-88 Biochemistry analyzer. All experiments were conducted in triplicates to ensure accuracy and reproducibility.

Figure 15: Antidiabetic test

5.3. Antioxidant activity (DPPH):

- To prepare DPPH solution, take 100 ml of ethanol and add 4 mg of DPPH to it. Mix the solution thoroughly until it is homogeneous, then store it in the dark (resulting in a concentration of 0.04 g/L)
- To prepare the stock solution, place 50 mg of each plant extract in a glass test tube. Add 50 ml of ethanol to the test tube and mix thoroughly until the solution is homogeneous, resulting in a concentration of 1 g/L.
- Take 9 test tubes and add varying volumes of the stock solution to each. Then, add a specific volume of ethanol to each tube so that the total volume in all tubes is 1 ml (dilution process)
- Add 1 ml of the previously prepared DPPH solution to each test tube and place them in the dark for half an hour. Then, measure the absorbance at a wavelength of λ=515 nm to compare the efficiency of the studied extracts. Ascorbic acid is used as a reference compound, and the inhibition percentage is calculated according to Chaouche et al. (2013):

$$I\ (\%)\ = \frac{(A_C - A_S)}{A_C} \times 100$$

I % = Percent inhibition

As: Absorbance intensity of DPPH assay in the presence of the sample

Ac: Absorbance intensity of DPPH in the absence of the sample

Figure 16: DPPH test before (purple) and after (yellow) scavenging

Chapter IV

Results & Discussion

This chapter presents the findings of the study and provides an in-depth analysis of the results. The data obtained from various experiments and tests are systematically discussed to elucidate the implications and significance of the outcomes. The results are interpreted in the context of existing literature, highlighting the advancements and contributions of this research. Through detailed discussion, this chapter aims to connect the empirical evidence with the theoretical framework, addressing the research questions and hypotheses formulated at the outset. The integration of results with discussion facilitates a comprehensive understanding of the study's impact and future directions.

1. Ethanolic Maceration :

1.1 Yield Estimation: The yield of extrcation can be calculated according to (Guettaf et al. (2016) as the following equation describes:

$$\text{Yield Percentage (\%)} = \frac{Mass\ of\ Extract}{Mass\ of\ Dry\ Plant\ Material} \times 100$$

$$\textbf{Ephedra (\%)} = \frac{\mathbf{7.170}g}{\mathbf{50}g} \textbf{ = 14.34 \%}$$

$$\textbf{Lavender (\%)} = \frac{\mathbf{9.560}g}{\mathbf{50}g} \textbf{ = 19.12 \%}$$

1.2 Rapid tests:

1.2.1 Mayer's test :

- We observed the appearance of white precipitate in the lavender test tube after 24 hours.
- We observed the appearance of very slight white precipitate in the ephedra test tube after more than 48 hours.

1.2.2 Wagner's test :

- We noticed the presence of dark brown sediment in lavender test tubes.
- We observed the absence of sediment in the ephedra test tube, with the indicator maintaining its original color.

1.2.3 Dragendorff's test :

- We observed the appearance of dark red sediment in both the ephedra and lavender test tubes.

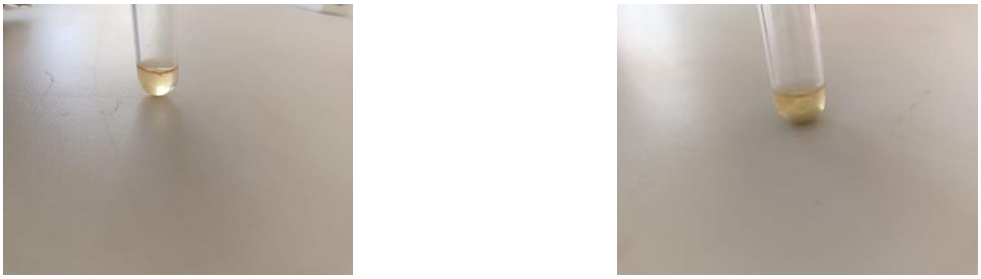
1.3 Explanation:

In this study, three standard qualitative tests (Mayer's, Wagner's, and Dragendorff's) were conducted to detect the presence of alkaloids in extracts from lavender and ephedra. The results of these tests are detailed below:

1.3.1 Mayer's Test:

Lavender: The appearance of a white precipitate after 24 hours indicates the presence of alkaloids in the lavender extract. Mayer's reagent reacts with alkaloids to form an insoluble precipitate, confirming their presence.

