PEOPLE'S DEMOCRATIC REPUBLIC OF ALGERIA
MINISTRY OF HIGHER EDUCATION
AND SCIENTIFIC RESEARCH
KASDI MERBAH OUARGLA UNIVERSITY
Faculty of Applied Sciences
Department of Process Engineering

Memory

Presented for the graduation of

ACADEMIC MASTER

Field: Science and Technology

Sector: Process Engineering

Specialty: Petrochemical engineering

Presented by: Hassan Al-ansi and Abd-Elhak Saidi

Theme

Valorization of the essential oils of the *Geranium Rose* plant

Publicly supported on: 13/06/2020

The jury:

Ms. Zobeidi naoual	**MCB**	**in KMU Ouargla**	**President**
Ms. Shoeeb hageera	**MCC**	**in KMU Ouargla**	**Examiner**
Mr. Ladjel Segni	**Pr**	**in KMU Ouargla**	**Supervisor**

Year of study: 2020 -2021

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DEDICATION

Thanks to almighty Allah who gave me the courage, the will and the strength to make this thesis.

I dedicate this humble work:

To my dear *parents*

Zaid and *Houria*

To my *Sisters* and my *Brothers*

Karima

Asma

Naseem

Somaya

Abdulohaab

yousef

Osama

Hassan

ACKNOWLEDGEMENT

At the end of this work, our thanks gose first to the almighty ALLAH who allowed us to complete our studies.

We would like to thank the Professor Ladjel Segni, the supervisor of this thesis, for agreeing to supervise this modest work and for his invaluable help on several levels (documentation, knowledge, guidance, moral support).

We address our sincere thanks in general to all the teachers who have taught us, always supported and encouraged us during our study and the preparation of this dissertation.

We thank the members of the jury; each has his name, for agreeing to judge our work.

Madame Zobeidi naoual

Madame Shoeeb hageera

Finally, we address more sincere thanks to all the friends who have always encouraged us during the preparation of this brief.

THANK YOU All

ABSTRACT

The objective of our work is to extract essential oils from Geranium Rose from the Toggourt region and to determine their chemical composition, physico-chemical analysis and industrial applications.

The extraction by hydro-distillation of the Geranium Rose gave a yield of 0.12%. Analysis by gas chromatography coupled with mass spectrometry the plant contains the majority compounds in the following proportions: Citronellol 18.44% and trans-Farnesol 7.05%.

The oils are separated and identified with GC / MS In addition, these oils are important and have great applications in different fields, pharmaceutical, medical and cosmetic also used in biological control as a biopesticide.

Keywords: Essential oil, Geranium Rose, GC / MS, Biopesticide.

RESUME

L'objectif de nos travaux est d'extraire les huiles essentielles de Géranium Rose de la région de Toggourt et de déterminer la composition chimique, l'analyse physico-chimique et applications industrielles.

L'extraction par hydro-distillation du Géranium Rose a donné un rendement de 0.12%. L'analyse par chromatographie en phase gazeuse couplée à la spectrométrie de masse la plante contient les composés magoritaires dans les proportions suivantes : Citronellol 18,44% et trans-Farnesol 7.05%.

Les huiles sont séparées et identifiées avec GC / MS de plus, ces huiles sont importantes et ont de grandes applications dans diffèrents domaines, pharmaceutiques, medicales et cosmétiques utilisés aussi dans la lutte biologique comme biopesticide.

Mots clés : Huile essentielle, Géranium Rose, GC / MS, Biopesticide.

الملخص

الهدف من عملنا هو استخراج الزيوت الأساسية من Geranium Rose من منطقة تقرت وتحديد التركيب الكيميائي والتحليل الفيزيائي والكيميائي والتطبيقات الصناعية.

أعطى الاستخلاص بالتقطير المائي ل Geranium Rose عائد 0.12% .

يحتوي المصنع على غالبية المركبات بالنسب التالية بالتحليل الكروماتوجرافي الغازي مقترنًا بقياس الطيف الكتلي: Citronellol 18.44٪ و Trans-Farnesol 7.05٪

يتم فصل الزيوت وتحديدها باستخدام GC / MS بالإضافة إلى أن هذه الزيوت مهمة ولها تطبيقات عديدة في مختلف المجالات الصيدلانية والطبية والتجميلية المستخدمة أيضًا في المكافحة البيولوجية كمبيد حيوي.

الكلمات المفتاحية: زيت عطري، Geranium Rose، GC / MS، مبيدات الآفات الحيوية.

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FIGURE CONTENT

ABBREVIATIONS CONTENT

AFNOR: French Standardization Association.

GC / MS: Gas chromatography coupled with a mass spectrometer.

EO: Essential Oil.

HD: Hydro distillation.

EOs: Essential Oils.

RI: Linear retention indices.

W: weight in grams.

W_{EO}: Weight of oil in gram.

W_S: Weight of the sample in gram.

AN: Acid Number.

M: Molar mass.

ρ**:** Specific mass (density)

W_0: Weight of empty specific mass needle.

W_1: Weight of water + specific mass needle.

W_2: Weight of test sample + specific mass needle.

CONTENT

CHAPTER I: GENERALITY ON ESSENTIAL OILS

CHAPTER II: TECHNIQUES FOR EXTRACTING ESSENTIAL OILS

CHAPTER III: GREEN CHEMISTRY

CHAPTER IV: GERANIUM ROSE

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GENERAL INTRODUCTION

GENERAL INTRODUCTION

The objective of the research is to evaluate the essential oils of Geranium Rose and study its use as a natural pesticide to compensate for chemical pesticides that cause serious problems like cancer.

Humankind has used plants for healing for many thousands of years, and it is from this tradition of that the use of aromatic plant compounds is medicine began. Oils were used in the embalming process, in medicine and in purification rituals. There are also over 200 references to aromatics, incense and ointments in the Old and New Testaments; Frankincense, Myrrh, Galbanum, Cinnamon, Cassia, Rosemary, Hyssop and Spikenard are noted for being used for anointing rituals and healing of the sick [1].

Essential oils are highly concentrated volatile substances extracted from various parts of certain plant species, each with specific therapeutical and energetic effects. These volatile liquids are very complex molecular substances, extremely potent and precise as action. Essential oil is not actually an oil because it contains no fatty substance. It is obtained from the essence rich in natural flavors and active ingredients that it secretes the cells of certain parts of the plant. Precious liquids are obtained by distilling or pressing the secretory organs. For example, citrus peel is cold pressed, and the other parts of the plant (stem, leaves, flowers, root, wood) are distilled [2-3]. These processes result in an aromatic concentrate and a genuine source of active substances. Essential oil is also known as volatile oil or ethereal oil [4].

Extraction of essential oils is expensive because of the large amount of raw material required to produce a few milliliters of oil. This explains the high prices required for genuine essential oils. For example, to obtain a single drop of essential rose oil, approx. 60 roses [5]. However, there are also less expensive oils due to the abundance of inexpensive raw materials and high productivity. Such oils are citrus–lemon, orange, bergamot, lime, lemongrass oil, tea tree oil. So, the essential oil is very precious, but only one drop is sufficient for beneficial results, moreover, overtaking a 2 % dosage is toxic and produces adverse effects [6-7].

New chemistry is required to improve the economics of chemical manufacturing and to enhance the environmental protection. The green chemistry concept presents an attractive technology to chemists, researchers, and industrialists for innovative chemistry research and applications.

Primarily, green chemistry is characterized as reduction of the environmental damage accompanied by the production of materials and respective minimization and proper disposal of wastes generated during different chemical processes. According to another definition, green chemistry is a new technique devoted to the synthesis, processing, and application of chemical materials in such manner as to minimize hazards to humankind and the environment [8].

Geranium for instance, yield oil both from the flowers and the leaves, and the oil from both parts differ in constituents, scents and some other properties. The quantity of essential oil extracted from the plant is determined by many interrelated factors, climatic, seasonal and geographical conditions, harvest period and extraction techniques [9]. This fraction is characterized by the complexity in the separation of its components, which belong to various classes of compounds and which are present in a wide range of concentrations. Therefore it is complicated to establish a composition profile of essential oils. The gas chromatographic method (GC) is almost exclusively used for the qualitative analysis of volatiles. The analysis of essential oils was developed in parallel with the technological developments in GC, such as stationary phases, detection devices, etc. However, advances in instrumentation were not the only important factor in the development of analytical methods for essential oils in plants. Sample extraction and concentration were also improved. The most outstanding improvements in the determination of the composition of essential oils came from the introduction of tandem techniques involving prior/further chromatography or spectroscopy.

The great amount of information on the application of GC and hyphenated techniques to essential oils has led to much research in this field [10].

The writing of this letter is organized according to the following diagram:

Theoretical part	**practice part**
Generality on essential oils	Materials and methods
Techniques for extracting	Results and discussions
Green chemistry	
Geranium rose.	

CHAPTER I
GENERALITY ON ESSENTIAL OILS

I.GENERALITY ON ESSENTIAL OILS:

I.1.What are Essential Oils?

Essential oils are aromatic substances present in the specialized cells or glands of certain plants used by them to protect themselves from predators and pests, but also to attract polinators. In other words, essential oils are part of the immune system of the plant. The famous alchemist, physician, physicist, astrologer, theologian and philosopher of Switzerland, Paracelsus, called distilled oils from herbs–quinta essentia–the quintessence of the plant, and hence the name of essential oils [11].

Essential oils are highly concentrated volatile substances extracted from various parts of certain plant species, each with specific therapeutical and energetic effects. These volatile liquids are very complex molecular substances, extremely potent and precise as action. Essential oil is not actually an oil because it contains no fatty substance. It is obtained from the essence rich in natural flavors and active ingredients that it secretes the cells of certain parts of the plant. Precious liquids are obtained by distilling or pressing the secretory organs. For example, citrus peel is cold pressed, and the other parts of the plant (stem, leaves, flowers, root, wood) are distilled. These processes result in an aromatic concentrate and a genuine source of active substances. Essential oil is also known as volatile oil or ethereal oil.

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I.2.Spreading

Essential oils are relatively widespread in the plant kingdom, some families being very rich in such substances, both in number and quantity. Typically, essential oils are found in superior plants (about 50 families) belonging to orders of angiosperms (Asterales, Laurales, Magnoliales, Zimgiberales, etc.) or ginsenosides (Pinales), but also known as sesquiterpenic lactone sesquiterpene volatile, or algae that produce halogenated sesquiterpenes. Although terpenic compounds are characteristic of the plant kingdom, some biosynthesized monoterpenes have been reported from soil bacteria, insects (probably pheromones), and some sesquiterpene and diterpenes of animal origin [12-13]. The synthesis and accumulation of essential oils occur either outside the plant, in the glandular brushes (Asteraceae, Geraniaceae, Laminaceae,etc.) and in the papillae, either inside the plant, in the secretory cells, in the intercellular spaces (secretory channels) secretory bags (Anacardiaceae, Rutaceae, Myrtaceae). Essential oils can accumulate in all plant organs, but in varying amounts. Thus we can meet them in: roots, leaves, flowers, fruits, wood of the stems or in the bark. The content in essential oils of plants is often below 1 %, rarely reaching 15 % or even more, in the dry product of some plants. The name of aromatic plants is attributed to those species which contain a higher amount of volatile oil (at least 0.1–0.2 %), which have a sufficiently perceptible odor or which lend themselves to economically viable exploitation [14-15]. In addition, there are other species that, although characteristically smell, still contain therapeutic substances that are comprised of essential oils.