Ephedra: The appearance of a very slight white precipitate after more than 48 hours suggests a lower concentration of alkaloids in the ephedra extract. The delayed and faint precipitate formation implies that alkaloids are present but in smaller amounts compared to lavender.



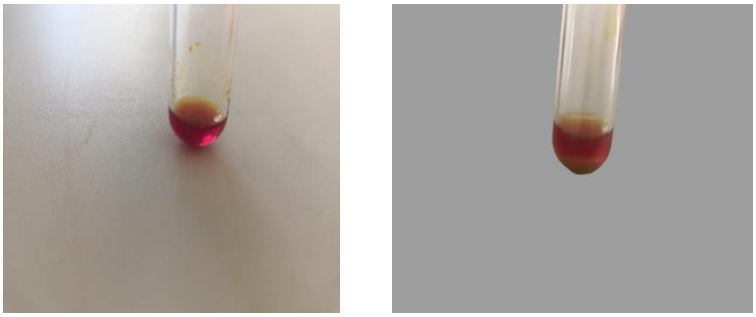
A **B**

Figure 01: Mayer's test observation

1.3.2 Wagner's Test:

Lavender: The presence of dark brown sediment indicates a positive reaction for alkaloids. Wagner's reagent, which contains iodine in potassium iodide, forms a brown precipitate with alkaloids, thereby confirming their presence in the lavender extract.

Ephedra: The absence of sediment and the maintenance of the indicator's original color suggest that alkaloids are either absent or present in very low concentrations in the ephedra extract. The lack of a visible reaction indicates a negative result for alkaloids.



A **B**

Figure 02: Wagner's test observation

1.3.3 Dragendorff's Test:

Lavender and Ephedra: The appearance of dark red sediment in both test tubes confirms the presence of alkaloids in both extracts. Dragendorff's reagent, which contains potassium bismuth iodide, reacts with alkaloids to form a red or orange precipitate. The positive results in both samples indicate that alkaloids are present in both lavender and ephedra extracts.

These observations collectively confirm the presence of alkaloids in both lavender and ephedra, with lavender showing a more robust response in the Mayer's and Wagner's tests, suggesting a higher alkaloid content compared to ephedra.

A **B**

Figure 03: Dragendorff's test observation

2. Methanolic maceration:

2.1 Yield Estimation: According(to Guettaf et al. (2016))

$$\textbf{Yield Percentage (\%)} = \frac{Mass\ of\ Extract}{Mass\ of\ Dry\ Plant\ Material} \times 100$$

Yield Percentage for Ephedra (%) =$\frac{1.5g}{20g}$ = 7.5 %

Yield Percentage for Lavender(%) =$\frac{0.52g}{20g}$ = 2.6 %

2.2 Alkaloid extraction :

The steps of total alkaloid extraction are the following:

	H_2SO_4	**$(C_2H_5)_2O$**	**$NH4^+$**	**$C_4H_8O_2$**
Lavender	Appearance of two phases a aqueous phase and an organic phase.	Dissolution of the organic phase and the appearance of two new aqueous phases.	Appearance of two phases accompanied by a color change.	Appearance of two distinct phases in thickness with a color change
Ephedra	Appearance of two phases an aqueous phase and an organic phase.	Dissolution of the organic phase and the appearance of two new aqueous phases.	Appearance of two phases accompanied by a color change.	Appearance of an extremely thin layer and another thick layer with a color change

2.3 Results :

2.3.1 Lavender plant:

The thickness of the alkaloid phase in the lavender plant differs due to the variation in alkaloid content between the two plants**.**

Figure 04: Separation by separatory funnel of Lavender extract

2.3.2 Ephedra plant :

Since the layer $C_4H_8O_2$ is very thin, it indicates a very low presence of alkaloids in the Ephedra plant.

Figure 05: Separation by separatory funnel of Ephedra extract

3. Biological activities:

To facilitate writing and to avoid repetition, we will symbolize the plant extract with the symbol RW + the abbreviation of the name of the plant. Therefore, RWAL will be the raw extract of the nightshade plant, and RWKHZ will be the abbreviation for the extract of the lavender plant.

3.1 Antioxidant activity:

3.1.1 Observation :

- We observe a change in the color of DPPH from purple to yellow**.**
- We notice an increase in the absorbance curve corresponding to concentration for the three plants.

3.1.2 Explanation :

- When the DPPH reagent interacts with antioxidant compounds in the plants, a color change from purple to yellow occurs. This transformation indicates that the antioxidant compounds have been utilized to neutralize (Inhibition) the free radicals present in the DPPH reagent.
- As for the increase in the curve, this signifies an enhancement in the effectiveness of the antioxidant compounds in the studied plants. This implies that the plants contain antioxidant compounds in varying quantities, depending on each plant.

3.1.3 Results :

- We conclude the presence of free radicals (carcinogenic substances) in the two extracts (Lavender and Ephedra). To determine which one has the highest and lowest free radicals, we follow the following results.

Concentration g/l / plants	0.01	0.025	0.050	0.075	0.1	0.125	0.150	0.175	0.2
I Lavender (%)	56	61	64	67	68	71	74	74	84
I Ephedra (%)	62	83	94	93	94	95	96	95	95

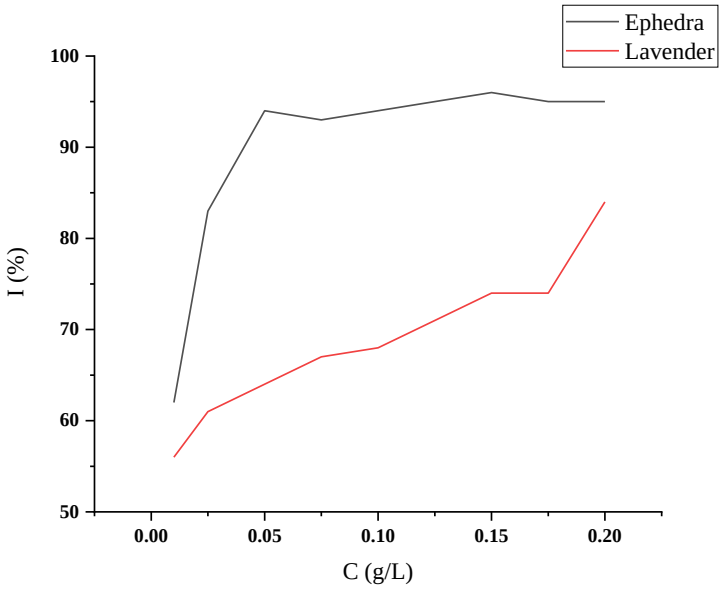


Figure 06: DPPH inhibition activity of Lavender and Ephedra plants

3.2 Antimicrobial activity:

3.2.1 Results :

Table 1. Results of antimicrobial tests: **RWAL**

	Microbial inhibition RWAL				
Strains used	80mg/ml	40mg/ml	20mg/ml	10mg/ml	CN
***Escherichia coli* ATCC 25922**	**10**	**8**	**8**	NI	33
***Pseudomonas aeruginosa* ATCC 27853**	**10**	NI	NI	NI	33
***Staphylococcus aureus* ATCC 25932**	**13**	**7**	**7**	NI	31
Bacillus subtilis ATCC 25973	NI	NI	NI	NI	28
	Anti-Candida activity				
Candida albicans ATCC 10231	NI	NI	NI	NI	/

NI = No Inhibition, CN = Gentamicin (CN) 30ug Discs.