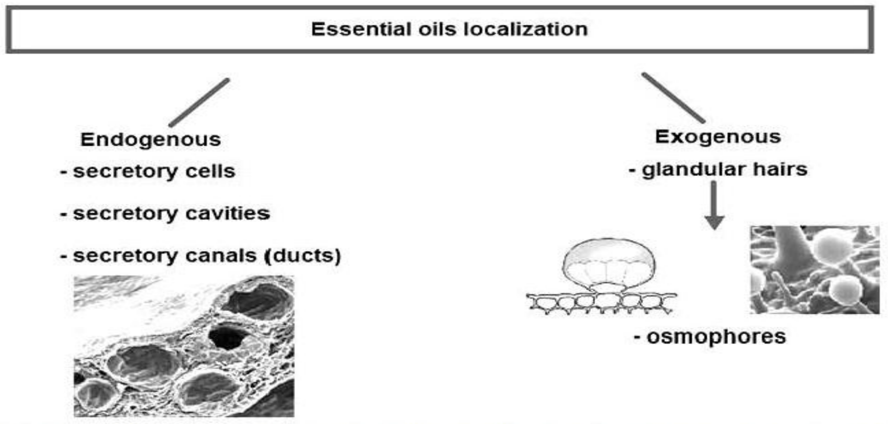

Figure 01: Presence of Essential oils in plant organs. Source: Preedy et al [16].

I.3.Biosynthesis of Essential Oils

The biosynthesis of odorous substances takes place in the leaves, where most of them are found and remains until flowering. Flowering, essential oils migrate into flowers, and part is consumed in the fertilization process. After fertilization, it accumulates in fruits and seeds or there is a migration to leaves, bark and root [17-18]. During the maturation of plants, the composition of essential oils changes: in young plants they contain mainly terpenic hydrocarbons and simpler molecules, while the reproductive organs contain etheric oils richer in oxygenated compounds. Although their role in the plant organism is partly known, ethereal oils have multiple uses. There are more than 3,000 essential oils that are physically and chemically characterized , about 150 of which are manufactured on an industrial scale [19-20].

I.4.Uses of Essential Oils

They are used in certain medicines, in perfumery, in herbal medicine or as a flavoring agent in food. We must distinguish the activity of the essential oil and that of the infused plant. There is often a threshold, beyond which they can become toxic. The use of plants and oils is controlled by the public health code. For several years, essential oils have invaded many products of everyday life. They are found more and more as food flavors as flavor enhancers (coffees, teas, tobacco, wines, yoghurts, ready meals, etc.). Cosmetics and mainly organic cosmetics is also a sector that uses more and more essential oils. They are found in many products such as: soaps, shampoos, shower gel, creams, etc. EOs are used for example as phyto products -sanitary to combat fungal, bacterial or viral infections in plant crops. They provide solutions in organic farming, reducing the harmful effects of synthetic pesticides such as pollution or the development of resistance.Akkadian texts dating back over four thousand years tell us that in Babylon, cypress was burned to stop epidemics. . The earliest texts relating to the use of fine oils and perfumes are Egyptian hieroglyphic papyri dating back over 2,800 years. Chinese and Indian civilizations also used essential oils for therapeutic and cosmetic care.

Some oils are dermocaustic (aggressive for the skin), like oregano, other photosensitizers like citrus. Therefore, we must act with great care and respect these few basic rules:

- Never apply a pure essential oil on the skin and especially on the mucous membranes.
- The essential oil must be very strongly diluted in a support such as a vegetable oil.
- Some essential oils can be irritating.
- Avoid expsosure to the sun after applying an essential oil, because some essential oils (especially those of Citrus) are photosensitizing (increased sensitivity to UV), or can cause the appearance of unsightly pigmented spots on the skin.
- In aromatic cosmetology, between 0.5% and 2% EO is used for the face, 2% and 5% for the body, and up to 10% for very localized treatments [21-22].

I.5.Major raw material used in extraction of essential oils:

Essential Oils are derived from various parts of Plants:

Leaves	Flowers	Peel	Seeds	Wood
Basil Bay leaf Cinnamon Eucalyptus Lemon Grass Melaleuca Oregano Patchouli Peppermint Pine Rosemary Spearmint Tea Tree Wintergreen Thyme	Chamomile Clary Sage Clove Geranium Hyssop Jasmine Lavender Manuka Marjoram Orange Rose Ylang-Ylang	Bergamot Grape fruit Lemon Lime Orange Tangerine	Almond Anise Celery Cumin Nutmeg Oil	Camphor Cedar Rosewood Sandalwood
Berries	**Bark**	**Resins**	**Rhizome**	**Root**
Allspice Juniper	Cassia Cinnamon	Frankincense Myrrh	Ginger	Valerian

Table 01: 1 Parts of plant material containing essential oils [23].

I.6.What is Aromatherapy?

"*The treatment of anxiety or minor medical conditions by rubbing pleasant smelling natural oils into the skin or breathing in their smell*."

It is the use of aromatic essential oils to benefit the body – in emotional and physical health and beauty. Science has discovered that our sense of smell plays a significant role in our overall health.

Many common essential oils have medicinal properties that have been applied in medicine since ancient times and are still widely used today. For example, many essential oils have antiseptic properties, though some are stronger than the other. In addition, many have an uplifting effect on the mind, though different essential oils have different properties.

The Beginnings of Modern Aromatherapy

The first modern-day distillation of essential oil was performed by the Persian philosopher Avicenna (980-1037 A.D.) who extracted the essence of rose petals through the *'enfleurage'* process. His discovery and subsequent use of a wonderful perfume substance eventually lead him to write a book on the healing properties of essential oil of Rose.

Early in the 20th century a French Chemist, Rene-Maurice Gattefosse, began studying what he called " *Aromatherapy* ." After several burning his arm in a laboratory accident, he thrust the arm into the nearest liquid, which happened to be tub of Lavender Oil. Surprised by the quick healing that followed, Dr. Gattefosse spent the remainder of his life researching the value of Essential Oils. His success made aromatherapy popular, and it became well-known in Europe.

How Essential Oil Works in Aromatherapy?

An Essential Oil is inhaled and directly by the olfactory system to the limbic System of the Brain. In true, the brain responds to the particular scent affecting our emotions and chemical balance. Essential Oils also absorbed by the skin and carried throughout the body via the circulatory system to reach all internal organs.

By carefully choosing one or more oils, you can experience benefical effects promoting overall health - and even specific targets. Benefits depend upon the unique nature of each person's response to an aromatic stimulus [24].

I.7. Pharmacological Properties of Essential Oils

- **Antiseptics:**

Essential oils have antiseptic properties and are active against a wide range of bacteria as well as on antibio-resistant strains. Moreover, they are also known to be active against fungi and yeasts (Candida).The most common sources of essential oils used as antiseptics are: Cinnamon, Thyme; Clover; Eucalyptus; Culin savory; Lavender. Citral, geraniol, linalool and thymol are much more potent than phenol**.**

- **Expectorants and diuretics:**

When used externally, essential oils like (L'essence de terebenthine) increase microcirculation and provide a slight local anaesthetic action. Till now, essential oils are used in a number of ointments, cream and gels, whereby they are known to be very effective in relieving sprains and other articular pains. Oral administration of essential oils like eucalyptus or pin oils, stimulate ciliated epithelial cells to secrete mucus. On the renal system, these are known to increase vasodilation and in consequence bring about a diuretic effect.

- **Spasmolytic and sedative:**

Essential oils from the Umbellifereae family, Mentha species and verbena are reputed to decrease or eliminate gastrointestinal spasms. These essential oils increase secretion of gastric juices. In other cases, they are known to be effective against insomnia.

- **Others**:

Cholagogue; anti-inflammatory; cicatrizing [25].

I.8.Chemical Constituents of Essential Oils

Pure essential oils are mixtures of more than 200 components, normally mixtures of terpenes or phenylpropanic derivatives, in which the chemical and structural differences between compounds are minimal. They can be essentially classified into two groups:

- **Volatile fraction:** Essential oil constituting of 90–95% of the oil in weight, containing the monoterpene and sesquiterpene hydrocarbons, as well as their oxygenated derivatives along with aliphatic aldehydes, alcohols, and esters.
- **Nonvolatile residue:** that comprises 1–10% of the oil, containing hydrocarbons, fatty acids, sterols, carotenoids, waxes, and flavonoids [26].

I.8.1. Hydrocarbon:

Essential Oils consist of Chemical Compounds that have hydrogen and carbon as their building blocks. Basic Hydrocarbon found in plants are isoprene having the following structure [27].

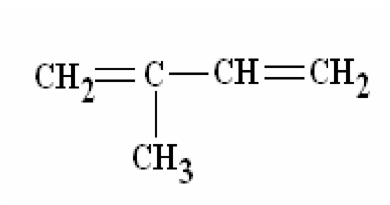

Figure 02: Chemical structures of Isoprene [27].

I.8.2. Terpenes:

Generally have names ending in "**ene**"

For examples: Limonene, Pinene, Piperene, Camphene, etc. Terpenes are anti-inflammatory, antiseptic, antiviral, and bactericidal. Terpenes can be further categorized in monoterpenes, sesquiterpenes and diterpenes. Referring back to isoprene units under the Hydrocarbon heading, when two of these isoprene units join head to tail, the result is a monoterpene, when three join, it is a sesquiterpene and four linked isoprene units are diterpenes [28].

I.8.2.a. Monoterpenes [$C_{10}H_{16}$]

Properties: Analgesic, Bactericidal, Expectorant, and Stimulant.

Monoterpenes are naturally occurring compounds, the majority being unsaturated hydrocarbons (C_{10}).But some of their oxygenated derivatives such as alcohols, Ketones, and carboxylic acids known as monoterpenoids.

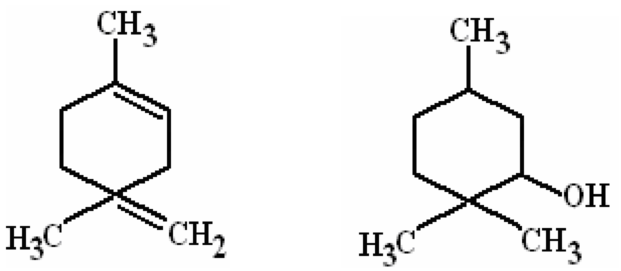

Figure 03: Chemical structures of Limonene and Menthol [28].

The branched-chain C10 hydrocarbons comprises of two isoprene units and is widely distributed in nature with more than 400 naturally occurring monoterpenes identified. Moreover, besides being linear derivatives (Geraniol, Citronellol), the monoterpenes can be cyclic molecules (Menthol – Monocyclic; Camphor – bicyclic; Pinenes (α and β) – Pine genera as well. Thujone (a monoterpene) is the toxic agent found in Artemisia absinthium (wormwood) from which the liqueur, absinthe, is made. Borneol and camphor are two common monoterpenes. Borneol, derived from pine oil, is used as a disinfectant and deodorant. Camphor is used as a counterirritant, anesthetic, expectorant, and antipruritic, among many other uses.

Example :

- Camphene and pinene in cypress oil.
- Camphene, pinene and thujhene in black pepper [29].

I.8.2. b. Sesquiterpenes

Properties: anti-inflammatory, anti-septic, analgesic, anti-allergic.

Sesquiterpenes are biogenetically derived from farensyl pyrophosphate and in structure may be linear, monocyclic or bicyclic. They constitute a very large group of secondary metabolites, some having been shown to be stress compounds formed as a result of disease or injury.

Sesquiterpene Lactones:

Over 500 compounds of this group are known; they are particularly characteristics of the Compositae but do occur sporadically in other families. Not only have they proved to be of interest from chemical and chemotaxonomic viewpoints, but also possess many antitumor, anti-leukemia, cytotoxic and antimicrobial activities. They can be responsible for skin allergies in humans and they can also act as insect feeding deterrents. Chemically the compounds can be classified according to their carboxylic skeletons; thus, from the germacranolides can be derived the guaianolides, pseudoguaianolides, eudesmanolides, eremophilanolides, xanthanolides, etc.