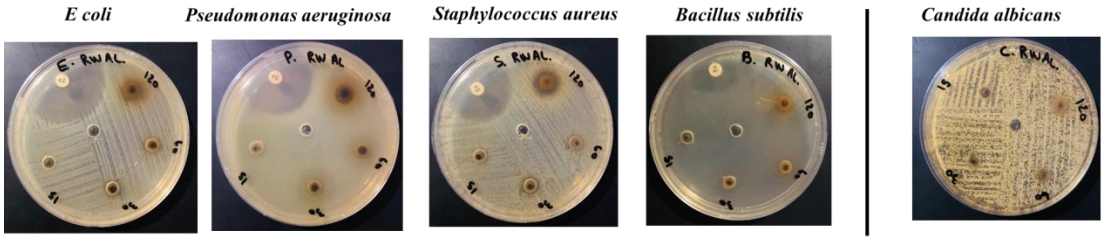


Figure 07: Antimicrobial test of RWAL

3.2.2 Interpretation

Escherichia coli ATCC 25922: Shows inhibition zones of 10 mm, 8 mm, and 8 mm at concentrations of 80 mg/ml, 40 mg/ml, and 20 mg/ml, respectively. No inhibition is observed at 10 mg/ml. The positive control (CN) shows a zone of 33 mm.

Pseudomonas aeruginosa ATCC 27853: Shows an inhibition zone of 10 mm at 80 mg/ml. No inhibition is observed at lower concentrations (40 mg/ml, 20 mg/ml, and 10 mg/ml). The positive control (CN) shows a zone of 33 mm.

Staphylococcus aureus ATCC 25932: Shows inhibition zones of 13 mm, 7 mm, and 7 mm at concentrations of 80 mg/ml, 40 mg/ml, and 20 mg/ml, respectively. No inhibition is observed at 10 mg/ml. The positive control (CN) shows a zone of 31 mm.

Bacillus subtilis ATCC 25973: No inhibition is observed at any concentration tested. The positive control (CN) shows a zone of 28 mm.

Anti-Candida Activity:

Candida albicans ATCC 10231: No inhibition is observed at any concentration tested.

The RWAL plant extract exhibits moderate antimicrobial activity against Escherichia coli and Staphylococcus aureus at higher concentrations (80 mg/ml), with some activity also observed against Pseudomonas aeruginosa. However, it shows no activity against Bacillus subtilis and Candida albicans.

Table 2. Results of antimicrobial tests: **RWKZ**

	Microbial inhibition RWKZ				
Strains used	80mg/ml	40mg/ml	20mg/ml	10mg/ml	CN
***Escherichia coli* ATCC 25922**	NI	NI	NI	NI	33
***Pseudomonas aeruginosa* ATCC 27853**	NI	NI	NI	NI	32
***Staphylococcus aureus* ATCC 25932**	7	NI	NI	NI	33
Bacillus subtilis ATCC 25973	NI	NI	NI	NI	26
	Anti-Candida activity				
Candida albicans ATCC 10231	NI	NI	NI	NI	/

NI = No Inhibition, CN = Gentamicin (CN) 30ug Discs.

Figure 08: Antimicrobial test of RWKHZ

- Escherichia coli ATCC 25922: No inhibition is observed at any concentration tested. The positive control (CN) shows a zone of 33 mm.
- Pseudomonas aeruginosa ATCC 27853: No inhibition is observed at any concentration tested. The positive control (CN) shows a zone of 32 mm.
- Staphylococcus aureus ATCC 25932: Shows an inhibition zone of 7 mm at 80 mg/ml. No inhibition is observed at lower concentrations (40 mg/ml, 20 mg/ml, and 10 mg/ml). The positive control (CN) shows a zone of 33 mm.