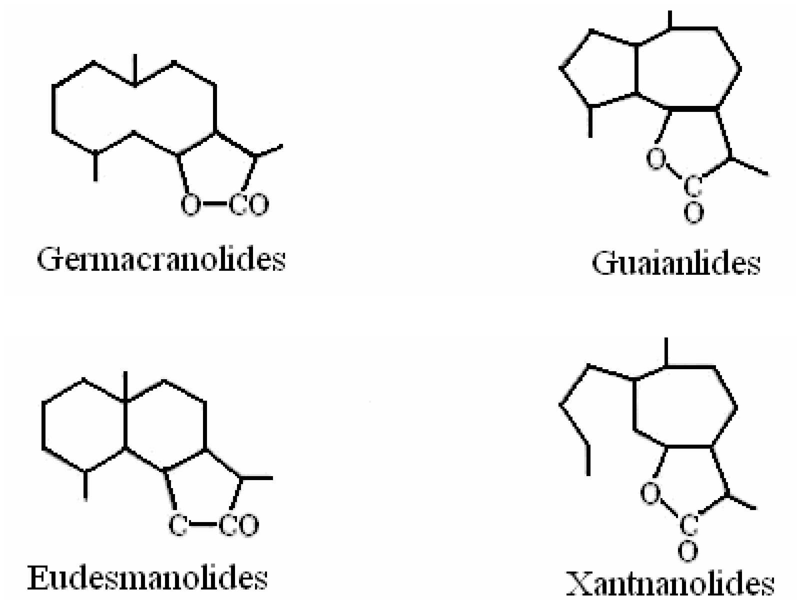

Figure 04: Chemical structures of all these compounds [30].

A structural feature of all these compounds, which appears to be associated with much of the biological activity, is the α, β -unsaturated- γ- lactones.

Example :

- Farnesene in chamomile and lavender.
- Beta-caryophyllene in basil and black pepper [30-31].

I.8.2.c. Diterpenes

Properties: anti-fungal, expectorant, hormonal balancers, hypotensive

Diterpenes are made of up four isoprene units. This molecule is too heavy to allow for evaporation with steam in the distillation process, so is rarely found in distilled essential oils. Diterpenes occur in all plant families and consist of compounds having a C20 skeleton. There are about 2500 known diterpenes that belong to 20 major structural types. Plant hormones Gibberellins and phytol occurring as a side chain on chlorophyll are diterpenic derivatives. The biosynthesis occurs in plastids and interestingly mixtures of monoterpenes and diterpenes are the major constituents of plant resins. In a similar manner to monoterpenes, diterpenes arise from metabolism of geranyl geranyl pyrophosphate (GGPP).

Diterpenes have limited therapeutical importance and are used in certain sedatives (coughs) as well as in antispasmodics and antoxiolytics.

Example :

- Sclareol in clary sage is an example of a diterpene alcohol [32].

I.8.3. Alcohols:

Properties: anti-septic, anti-viral, bactericidal and germicidal.

Alcohols are the compounds which contains Hydroxyl compounds. Alcohols exist naturally, either as a free compound, or combined with a terpenes or ester. When terpenes are attached to an oxygen atom, and hydrogen atom, the result is an alcohol. When the terpene is monoterpene, the resulting alcohol is called a monoterpenol. Alcohols have a very low or totally absent toxic reaction in the body or on the skin. Therefore, they are considered safe to use.

Example :

- linalool found in ylang-ylang and lavender.
- Geraniol in geranium and rose.
- Nerol in neroli [33].

I.8.4. Aldehydes:

Properties : anti-fungal, anti-inflammatory, anti-septic, anti-viral, bactericidal, disinfectant, sedative.

Medicinally, essential oils containing aldehydes are effective in treating Candida and other fungal infections.

Example :

- Citral in lemon.
- Lemongrass and lemon balm.
- Citronellal in lemongrass, lemon balm and citrus eucalyptus.

I.8.5. Acids:

Properties: anti-inflammatory.

Organic acids in their free state are generally found in very small quantities within Essential oils. Plant acids act as components or buffer systems to control acidity.

Example :

- Cinnamic and benzoic acid in benzoin.
- Citric and lactic.

I.8.6. Esters:

Esters are formed through the reaction of alcohols with acids. Essential oils containing esters are used for their soothing, balancing effects. Because of the presence of alcohol, they are effective antimicrobial agents. Medicinally, esters are characterized as antifungal and sedative, with a balancing action on the nervous system. They generally are free from precautions with the exception of methyl salicylate found in birch and wintergreen which is toxic within the system.

Example :

- linlyl acetate in bergamot and lavender.
- Geranyl formate in geranium [34].

I.8.7. Ketones:

Properties: anti-catarrhal, cell proliferant, expectorant, vulnery.

Ketones often are found in plants that are used for upper respiratory complaints. They assist the flow of mucus and ease congestion. Essential oils containing ketones are beneficial for promoting wound healing and encouraging the formation of scar tissue. Ketones are usually (not always) very toxic**.** The most toxic ketone is Thujone found in mugwort, sage, tansy, thuja and wormwood oils. Other toxic ketones found in essential oils are pulegone in pennyroyal, and pinocamphone in hyssops. Some non-toxic ketones are jasmone in jasmine oil, fenchone in fennel oil, carvone in spearmint and dill oil and menthone in peppermint oil.

Example :

- fenchone in fennel, carvone in spearmint and dill
- Menthone in peppermint.

I.8.8. Lactones:

Properties: anti-inflammatory, antiphlogistic, expectorant, febrifuge.

Lactones are known to be particularly effective for their anti-inflammatory action, possibly by their role in the reduction of prostaglandin synthesis and expectorant actions. Lactones have an even stronger expectorant action then ketones [34].

I.9.What is the Life Span of an Essential Oil

Some the oils, such as citrus, have a lower life expectancy than others, such as patchouli and sandalwood, the latter seeming to be getting better as time passes [35]. The average life span of essential oils varies, depending on both the manufacturing process and the conservation methods. Oils should be stored in hermetic and resistant glass containers at temperatures between 15 and 20 degrees C. If the optimal conditions are met, most oils can be stored for at least 3 years. The shorter life span of citrus oils (lemon, orange, etc.) is due to the fact that they are extracted from the fruit bark and the unstable components, such as waxes and fatty acids, also remain in the essential oil [36]. Most the essential oils are extracted using the steam distillation method, and the heat produced during this process changes and stabilizes the natural components of the plant [37-38]. After the distillation, the extracted oils undergo a maturation process and at this stage additional chemical changes occur before the odor can stabilize. This stabilization may take a few weeks, as in the case of peppermint oil [39]. Heavier, heavier oils such as patchouli and sandalwood need more time to manifest their full potential.

I.10 .Essential Oils: Worldwide production

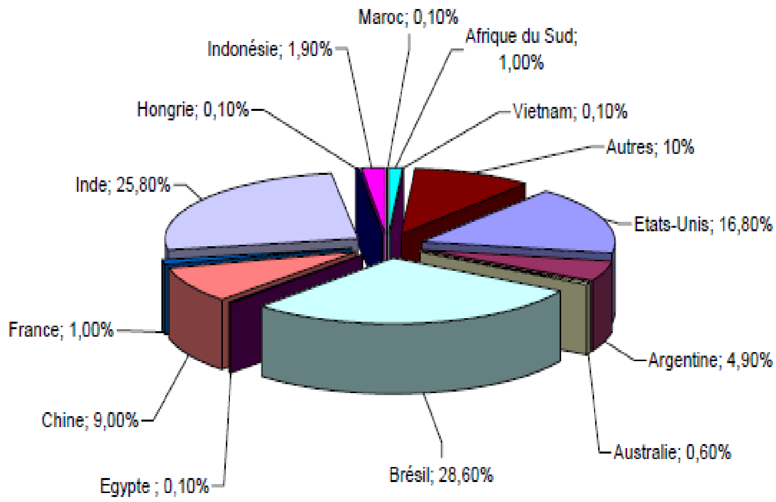

Figure 05: Production countries and Essential oil production worldwide (2008) [40].

- 1850-1950 : France, Italy, Spain, Portugal
- 1950 : North Africa
- 1990 : China, India
- 2010 : India

CHAPTER II

TECHNIQUES FOR EXTRACTING ESSENTIAL OILS

II. TECHNIQUES FOR EXTRACTING ESSENTIAL OILS

II.1.What is the extraction of Essential Oils?

Several parts of various aromatic plants can be extracted and form essential oils which subsequently have many applications in cosmetics, pharmaceutical and food safety fields. The manufacturing method and technique used to extract essential oils are dependent on the characteristics and components required in the botanical extract. The main factor to ensure the quality of essential oils is the extraction method used, since inappropriate extraction procedures may cause the destruction and vary the action of phytochemicals present in aromatic oils. The resulting effects can be, for example, the loss of pharmacological constituents, stain effect, offflavor/ odour, and physical change of essential oils [41].

Such extraction techniques can be categorized into two categories:

classical methods and innovative methods. The application of innovative techniques, such as ultrasonic and microwave enhanced processes, has improved the efficiency of extraction process in terms time required for isolation of the essential oil and energy dissipation, as well as improvement in production yield, and high quality of essential oils [42].

Methods: Several advanced (supercritical fluid extraction, subcritical extraction liquid, solvent-free microwave extraction) and conventional (hydrodistillation, steam distillation, hydrodiffusion, solvent extraction) methods have been discussed for the extraction of essential oils. Advanced methods are considered as the most promising extraction techniques due to less extraction time, low energy consumption, low solvent used and less carbon dioxide emission.

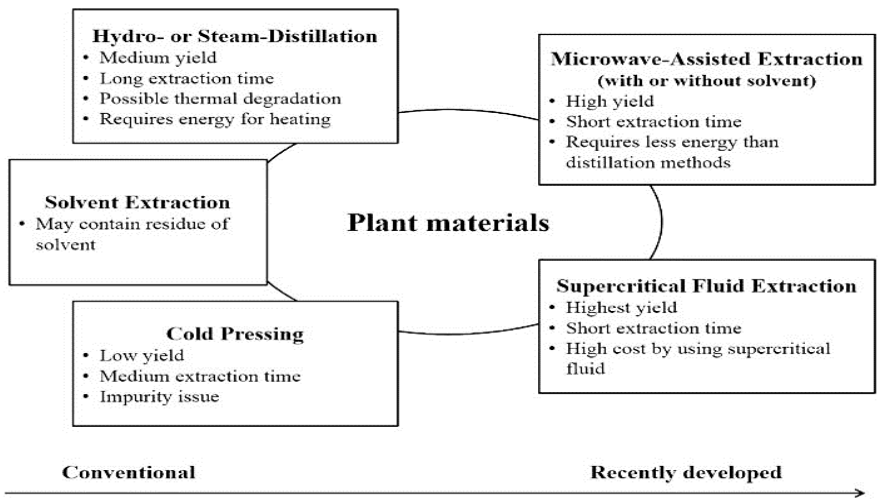

Figure 06: Different methods to extract essential oils adapted from Preedy et al. [16].

II.2. Extraction Methods for Essential Oils

II.2.1. Hydrodistillation

Hydrodistillation is the oldest and simplest oils extraction method which was discovered by Avicenna and the first to develop extraction through the alembic. Rose was the first plant extract used and purified by this method. The procedures start with immersing the plant materials directly into water inside the alembic (vessel), and whole mixture was boiled. The devices include a heating source, vessel (Alembic), a condenser to convert vapor from vessel onto liquid, and a decanter to collect the condensate and to separate essential oils with water (Fig II.2).

This extraction technique is considered as a unique method to extract plant materials like wood or flower and is frequently used for extractions involving hydrophobic natural plant material with a high boiling point. As the oils are surrounded by water, this method is able to protect essential oils to be extracted at a certain degree

without being overheated. The main advantage of this extraction technique is its ability to isolate plant materials below 100°C [42].

Few studies have been conducted on the extraction of essential plant oils by using hydrodistillation. Okoh *et al.* investigated the comparison between extraction process, Hydrodistillation (HD) and Solvent-free Microwave Extraction (SFME) on the properties and yield of essential oil from rosemary (*Rosmarinus officinalis* L.).

Through hydrodistillation, a total yield of the volatile fraction was 0.31%, while 0.39% was obtained for the SFME method [43].

The general hydrodistillation process has been modified by using new technologies as reported by a few researchers. Golmakani and Rezaei developed advanced HD extraction process technique named Microwave-assisted HD (MAHD) which showed superiority in energy dissipation and isolation period (75 min compared to 4 h in HD) [44]. Additionally, Ohmic-assisted HD (OAHD) is the other advanced HD extraction technique, discovered by Gavahian and co-workers. Through OAHD, thyme essential oil can be extracted in a period of only 25 min compared to HD method. No change was observed in characteristics of components in thyme obtained by OAHD and HD [45].

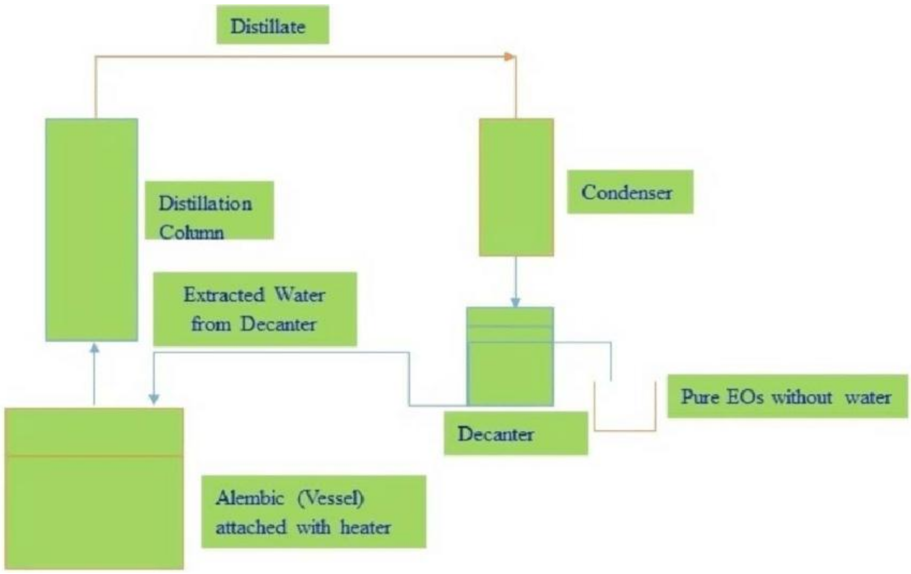

Figure 07: Flow diagram of hydrodistillation extraction process [42].

II.2.2. Steam Distillation

In essential plant oil extraction, steam distillation method is the broadest technique applied. The percentage of essential oils being extracted by this technique is 93% and the remaining 7% can be further extracted by other methods [46]. Basically, the process started by heating of plant material using steam which is supplied from steam generator (Fig II.4). Heat is the main factor determining how effectively the plant material structures break down and burst and release the aromatic components or essential oils [47].

Masango developed an innovative steam distillation extraction technique to increase the isolated essential oil yields and reduce the amount of wastewater produced during the extraction process. The system uses a packed bed of the plant samples, placed above the steam source. Only steam is allowed to pass through the plants and boiling water does not mix with the botanical materials. Therefore, the process requires less steam and the amount of water in the distillate can be reduced [46].

In another study, Yildirim *et al.* reported a component 2,2- diphenyl-1-picryl hydrazyl (DPPH) used to evaluate the antioxidant properties of essential oils by using steam distillation extraction process. It was reported to have a higher yield of antioxidant components than the oils extracted by hydrodistillation (HD) [48].

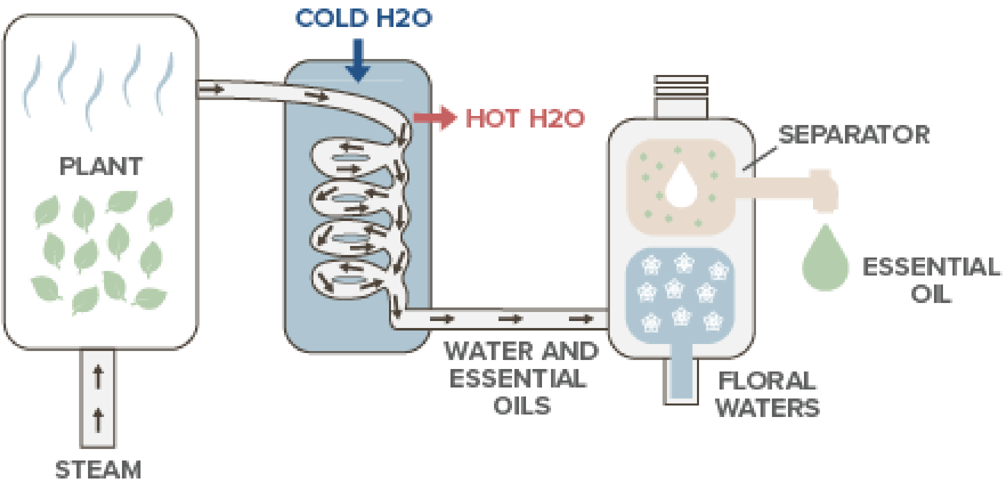

Figure 08: Diagrammatic illustration of steam distillation method [41].

II.2.3. Hydrodiffusion

Hydrodiffusion extraction method is an extraction process in which steam is supplied to a container which holds plant materials.

This technique is only applied on dried plant samples that can be damaged at boiling temperature. In the steam distillation process, steam is applied from the bottom of the steam generator, whereas in the hydrodiffusion method, steam is supplied from the top of the generator. This process was carried out at low pressure or vacuum and steam temperature can be reduced below 100°C [49].

This steam diffusion method was further enhanced by adding microwave technology. Bousbia and research team have investigated the difference in performance between innovative Microwave Hydrodiffusion and Gravity (MHG) and a traditional method like hydrodistillation [28]. In another study, the isolation of essential oil from orange peel was studied using an innovative steam diffusion technique (SDf) called microwave steam diffusion (MSDf). The extraction performance results showed that the isolation period of the essential oils by MSDf technique is within 12 minutes and had similar yield and aromatic profile to those obtained by SDf for 40 minutes [50].

II.2.4. Solvent Extraction

Ordinary solvents like acetone, petroleum ether, hexane, methanol, or ethanol have been implemented by this technique to extract fragile or delicate flower materials which cannot be extracted using heat or steam supplied [41]. Generally, the plant samples are mixed with solvents to be extracted by mildly heating the mixture, and the process is followed by filtration and evaporation of the solvents. The filtrate contains a resin (resinoid), or the mixture of wax, fragrance, and essential oil. Alcohol is combined with the filtrate mixture in order to dissolve the essential oil into it and thereafter distilled at low temperature. During the distillation process, the alcohol absorbs fragrance and is evaporated while the aromatic absolute oil remains in the pot residue. Compared to other methods, this method is more complicated for essential oils extraction, and as a result, time-consuming and more expensive [51].

In another study, authors investigated the antioxidant activity of Ptychotisverticillata by solvent extraction technique for essential oils extraction. It was found that 48% of phenolic compounds are present and contain 44.6% and 3.4% of carvacol and thymol, respectively, as the main compounds [52]. In other studies, the essential oils were separated from Thymus praecox subsp. Skorpilii var. Skorpilii (TPS), and its chemical constituents and antioxidant activity were investigated by mixing the plant extractant with different solvents like ethanol, methanol, and water. These extracts showed significant free-radical scavenging activity with 40.31% of thymol and 13.66% of o-cymene. The results also showed that the extraction process using water as the solvent gives highest phenolics and flavonoids compared to other types of solvents [53].

II.2.5. Supercritical Fluid Extraction

Conventional extraction techniques such as solvent extraction and steam distillation need more time to undergo the extraction process and a large amount of organic solvents are required [54]. Additionally, the disadvantages of these techniques like various volatile components losses, poor efficiency of oils extraction, degradation of unsaturated compounds, and toxic residues from extraction process need to be encountered [55-56].

The supercritical fluid state is mainly depending on two factors which are the fluids critical pressure, Pc and critical temperature, Tc. Fluids with these critical parameters exhibit very interesting properties such as low viscosity, high diffusivity, and density closer to liquids [42].

Carbon dioxide is used as a supercritical solvent for the extraction of essential oils due to its numerous attractive properties:

- easily reach critical point (low critical pressure, Pc : 72.9 atm, and temperature, Tc31.2°C);
- unaggressive for thermo labile molecules of the plant essence;
- chemically inert and toxic;
- nonflammable;
- available in high purity at relatively low cost;
- easily eliminated;
- its polarity similar to pentane which makes it suitable for extraction of lipophilic compounds [57-58].

Generally, the principle of supercritical fluid extraction process involves the use and recycling fluid in repeated steps of compression/ decompression. The supercritical state of CO_2 can be achieved by highly compressing and heating this fluid. Then, it passes through the raw plant material to load volatile matter and plant extracts. The process is followed by decompression steps, where the mixture of CO_2 and plant extracts are routed to two separators where the fluid is gradually decompressed to separate the obtained extracts from the CO_2. The CO_2 is released from second separator and recycled into storage tank, and no solvent residue remains in the final product since CO_2 easily reverts to a gas under normal atmospheric pressure and temperature [59].

Several plant materials have been extracted by using supercritical carbon dioxide extraction method such as rose geranium, Eugenia caryophyllata, clove buds, and marchantia convolute, and their chemical constituents are revealed by some researches [60]. In a study about the comparison between supercritical fluid extractions with hydrodistillation method, by using the supercritical fluid technique, an essential oil was successfully isolated and revealed as advance aromatic oil, with superior performance and pharmacological activities [47]. Other than that, a carrot essential oil obtained by supercritical fluid extraction method was found to give better antibacterial and antifungal properties against Bacillus cereus as compared to the oil obtained by hydrodistillation.

II.2.6. Subcritical Fluid Extraction

The use of water at subcritical state has been reported by many researchers and found that this is a better and powerful alternative of essential oils extraction technique [61].

The definition of subcritical stage of liquid is the time when liquid reaches pressure higher than the critical pressure, Pc and lower than the critical temperature, Tc or *vice-versa*.

The fluids that are used to extract essential oils using this method are water and CO2. The subcritical state of fluid offers several superior characteristics such as lower viscosity, lower density, and enhanced diffusivity between gas and liquids.

This extraction technique is considered the best alternative approach as it enables a fast essential oil isolation process, conducted at a low working temperature. Moreover, it is a costefficient extraction, simple and environmental friendly process [61].

In this process, the required duration of extraction is only 15min compared to 3h required to extract essential oils by using conventional methods. Essential oils with more valuable properties which are a higher amount of oxygenated components with no significant presence of terpenes can be obtained and allow substantial cost saving in terms of both energy and plant materials [62].

Kubatova and co-workers investigated the lactones extraction from a Piper methysticum root by using subcritical water extraction, and this method was compared with Soxhlet extraction with water. The working temperature for subcritical water extraction was at 100°C and 175°C, and the extraction time required to extract the lactones was 20 min and 2h, respectively. Soxhlet extraction method showed a large difference in extraction time compared to subcritical water method, and required 6 hours to extract the oils and produced lower yields by 40% to 60% [62].

II.2.7. Solvent Free Microwave Extraction

The impediments of ordinary extraction techniques, such as solvent and hydrodiffusion, are the losses of several evaporative constituents, poor isolation coherence, and toxic solvent residues at the final product stage. These challenges prompted the consideration of Solvent-Free Microwave Extraction (SFME) for various applications [50]. This technique is an expeditious isolation of essential oils from spices, aromatic herbs, and dry seeds. Several advantages of SFME have been reported by researchers, which can be summarized: to obtain essential oils with high yield and selectivity, shorter extraction time, and environmentally friendly process [50].

SFME involves a combination of two techniques which are heating plant samples using microwave technology followed by dry distillation which operates at an atmospheric pressure in the absence of any solvent. Bayramoglu *et al*. applied SFME method to extract oregano at different microwave power; 622W, 498W, 373W, and 249W, while the essential oil yields were determined depending on each different microwave power used. The results showed maximum yields achieved at 0.054, 0.053, 0.052 and 0.049 mL/g of oregano essential oil at 622W, 498W, 373W, and 249W power levels, respectively. Exception with working at lowest microwave power (249W), all other yields were found to be higher ($p \leq 0.05$) [62].

Compared to hydrodistillation, the yield extracted oregano essential oil was only 0.048 mL/g which about 6% slightly lower than SFME oregano oil highest yield. Later, Ferhat *et al*. presented the comparison of SFME method with traditional methods in terms of extraction periods, yields, impact of the technique used towards the environment, solvent residues content, and antimicrobial activities.