- Bacillus subtilis ATCC 25973: No inhibition is observed at any concentration tested. The positive control (CN) shows a zone of 26 mm.

Anti-Candida Activity:

- Candida albicans ATCC 10231: No inhibition is observed at any concentration tested.

The RWKZ plant extract exhibits very limited antimicrobial activity, with only a weak inhibition observed against Staphylococcus aureus at the highest concentration (80 mg/ml). It shows no activity against Escherichia coli, Pseudomonas aeruginosa, Bacillus subtilis, and Candida albicans.

Overall Conclusion:

- RWAL demonstrates broader antimicrobial activity compared to RWKZ, particularly against Escherichia coli and Staphylococcus aureus at higher concentrations.
- RWKZ shows minimal antimicrobial activity, with slight effectiveness only against Staphylococcus aureus at the highest concentration tested.
- Neither plant extract exhibits anti-Candida activity under the tested conditions.

These results suggest that RWAL may have potential applications as an antimicrobial agent against specific bacterial strains, while RWKZ shows limited potential in this regard.

3.3 Antidiabetic activity:

3.3.1 In vitro antidiabetic activity of RWAL

Analysis of Results

The in vitro antidiabetic activity of RWKL was assessed using a yeast cell model to measure glucose uptake at different concentrations of the extract. The results, as shown in Figure 1, indicate that the glucose uptake by yeast cells increases over time across all concentrations.

At T15, the 20 mg/mL concentration showed the highest glucose uptake (5.68%), while at T30, the 50 mg/mL concentration led to a notable increase (6.28%). By T60, the 10 mg/mL concentration resulted in the highest uptake (12.47%), and at T120, both the 10 mg/mL (15.3%) and 20 mg/mL (15.06%) concentrations demonstrated the most significant glucose uptake.

Overall, the 10 mg/mL concentration consistently showed a significant increase in glucose uptake across all time points, indicating that it may be the most effective concentration for enhancing glucose absorption by yeast cells. This suggests a potential optimal concentration for antidiabetic activity in this model.

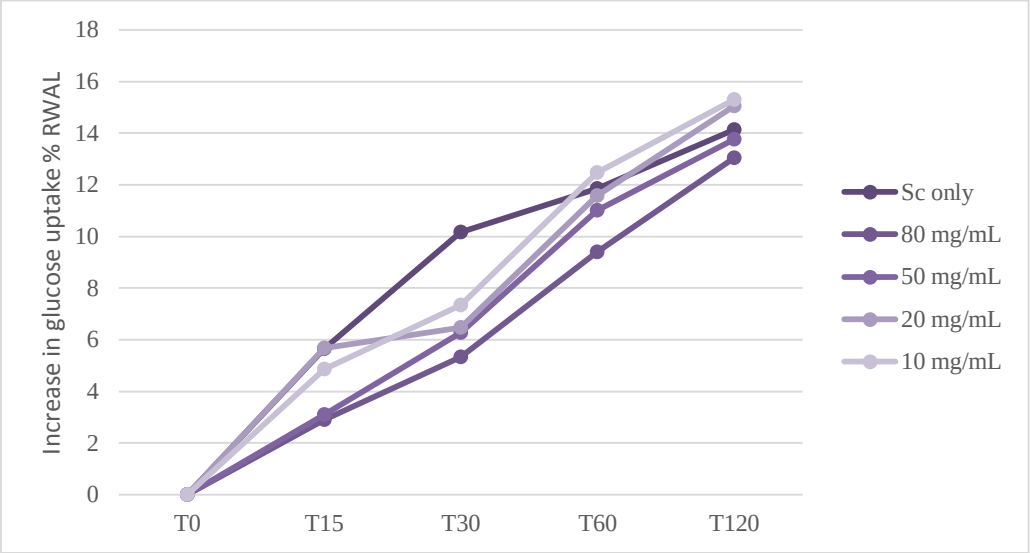


Figure 09: In vitro antidiabetic activity of RWAL using a yeast cell model represented % glucose uptake by yeast cells mediated by different extract concentration; Sc = yeast of *Saccharomyces cerevisiae.*