It was demonstrated that microwave extraction offers a shorter isolation period of essential oil (30 min compared to 3 h for hydrodiffusion and 1 h for cold pressing), 0.24% of yields from SFME which is much better than hydrodiffusion and cold pressing with 0.21% and 0.05%, respectively; high energy consumption for performing hydrodiffusion and cold pressing (using mechanical motors) compared to rapid microwave extraction; no water and solvent used in SFME make the extraction process as cleaner features, and high antimicrobial activities of essential oils obtained by SFME technique [63].

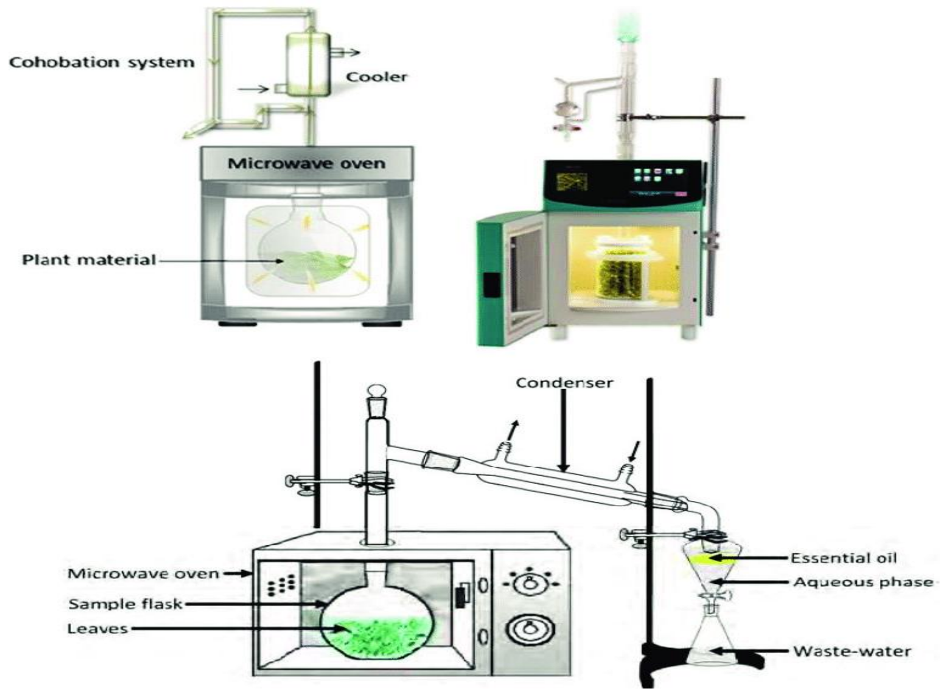

Figure 09: Solvent Free Microwave Extraction process [63].

CHAPTER III

GREEN CHEMISTRY

III.GREEN CHEMISTRY:

III.1.What is Green Chemistry?

The limitations of a command and control system for environmental protection have become more obvious even as the system has become more successful. In industrialized societies with good, well-enforced regulations, most of the easy and inexpensive measures that can be taken to reduce environmental pollution and exposure to harmful chemicals have been implemented. Therefore, small increases in environmental protection now require relatively large investments in money and effort. Is there a better way? There is, indeed. The better way is through the practice of green chemistry.

Green chemistry can be defined as the practice of chemical science and manufacturing in a manner that is sustainable, safe, and non-polluting and that consumes minimum amounts of materials and energy while producing little or no waste material. The practice of green chemistry begins with recognition that the production, processing, use, and eventual disposal of chemical products may cause harm when performed incorrectly. In accomplishing its objectives, green chemistry and green chemical engineering may modify or totally redesign chemical products and processes with the objective of minimizing wastes and the use or generation of particularly dangerous materials. Those who practice green chemistry recognize that they are responsible for any effects on the world that their chemicals or chemical processes may have. Far from being economically regressive and a drag on profits, green chemistry is about increasing profits and promoting innovation while protecting human health and the environment.

To a degree, we are still finding out what green chemistry is. That is because it is a rapidly evolving and developing subdiscipline in the field of chemistry. And it is a very exciting time for those who are practitioners of this developing science. Basically, green chemistry harnesses a vast body of chemical knowledge and applies it to the production, use, and ultimate disposal of chemicals in a way that minimizes consumption of materials, exposure of living organisms, including humans, to toxic substances, and damage to the environment. And it does so in a manner that is economically feasible and cost effective [64].

In one sense, green chemistry is the most efficient possible practice of chemistry and the least costly when all of the costs of the practice of chemistry, including hazards and potential environmental damage are taken into account.

Green chemistry is sustainable chemistry. **There are several important respects in which green chemistry is sustainable:**

- Economic: At a high level of sophistication green chemistry normally costs less in strictly economic terms (to say nothing of environmental costs) than chemistry as it is normally practiced.
- Materials: By efficiently using materials, maximum recycling, and minimum use of virgin raw materials, green chemistry is sustainable with respect to materials.
- Waste: By reducing insofar as possible, or even totally eliminating their production, green chemistry is sustainable with respect to wastes.

III.2.Basic principles of green chemistry

Green chemistry is generally based on the 12 principles proposed by Anastas and Warner Nowadays, these 12 principles of green chemistry are considered the fundaments to contribute to sustainable development. The principles comprise instructions to implement new chemical products, new synthesis, and new processes as illustrated in Figure III.7: [65].

Figure 10: The 12 principles of Green Chemistry [65].

III.2.1. The “better to prevent than to cure” principle

It is beneficial to a priori prevent the generation of waste instead of later on treating and cleaning up waste

III.2.2. The “atom economy” principle

Synthetic production routes have to be planned in a way maximizing the incorporation of all the compounds used in the synthesis into the desired product

III.2.3. The “less precarious chemical syntheses” principle

Wherever feasible, such synthetic methods have to be aspired, which resort to and generate compounds of no or only insignificant noxiousness to the environment and human health

III.2.4. The “designing safer chemicals” principle

Chemicals should be developed in a way affecting their desired functionality, while, at the same time, considerably reducing their toxicity

III.2.5. The “safer solvents and safer auxiliaries” principle

Expenditure of auxiliary substances, such as solvents, separation agents, and others, should be avoided wherever possible; if not possible, harmless auxiliaries should be used

III.2.6. The “design for energy efficiency” principle

The environmental and economic impact of energy demands for chemical processes should be analyzed in terms of followed by optimizing the required energy input. Wherever practicable, chemical synthesis should be carried out under mild process conditions, hence, at ambient temperature and pressure

III.2.7. The “renewable feedstocks” principle

Whenever feasible in technological and economic terms, synthetic processes should resort to such raw materials and feedstocks, which are renewable rather than limited

III.2.8. The “derivative reduction” principle

Redundant derivatization, e.g., protection/deprotection, the use of blocking groups, or temporary modification of physical/chemical processes, requires additional reagents and often contributes to additional waste generation. Therefore, wherever possible, they should be avoided or reduced to a minimum

III.2.9. The “catalysis” principle

Generally, catalytic reagents are intrinsically superior to stoichiometric reagents; these catalysts should be as selective as possible

III.2.10. The "degradation" principle

Chemical products have to be designed in such a way that, at the end of their life span, they do not resist in the biosphere, but disintegrate into nontoxic degradation products

III.2.11. The "real-time analysis for pollution prevention" principle

Advanced analytical methods have to be developed, which permit the real-time, in-line process monitoring and control well before hazardous substances are generated

III.2.12. The "accident prevention by inherently safer chemistry" principle

Compounds and the compound's formula applied in a chemical process should be chosen in a way minimizing the risk of chemical accidents, encompassing the release of chemicals, detonations, or fire formation [65].

CHAPTER IV

GERANIUM ROSE

IV.GERANIUM ROSE:

IV.1.What is Geranium Rose (Pelargonium graveolens)?

In the 18th century, pelargonium, native to South Africa, also called geranium, was in high demand. He enjoyed, in the parks of the princely houses of England, France and later also of Italy and from Germany, of almost royal status. But in the 20th century, preference was given to large plants. flowers, and from then on geraniums began to lead a shadowy existence. The main growing areas for extracting essential oil from geranium nowadays are parts of France, Italy, India and the former Soviet Union as well as an island in the Caribbean, Réunion, where the most famous essential oil of geranium comes from, "Bourbon oil."

The geranium rose is an aromatic plant belonging to to the Geraniaceae family, it is native to Africa southern and cultivated in many Mediterranean regions and subtropical [66-67]. In Algeria, this plant annual and spontaneous is widespread. Although the essential oil of this pelargonium has anti-infectious, anti-inflammatory, spasmolytic properties, hemostatic and healing [68-69], little work were dedicated to him in Algeria. From the perspective of using the essential oil of Pelargonium graveolens in the food industry, in practice medical and cosmetics, we have offered to determine its chemical composition and to search for in vitro its activity on one hundred and thirty bacterial strains community infections, including strains multiresistant.

IV.2.Etymology

Pelargonium comes from the Greek πελαργός pelargos which means stork. Another name for pelargoniums is stork's-bills due to the shape of their fruit. The specific epithet graveolens refers to the strong-smelling leaves [70].

IV.3.Color and Fragrance

Geraniums rose have small lavender or pink flowers that dot a mound of grayish, silvery green foliage, making them appear much different that the typical ‘cranesbill’ rounded-head varieties you may envision when you think of geraniums. The leaves are the prized portion of the plant, because this is where the scent is produced. This foliage is often used in perfumed oils, and can even be added to culinary creations including beverages and desserts. From the popular ‘Attar of Rose’ and ‘Old Fashioned Rose’ varieties, which have a strong rose scent, to those that have a lemon and rose blend, such as ‘Ice Crystal Rose’ and ‘Candy Dancer,’ the fragrances are a delightful addition to any garden [70].

IV.4.Description

Pelargonium graveolens is an erect, multi-branched shrub, that grows up to 1.5 m and has a spread of 1 m. The leaves are deeply incised, velvety and soft to the touch (due to glandular hairs). The flowers vary from pale pink to almost white and the plant flowers from August to January. The leaves may be strongly rose-scented, although the leaf shape and scent vary. Some plants are very strongly scented and others have little or no scent. Some leaves are deeply incised and others less so [5], being slightly lobed like P. capitatum.

Figure 11: Experimental Geranium Rose (Pelargonium graveolens).

IV.5.Cultivars and hybrids

Many plants are cultivated under the species name "Pelargonium graveolens" but differ from wild specimens as they are of hybrid origin [1] (probably a cross between P. graveolens, P. capitatum and/or P. radens). There are many cultivars and they have a wide variety of scents, including rose, citrus, mint and cinnamon as well as various fruits. Cultivars and hybrids include:

- P. 'Graveolens' (or Pelargonium graveolens hort.) - A rose-scented cultivar; possibly a hybrid between P. graveolens and P. radens or P. capitatum. This cultivar is often incorrectly labeled as Pelargonium graveolens (the species). The main difference between the species and this cultivar is the dissection of the leaf. The species had about 5 lobes but the cultivar has about 10.

- P. 'Citrosum' - A lemony, citronella-scented cultivar, similar to P. 'Graveolens'. It is meant to repel mosquitos and rumour has it that it was made by genetically bonding genes from the citronella grass but this is highly unlikely.
- P. 'Cinnamon Rose' - A cinnamon-scented cultivar.
- P. 'Dr Westerlund' - A lemony rose-scented cultivar, similar to P. 'Graveolens'.
- P. 'Graveolens Bontrosai' - A genetically challenged form; the leaves are smaller and curl back on themselves and the flowers often do not open fully. Known as P. 'Colocho' in the US.
- P. 'Grey Lady Plymouth' - A lemony rose-scented cultivar similar to P. 'Lady Plymouth'. The leaves are grey–green in colour.
- P. 'Lady Plymouth' - A minty lemony rose-scented cultivar. A very popular variety with a definite mint scent. Possibly a P. radens hybrid.
- P. 'Lara Starshine' - A lemony rose-scented cultivar, similar to P. 'Graveolens' but with more lemony scented leaves and reddish pink flowers. Bred by Australian plantsman Cliff Blackman.
- P. 'Lucaeflora' - A rose-scented variety, much more similar to the species that most other cultivars and varieties.
- P. × melissinum - The lemon balm pelargonium (lemon balm - Melissa officinalis). This is a hybrid between P. crispum and P. graveolens.
- P. 'Mint Rose' - A minty rose-scented cultivar similar to P. 'Lady Plymouth' but without the variegation of the leaves and lemony undertones.
- P. 'Secret Love' - An unusual eucalyptus-scented cultivar with pale pink flowers.
- P. 'Van Leeni' - A lemony rose-scented cultivar, similar to P. 'Graveolens' and P. 'Dr Westerlund'.