3.3.2 In vitro antidiabetic activity of RWKZ

Analysis of Results

The in vitro antidiabetic activity of RWKZ was evaluated by measuring glucose uptake in yeast cells at different concentrations of the extract over various time points. The table presents the percentage increase in glucose uptake.

At T15, the 20 mg/mL concentration showed the highest increase in glucose uptake (4.98%), which is slightly higher than the Sc only control (5.65%). At T30, the 10 mg/mL concentration demonstrated the highest uptake (6.85%), indicating a strong response at this concentration. By T60, the 20 mg/mL concentration again showed the highest uptake (13.79%), outperforming other concentrations. Finally, at T120, the 20 mg/mL concentration

continued to lead with the highest uptake (16.43%), followed closely by the 10 mg/mL concentration (15.43%).

The 20 mg/mL concentration consistently showed a significant increase in glucose uptake, particularly at T60 and T120, suggesting it may be the most effective concentration for enhancing glucose absorption by yeast cells in this model. This pattern highlights its potential optimal concentration for antidiabetic activity.

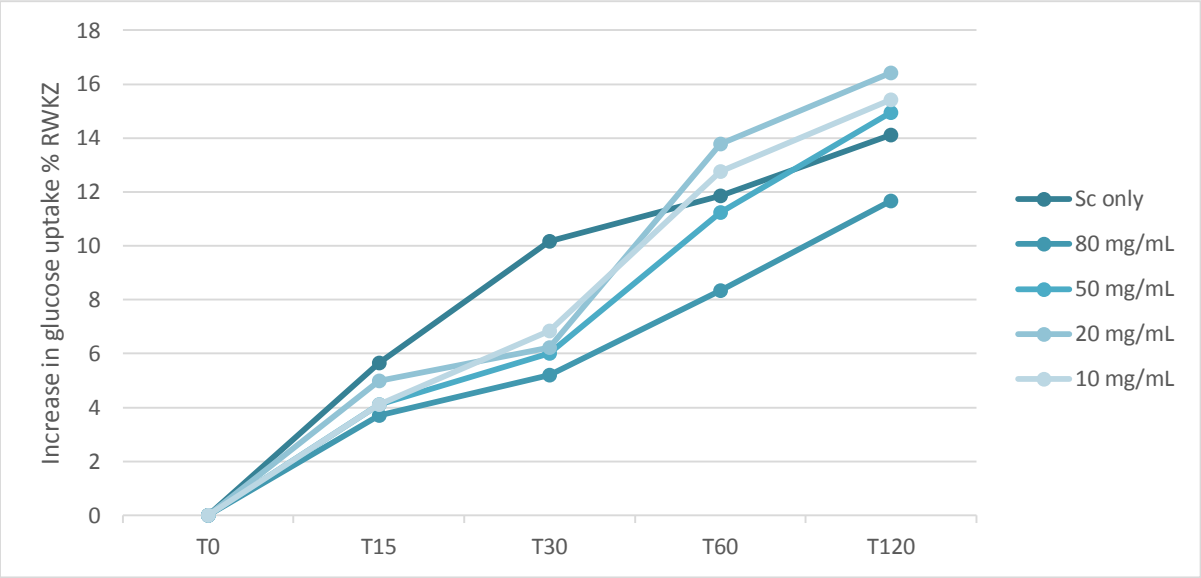


Figure 10: In vitro antidiabetic activity of RWKZ using a yeast cell model represented % glucose uptake by yeast cells mediated by different extract concentration; Sc = yeast of *Saccharomyces cerevisiae.*

3.3.3 Comparative Analysis of RWKL and RWKZ

The effectiveness of the two samples RWKL and RWKZ in increasing glucose uptake in yeast cells was assessed at various concentrations over multiple time points. Here is a comparative analysis:

RWKL:

- **Most Effective Concentration**: 10 mg/mL
- **Key Observations**:
    - At T60, 10 mg/mL showed the highest uptake (12.47%).