Others known; Camphor Rose, Capri, Granelous and Little Gem [70].

IV.6.Use in natural medicine

Only a few of the msore than 230 kinds / species of this abundant genus play a role as medicinal plants. These include Pelargonium reniforme, P. sidoides, P. sad and well heard Pelargonium graveolens. Internally, the plant is used for nausea, inflammation of the tonsils and circulatory weakness. The essential oil of geranium contained in face creams has a superfatting action on the skin, inhibits yeast infections and also heals eczema. Geranium essential oil is also used as a secondary treatment for depression and hormonal disorders. Essential oil extracted from P. graveolens is an important ingredient in skin care products and perfumes and is also used successfully in aromatherapy. Its leaves, wrapped in a scent bag, can also serve as a fragrance diffuser [71].

IV.7.Use in the kitchen

It is amazing how many ways we can use leaves and flowers generally delicate plant in the kitchen. Flowers for decorating a salad not only put a colorful note, but also tasty accents. Finely chopped leaves can serve to season sauces, custards and to make jams and syrup. But the cookies, cakes and breads can also be flavored with the different fragrances of pelargoniums. The fragrant leaves and slender flowers of the plant are of course also perfect for decorate a festive table, and a potpourri of flowers and bewitching aromas is a wonderful gift very personal [71].

PRACTICE PART

I.MATERIALS AND METHODS

I.1.Study goal:

Our work was carried out in the laboratory of L.G.P. at the University of Kasdi Merbah Ouargla. The objective of our work is Valorization of the essential oils of the geranium rose plant and used as biopesticide. The purpose of this phase is to extract the essential oil from geranium rose. The hydro-distillation extraction method is used.

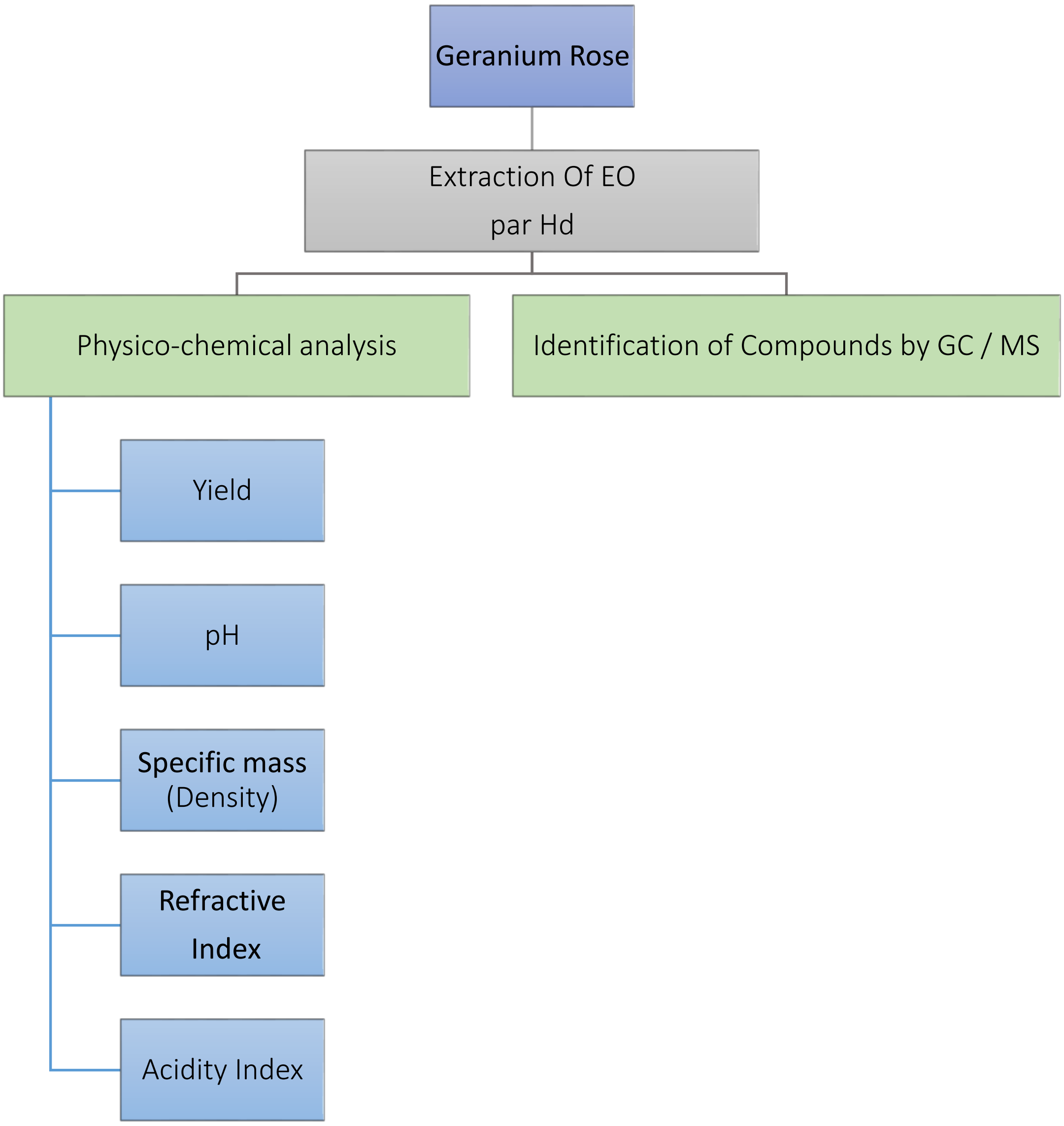

Figure 12: The general diagram of the experimental procedure.

I.2.Vegetal material:

The plant material that is used during the realization of this work consists of geranium rose leaves species harvested in April 2021 in the region of Toggort (Southeast Algeria) and then dried in the shade at an ambient temperature during 05 days.

Figure 13: Experimental Geranium Rose (Pelargonium graveolens).

I.3.Extraction of EO:

The essential oils of the plant studied were extracted by hydrodistillation using an apparatus of the Clevenger (Figure.V.3). This method involves directly immersing the plant material to be treated in distilled water which is brought to the boil. The volatile constituents are then entrained by the water vapor and after condensation of the distillate, are separated by decantation.

Figure 14: Experimental setup of hydrodistillation.

- **Operating mode:**

One hundred grams (100g) of the Geranium rose plant material is placed in a 2000ml vacuum flask supplemented with 900ml of water. The whole is brought to the boil, after the appearance of the first drop of distillate at the outlet of the steam condenser; the essential oil is then carried away by the water vapor. It is then condensed by passing through a condenser, which is fixed by a vertical support which facilitates the flow of the distillate. The time for this extraction is approximately three hours. The distillate is left to stand for a few minutes, which results in the appearance of two phases, one is organic (essential oil) loaded with volatile species contained in the plant and the other is aqueous (or the hydrosol) having higher density. The volume of essential oil obtained is collected in a test tube. The mass of EO is noted for the calculation of efficiency.

I.3.1. Oil conservation:

The storage of essential oil requires certain essential precautions [72]. This is why we dehydrated the essential oil with chlorure calcium ($CaCl_2$) [73] and stored it at a temperature close to 4 ° C, in tightly closed tubes, covered with aluminum foil to preserve it from the air and light.

I.3.2 Determination of the chemical composition of the EO of Geranium rose by GC / MS

The analysis of T. capitatus EO was performed at the L.G.P (Process Engineering laboratory) in Kasdi Merbah Ouargla University. The gas chromatograph adopted is a Bruker SCION 436 GC, coupled to a mass spectrometer quadrupole ionization voltage of 70 ev. The column that is used is an HP-5MS; 5% Phenyl Methyl Siloxane with a length of 30 m and an internal diameter of 0.25 mm. The wire thickness being 0.25 mm.

The operating conditions are:

- The temperature of the injector (split mode 1:50): 250 °C

- Temperature programming: from 50 °C to 280 °C at a rate of 5 °C/min.

- The vector gas used is helium with a flow rate of 1.2 ml/min.

The temperatures of the quadrupole source are fixed, respectively, at 250 °C and 280 °C. Linear retention indices (RI) for all compounds were determined using n-alkanes as standards.

Identification of individual compounds was performed by matching their mass spectral fragmentation patterns with corresponding data available (Wiley 275 library (6th edition)).

I.3.3.Evaluation of some physicochemical indices of EOs

***I.3.3.1.Yield*:**

According to the AFNOR standard (2000), the yield of essential oil is defined as the ratio between the weight of essential oil obtained after extraction and the weight of the plant material used . It is given by the following formula [74]:

Percent yield of oil (w/w) = (W_{EO} / W_S)*100

Percent yield of oil (w/w) (%)

W_{EO}: weight of oil in gram

W_S: weight of the sample in gram

I.3.3.2.Determination of the Acid Number:

The AN Acid Number is the number of milligrams (mg) of potash needed to neutralize the free acids in 1 gram (g) of essential oil according to the reaction [75]:

RCOOH + KOH $\longrightarrow$ **RCOOK + H2O** (reaction at room temperature)

- **Operating mode:**

1 g of EO is placed in a beaker. Add 5 ml of ethnol and a maximum of 3 drops of indicator, or Phenolphthalein solution. Titrate the liquid with the KOH solution (C (KOH) = 0.1 mol / l), contained in the burette for a few seconds. After the color has turned to pink, the titration is stopped. Note the volume of KOH solution used.

The Acid index is expressed by the formula:

$$AN = (56,11 \times N \times V) / W$$

56.11 = Molecular weight of KOH

N: Normality of KOH.

V: Volume in ml of the ethanoic solution of KOH used for the titration.

W: weight in grams of the essential oil.

Figure 15: Experimental setup of the Acid Number.

I.3.3.3. Determination of Specific mass (Density ρ):

Density varies from oil to oil, which is lower than the density of water, so the oil floats above water.

- **Operating mode :**

Determination of specific mass of oils, a clean 50 ml specific mass needle was weighted (W_0). Then the needle was filled to the brim with water and stopper was inserted.

Extra water spilled out. The water on the stopper and needle were carefully wiped off and reweighed (W_1). Same process was repeated, but using oil samples instead of water and weighted again (W_2). The specific gravity of the all oil samples were calculated using the following formula [76].

Specific mass of test sample $\rho = (W_2 - W_0 / W_1 - W_0)$

W_0: Weight of empty specific mass needle

W_1: Weight of water + specific mass needle

W_2: Weight of test sample + specific mass needle.

Figure 16: Experimental determination of specific mass of oil.

I.3.3.4.Refractive index:

The refractive index (change in direction of light when passing from one medium to another) of an essential oil is the ratio between the sine of the angle of incidence and the sine of the angle of refraction of the light ray of determined wavelength, passing from air to essential oil maintained at a constant temperature [62].

- **Operating mode:**

The secondary prism is opened and then 2 or 3 drops of the liquid sample are placed on the central part of the main prism. Then the secondary prism is gently closed. The sample spreads between the main prism and the secondary prism in a thin film. The temperature is left to wait for the measurement to be stable. The measurement value for a liquid sample being modified according to the temperature change, read the temperature indicator to know the actual measurement degree, and attach it without fail to the measured value.

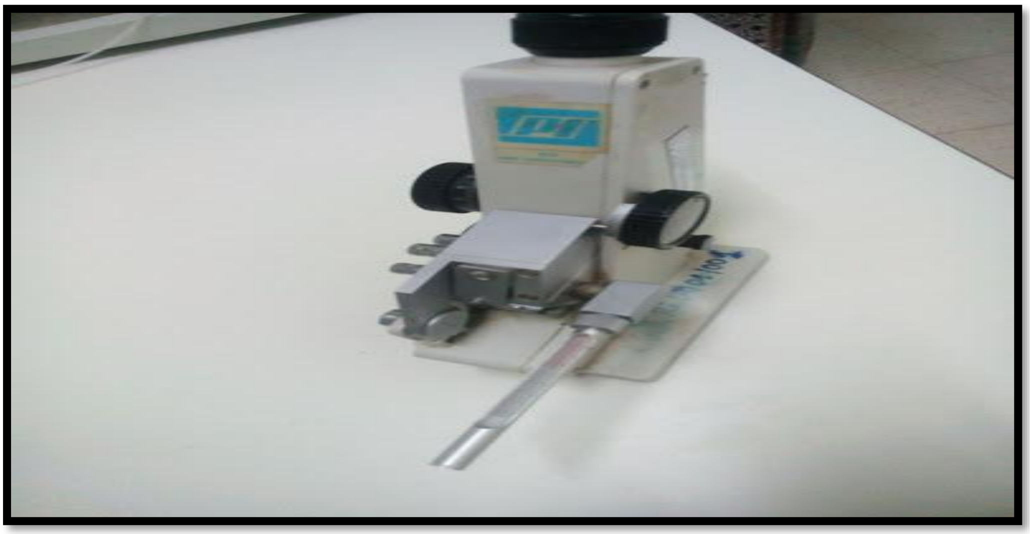

Figure 17: Experimental setup of Refractometer.

I.3.3.5.pH measurement:

pH: the abbreviation for hydrogen potential measures the chemical activity of hydrogen ions (H^+) in solution

• Measured by a pH meter

• Of paper pH.

We put a few drops of EO on a piece of pH paper, after changing the color of the paper we compare it with a range of colors that vary according to the pH

Figure 18: Experimental setup of pH paper.

II.RESULTS AND DISCUSSIONS

II.Results and Discussions:

In this chapter, the extraction of essential oils from geranium rose, organoleptic property and Physico-Chemical analysis were discussed.

II.1.Organoleptic properties of essential oils:

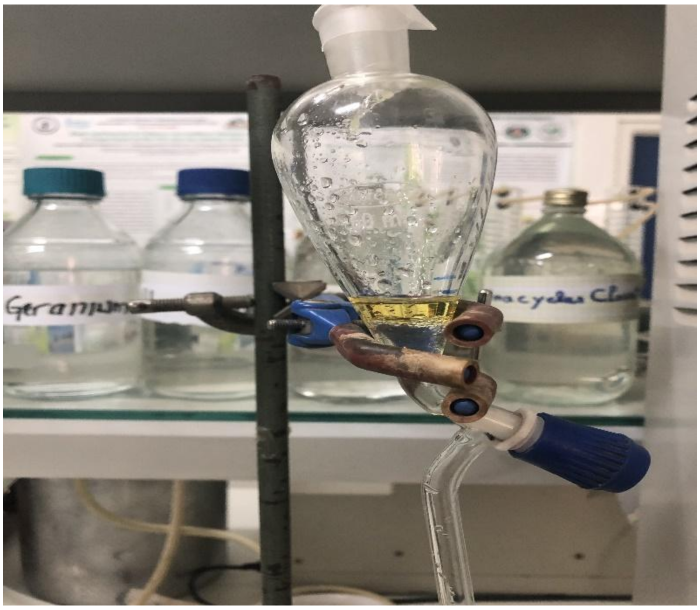

Figure 19: Experimental setup the EO of geranium rose extracted by HD.

The information is summarized in the following table:

Geranium Rose	
Aspect	Clear liquid
Color	Dark yellow
Odour	Strong
Taste	Bitter

Table 02: Organoleptic characteristics of the EO of geranium rose.

II.2. Physico-chemical analyzes:

II.2.1. Extraction yield:

- **Results :**

The information is summarized in the following table:

EO	**Yield %**	**AFNOR [74].**
Geranium Rose	0.12	0.1-0.15%

Table 03: The yield of essential oils.

- **Discussion :**

The essential oil yield of geranium rose a maximum of 0.12%. The yield of the essential oil is clearly between that found by AFNOR (0.1-0.15%) [74].

It should be noted that the yield and the chemical composition of EOs depends on several factors namely the species, natural factors such as climate, soil quality, ..etc. the harvest environment, harvest time, cultural practices.

II.2.2. physico-chemical properties of EO:

- **Results :**

Properties	**Practical value**	**AFNOR [74].**
The Acid Number	3.92	2 - 4
pH	5	4 - 6
Specific mass (density)	0.91	0.905-0.921
The refractive index	1.4771	1.461-1.470

Table 04: Physicochemical property of essential oils.

- **Discussion :**

The pH obtained indicates that our extracted oil is acidic.

The Specific mass (density) of our essential oil is 0.91. This physical property is used in the classification of essential oils; these data are insufficient for the classification of oils. This parameter is related to the chemical composition of this oil which is affected by a large number of factors such as phenotype, time of harvest, type of soil, storage, process and extraction conditions [77].

The measured refractive index is : 1.4771. This index depends on the chemical composition which increases as a function of the lengths of the acid chains, their degrees of establishment and the temperature, it varies mainly with the content of monoterpenes and oxygenated derivatives. A high content of monoterpenes will give a high index [78].

A low refractive index of EO indicates its low refraction of light, which could favor its use in cosmetic products. The results indicate that the physico-chemical parameters of the samples analyzed are found within the reference ranges established by the standards.

An acid number of less than 2 is proof of good preservation of the gasoline (low amount of free acids) to characterize essential oils our results more than 2 it is less preservation.

The determination of the physicochemical properties is a necessary step but remains not sufficient to characterize the EO. It will therefore be essential to supplement it with chromatographic analyzes (GC and GC / MS).

II.2.3. Chromatographic analyzes:

The characterization of essential oils was carried out by GC / SM.

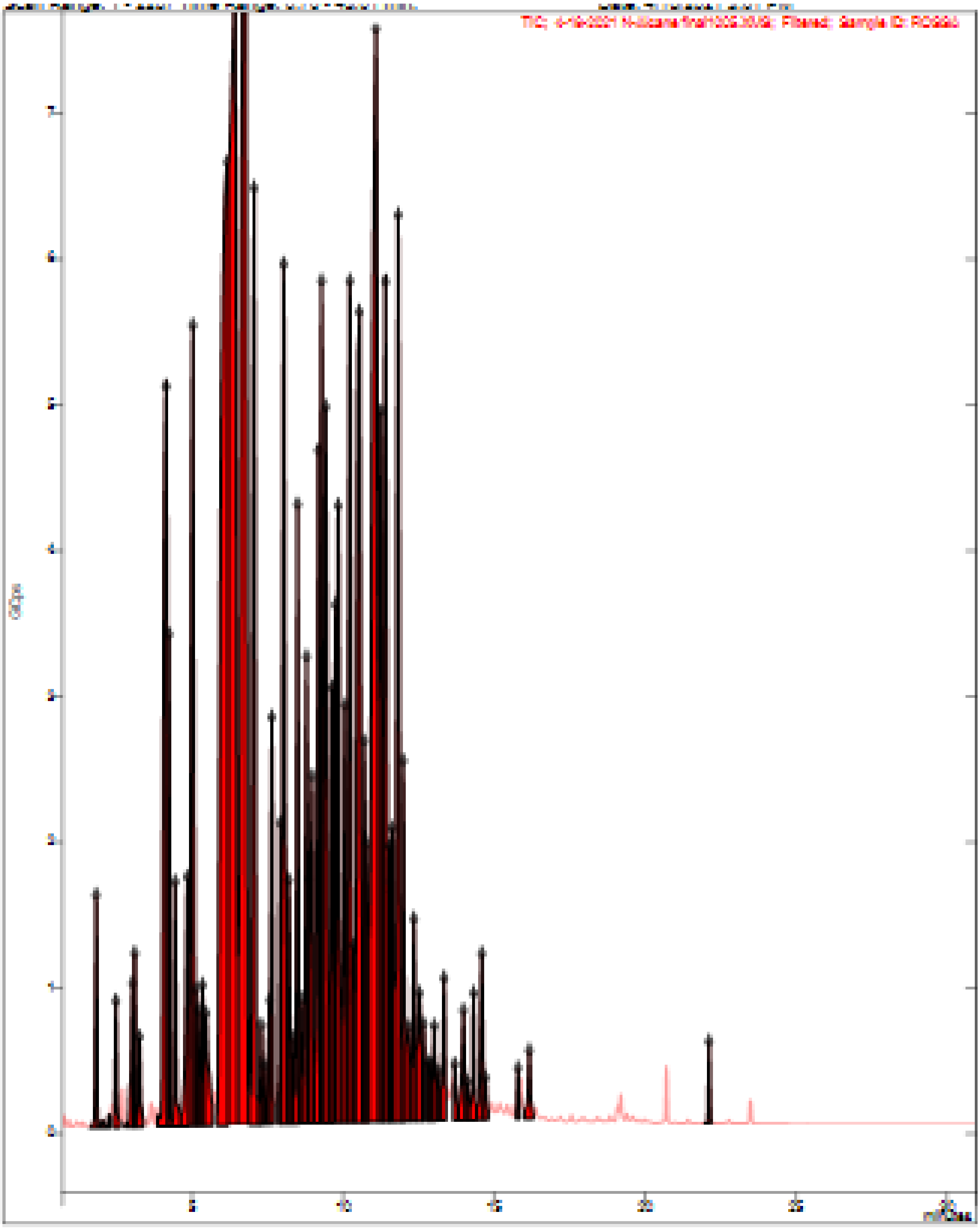

Figure 20: Chromatographic profile of essential oil of Geranium rose.

N°	Compound Name	RT	Area %
01	Linalool	4.140	3.30
02	(2S,4R)-4-Methyl-2-(2-methylprop-1-en-	4.226	1.46
03	l-Menthone	5.029	3.44
04	***Citronellol***	***6.160***	***18.44***
05	**trans-Farnesol**	**6.727**	**7.05**
06	2,6-Octadien-1-ol, 3,7-dimethyl-, format	7.058	3.21
07	6-Octen-1-ol, 3,7-dimethyl-, acetate	7.642	2.04
08	Caryophyllene	8.499	1.34
9	(1R,9R,E)-4,11,11-Trimethyl-8-methylen	8.988	0.71
10	Lavandulyl propionate	9.189	1.91
11	(1R,2S,6S,7S,8S)-8-Isopropyl-1-methyl-3	9.298	2.42
12	1H-Cycloprop[e]azulene, 1a,2,3,5,6,7,7a,	9.422	1.94
13	Butanoic acid, 3,7-dimethyl-2,6-octadien	9.625	1.03
14	1-Isopropyl-4,7-dimethyl-1,2,3,5,6,8a-he	9.741	1.75
15	Citronellyl butyrate	9.838	1.55
16	(3R,5aS,9aR)-2,2,5a,9-Tetramethyl-3,4,5	10.065	1.14
17	Butanoic acid, 3,7-dimethyl-2,6-octadien	10.226	2.55
18	2-Phenylethyl tiglate	10.556	1.88
19	***2-((2S,4aR)-4a,8-Dimethyl-1,2,3,4,4a,5,6***	***11.094***	***7.00***
20	Agarospirol	11.171	0.98
21	.tau.-Cadinol	11.250	2.47
22	(1aR,3aS,7S,7aS,7bR)-1,1,3a,7-Tetrame	11.406	3.20
23	1-Heptatriacotanol	11.539	0.72
24	3,7-Dimethyloct-6-en-1-yl	11.620	0.77
25	Geranyl tiglate	11.835	2.99
26	Hexanoic acid, 3,7-dimethyl-2,6-octadien	11.986	0.93
	Totale %		76.22%

Table 05: Chemical composition of Geranium rose.