- At T120, both 10 mg/mL (15.3%) and 20 mg/mL (15.06%) demonstrated significant glucose uptake.

RWKZ:

- **Most Effective Concentration**: 20 mg/mL
- **Key Observations**:
  - At T60, 20 mg/mL showed the highest uptake (13.79%).
  - At T120, 20 mg/mL led with the highest uptake (16.43%).

General Conclusion

Phytochemistry, the study of the chemical compounds in plants, reveals the presence of bioactive compounds that have various biological activities and potential therapeutic applications. In this study, the focus was on the phytochemical analysis of two plants, Lavender (Lavandula) and Ephedra, particularly their alkaloid content.

The ethanolic extraction yield was found to be 19.12% for Lavender and 14.34% for Ephedra.

Rapid tests:

Mayer's Test : A white precipitate appeared in the lavender test tube after 24 hours, indicating the presence of alkaloids. For Ephedra, a very slight white precipitate appeared after more than 48 hours, suggesting a lower concentration of alkaloids compared to Lavender.

Wagner's Test: Dark brown sediment was present in the lavender test tube, confirming the presence of alkaloids. In contrast, no sediment was observed in the Ephedra test tube, indicating either the absence or a very low concentration of alkaloids.

Dragendorff's Test: Dark red sediment appeared in both Lavender and Ephedra test tubes, confirming the presence of alkaloids in both plant extracts.

This study demonstrates that both Lavender and Ephedra contain alkaloids, although in differing concentrations. Lavender showed a more significant presence of alkaloids across all tests compared to Ephedra, which had a lower concentration as evidenced by the delayed and less intense reactions in Mayer's and Wagner's tests. The presence of alkaloids in both plants supports their traditional use in medicine and suggests potential for further pharmaceutical applications.

Biological Activities

Lavender (Lavandula):

Anti-inflammatory and Antioxidant Activity: Lavender contains compounds like linalool and linalyl acetate, which exhibit significant anti-inflammatory and antioxidant properties. These compounds help reduce inflammation and oxidative stress, contributing to the plant's therapeutic effects.

Antimicrobial Activity: Lavender oil has been shown to possess strong antimicrobial properties against a variety of bacterial and fungal pathogens. This makes it useful in treating infections and in the formulation of antiseptic products.

Sedative and Anxiolytic Effects: The presence of linalool is also associated with sedative and anxiolytic effects, making lavender useful in managing anxiety and promoting relaxation.

Ephedra:

Stimulant and Bronchodilator Activity: Ephedrine, the primary alkaloid in Ephedra, is a well-known stimulant that increases heart rate and blood pressure. It also acts as a bronchodilator, making it effective in treating conditions like asthma and bronchitis.

Weight Loss and Metabolic Effects: Ephedrine is known to boost metabolism and promote weight loss. This is due to its ability to increase thermogenesis and fat oxidation.

Anti-inflammatory and Analgesic Effects: Compounds in Ephedra, including ephedrine, have been shown to possess anti-inflammatory and analgesic properties, which help in managing pain and inflammation.

Perspectives

Future investigations should focus on the quantitative analysis of specific alkaloids present in these plants and their pharmacological activities. Given the lower concentration of alkaloids in Ephedra, it may be beneficial to explore other extraction methods or conditions that might enhance yield. Additionally, expanding the study to include other bioactive compounds such as flavonoids and terpenes could provide a more comprehensive understanding of the medicinal potential of these plants. This research contributes to the growing body of evidence supporting the use of plant-derived compounds in the development of new therapeutic agents.

Appendices