The results of the gas chromatography analysis -Mass spectrometry of the chemical composition of EO are presented in the table in which the identified compounds are listed. In total 27 compounds have been identified which correspond to at a percentage of 76.22% relative to all of the isolated constituents. Citronellol appears as the major constituent of EO (18.44%), followed by trans-Farnesol (7.05%).

GENERAL CONCLUSION

GENERAL CONCLUSION

Essential oils are highly concentrated volatile substances extracted from various parts of certain plant species for example (stem, leaves, flowers, root, wood) each with specific therapeutic and energetic effects.

Green chemistry can be defined as the practice of chemical science and manufacturing in a manner that is sustainable, safe, and non-polluting and that consumes minimum amounts of materials and energy while producing little or no waste material.

The Geranium rose is an aromatic plant belonging to to the Geraniaceae family. In Algeria, this plant annual and spontaneous is widespread. Although the essential oil of this pelargonium has anti-infectious, anti-inflammatory, spasmolytic properties, hemostatic and healing, little work were dedicated to him in Algeria. From the perspective of using the essential oil of Pelargonium graveolens in the food industry, in practice medical and cosmetics.

The aim of this research is to study and evaluate the essential oils of the aromatic plant Geranum rose and used as biopstcide using the hydrodistillation extraction method. The physical and chemical characteristics of these oils are also been studied. The study found that the plant contains the compounds in the following proportions:

Citronellol 18.44 % and trans-Farnesol 7.05 %.

Citronellol is used in perfumes and insect repellents and as a mite attractant. Citronellol is a good mosquito repellent at short distances, but protection greatly lessens when the subject is slightly further from the source and Citronellol is used as a raw material for the production of rose oxide.

Oils are separated and identified with GC / MS in addition, these oils are important and have great applications in different fields, pharmaceutical, medical and cosmetic, also used in biological control as a biopesticide.

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ANNEXES

N°	Compound Name	RT	Area %	CAS#
01	Bicyclo[3.1.1]hept-2-ene, 3,6,6-trimethy	1.823	0.50	4889-83-2
02	.beta.-Pinene	2.338	0.04	127-91-3
03	.beta.-Myrcene	2.475	0.24	123-35-3
04	.alpha.-Phellandrene	2.694	0.08	99-83-2
05	p-Cymene	2.943	0.09	99-87-6
06	D-Limonene	3.014	0.37	5989-27-5
07	Bicyclo[3.1.1]hept-2-ene, 3,6,6-trimethy	3.104	0.36	4889-83-2
08	.beta.-Ocimene	3.251	0.15	13877-91-3
09	8-hydroxymenthol	3.629	0.07	None
10	Cyclohexene, 3-methyl-6-(1-methylethyli	3.804	0.03	586-63-0
11	Linalool	4.140	3.30	78-70-6
12	(2S,4R)-4-Methyl-2-(2-methylprop-1-en-1	4.226	0.99	3033-23-6
13	(2R,4R)-4-Methyl-2-(2-methylprop-1-en-1	4.432	0.47	5258-11-7
14	l-Menthone	4.842	0.81	14073-97-3
15	l-Menthone	5.029	2.63	14073-97-3
16	endo-Borneol	5.107	0.31	507-70-0
17	Cyclohexanol, 5-methyl-2-(1-methylethyl)	5.202	0.32	15356-70-4
18	Cyclohexanol, 5-methyl-2-(1-methylethyl)	5.339	0.33	15356-70-4
19	.alpha.-Terpineol	5.454	0.46	98-55-5
20	Isogeranial	5.740	0.09	None
21	Citronellol	6.160	8.69	106-22-9
22	6-Octen-1-ol, 3,7-dimethyl-, (R)-	6.429	9.75	1117-61-9
23	trans-Farnesol	6.727	7.05	106-28-5
24	6-Octen-1-ol, 3,7-dimethyl-, formate	6.780	2.19	105-85-1
25	(1S,3S,4S,5R)-1-Isopropyl-4-methylbicyc	6.844	0.05	7712-79-0
26	2,6-Octadien-1-ol, 3,7-dimethyl-, format	7.058	3.21	2142-94-1
27	Santolina triene	7.157	0.06	2153-66-4
28	cis-3-Hexenyl cis-3-hexenoate	7.282	0.23	61444-38-0
29	Elemene isomer	7.378	0.12	None
30	2-Cyclohexen-1-one, 3-methyl-6-(1-meth	7.510	0.13	491-09-8
31	.alpha.-Cubebene	7.561	0.21	17699-14-8
32	6-Octen-1-ol, 3,7-dimethyl-, acetate	7.642	0.88	150-84-5
33	2,6-Octadien-1-ol, 3,7-dimethyl-, acetat	7.751	0.04	141-12-8
34	.alpha.-ylangene	7.830	0.07	None
35	.alfa.-Copaene	7.926	0.60	None
36	(R)-lavandulyl acetate	8.039	2.34	None
37	Cyclohexane, 1-ethenyl-1-methyl-2,4-bis(	8.118	0.55	33880-83-0
38	Oxalic acid, isohexyl 2-phenylethyl este	8.193	0.64	None
39	1H-Cycloprop[e]azulene, 1a,2,3,4,4a,5,6,	8.313	0.17	489-40-7
40	Citronellol epoxide (R or S)	8.399	0.03	None
41	Caryophyllene	8.499	1.34	87-44-5
42	.beta.-copaene	8.601	0.27	None
43	trans-.alpha.-Bergamotene	8.657	0.10	13474-59-4
44	6-Octen-1-ol, 3,7-dimethyl-, propanoate	8.788	1.12	141-14-0
45	(1R,3aS,8aS)-7-Isopropyl-1,4-dimethyl-1,	8.848	0.51	36577-33-0

46	Humulene	8.935	0.47	6753-98-6
47	(1R,9R,E)-4,11,11-Trimethyl-8-methylen	8.988	0.71	68832-35-9
48	Germacrene D	9.084	0.11	23986-74-5
49	Lavandulyl propionate	9.189	1.91	59550-34-4
50	(1R,2S,6S,7S,8S)-8-Isopropyl-1-methyl-3	9.298	2.42	18252-44-3
51	1H-Cycloprop[e]azulene, 1a,2,3,5,6,7,7a,	9.422	1.94	21747-46-6
52	(1S,2E,6E,10R)-3,7,11,11-Tetramethylbi	9.471	0.61	24703-35-3
53	2H-3,9a-Methano-1-benzoxepin, octahyd	9.558	0.33	5956-09-2
54	Butanoic acid, 3,7-dimethyl-2,6-octadien	9.625	1.03	106-29-6
55	1-Isopropyl-4,7-dimethyl-1,2,3,5,6,8a-he	9.741	1.75	16729-01-4
56	Citronellyl butyrate	9.838	1.55	141-16-2
57	Phenyl ethyl angelate,2-	9.934	0.15	None
58	.alpha.-Calacorene	9.991	0.17	21391-99-1
59	(3R,5aS,9aR)-2,2,5a,9-Tetramethyl-3,4,5	10.065	1.14	5956-12-7
60	Butanoic acid, 3,7-dimethyl-2,6-octadien	10.226	2.55	106-29-6
61	(1S,3aR,4R,8R,8aS)-1-Isopropyl-3a-met	10.292	0.14	88395-47-5
62	3,7-Dimethyloct-6-en-1-yl heptanoate	10.335	0.31	72934-17-9
63	No Match	10.388	0.25	None
64	Phenyl ethyl angelate, 2-	10.504	2.12	None
65	2-Phenylethyl tiglate	10.556	1.88	55719-85-2
66	Neryl (S)-2-methylbutanoate	10.685	0.97	None
67	Ledol	10.756	0.50	577-27-5
68	(1R,3E,7E,11R)-1,5,5,8-Tetramethyl-12-	10.798	0.43	19888-34-7
69	2-((2S,4aR)-4a,8-Dimethyl-1,2,3,4,4a,5,6	11.094	7.00	117066-77-0
70	Agarospirol	11.171	0.98	1460-73-7
71	.tau.-Cadinol	11.250	2.47	5937-11-1
72	(1aR,3aS,7S,7aS,7bR)-1,1,3a,7-Tetrame	11.406	3.20	527-90-2
73	1-Heptatriacotanol	11.539	0.72	105794-58-9
74	3,7-Dimethyloct-6-en-1-yl tetradecanoate	11.620	0.77	3681-72-9
75	(1R,7S,E)-7-Isopropyl-4,10-dimethylenec	11.714	0.17	81968-62-9
76	Geranyl tiglate	11.835	2.99	7785-33-3
77	No Match	11.916	0.12	None
78	Hexanoic acid, 3,7-dimethyl-2,6-octadien	11.986	0.93	68310-59-8
79	Methyl 5,11,14-eicosatrienoate	12.100	0.13	None
80	1,6,10,14-Hexadecatetraen-3-ol, 3,7,11,1	12.157	0.15	1113-21-9
81	Cyclohexene, 1,5,5-trimethyl-6-acetylmet	12.192	0.13	211563-96-1
82	No Match	12.284	0.11	None
83	Hexanoic acid, 3,7-dimethyl-2,6-octadien	12.341	0.48	68310-59-8
84	Bicyclo[4.1.0]heptan-2-one, 3,4,4-trimet	12.445	0.13	102146-81-6
85	Tricyclo[20.8.0.0(7,16)]triacontane, 1(2	12.506	0.19	None
86	Cholesterol 3-O-[[2-acetoxy]ethyl]-	12.544	0.19	30656-76-9
87	((4aS,8S,8aR)-8-Isopropyl-5-methyl-3,4,	12.599	0.10	135118-51-3
88	1-Hexadecanol	12.669	0.19	36653-82-4
89	Bicyclo[4.1.0]heptane, 3,7,7-trimethyl-	12.741	0.11	554-59-6
90	((8R,8aS)-8-Isopropyl-5-methyl-3,4,6,7,8	12.818	0.11	135118-52-4
91	Ylangenal	12.898	0.10	41610-68-8

92	3,7-Dimethyloct-6-en-1-yl heptanoate	13.010	0.21	72934-17-9
93	6,11-Dimethyl-2,6,10-dodecatrien-1-ol	13.062	0.06	None
94	2-Butenal, 2-methyl-4-(2,6,6-trimethyl-1	13.156	0.10	3155-71-3
95	Cetene	13.231	0.05	629-73-2
96	Heptanoic acid, 3,7-dimethyl-2,6-octadie	13.344	0.33	73019-15-5
97	Phthalic acid, 8-bromoctyl isobutyl este	13.454	0.05	None
98	Pentadecanoic acid	13.545	0.04	1002-84-2
99	3,7-Dimethyloct-6-en-1-yl heptanoate	13.620	0.07	72934-17-9
100	1-Hexadecanol	13.708	0.13	36653-82-4
101	2-Butyloxycarbonyloxy-1,1,10-trimethyl-6	13.897	0.04	108533-24-0
102	Farnesol isomer a	13.996	0.28	None
103	(4aS,7R)-7-(2-Hydroxypropan-2-yl)-1,4a-	14.113	0.08	473-10-9
104	9-Nonadecene	14.244	0.03	31035-07-1
105	Geranyl caprylate	14.321	0.22	51532-26-4
106	Pentadecanoic acid	14.596	0.45	1002-84-2
107	3-Eicosene, (E)-	14.709	0.09	74685-33-9
108	(9Z,12Z,15Z)-3,7-Dimethyloct-6-en-1-yl o	14.946	0.02	None
109	Farnesol isomer a	15.169	0.02	None
110	3-Eicosene, (E)-	15.209	0.02	74685-33-9
111	Cyclopentane, 1-isobutylidene-3-methyl-	15.685	0.03	None
112	5-(7a-Isopropenyl-4,5-dimethyl-octahydro	15.785	0.11	None
113	Phytol	15.879	0.08	150-86-7
114	3,7-Dimethyloct-6-en-1-yl nonanoate	15.968	0.02	72934-18-0
115	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-	16.168	0.15	463-40-1
116	2-methyloctacosane	19.191	0.05	None
117	2-methyloctacosane	20.706	0.12	None
118	2-methyloctacosane	22.117	0.17	None
119	2-methyloctacosane	23.491	0.05	None
		Totale	**99,97%**	