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Antibacterial, antifungal activity and chemical composition study of essential oil of *Mentha pepirita* from the south Algerian

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ABSTRACT

Natural substances from plants have multiple interests put to use in the industry. Essential oils are a very interesting group of these metabolites that feature different properties that make them interesting. We did this study to obtain an extract essential oil from the plant Mentha pepirita South region Algerian cultivated to the Municipality of Ouargla. the Mint, widespread in Algeria, of the family Lamiaceae is a native herb of several therapeutic properties (antiseptic, anti-neuralgic, analgesic ...). The extraction of its aromatic fraction offers new perspectives in aromatherapy by hydrodistillation method by a Clevenger-type apparatus. The extract obtained was the subject of Antibacterial and antifungal activity and a chemical study includes determining the chemical essential oil composition by GC-MS which showed the product exsistance which might present different interests. The Antibacterial and antifungal activity of oils was tested using the direct contact method against the growth of different Fusarium. The results showed great potential of natural antimicrobial activity against strains tested. The analysis of this oil by GC-MS, lets notice the presence of several components which (23.3%), Followed by 7-Oxabicyclo[4,1,0]heptane,1-methyl-4-(methylethenyl)- (14,6%), Cis-(-)-1,2-Epoxy-p-menth-8-ene (5,72%), and Bicyclo[2.2.1] heptane-2,5-diol,1,7,7-trimethyl-,(2-endo,5-exo)- (4,04%) are the major components.

Keywords: Mints, Essential oils, chemical composition, Antibacterial and antifungal activity and GC-MS analysis.

INTRODUCTION

For thousands of yearshumanity has used various plants found in its environment, to treat and cure all kinds of diseases, these plants represent a huge reservoir of potential compounds attributed to secondary metabolites that have the advantage of being a great diversity of chemical structure and they possess a very wide range of biological activities. However the assessment of these activities remains a very interesting task that may be the interest of many studies [1].

Only at the end of the 18th century that drugs are part of a scientific logic. Initially, it was discovered that plants contained several components, the first pure medicinal substance derived from the plants was morphine, which was extracted from the poppy at the beginning of the 19th century [2].

Currently, aromatic plants have a major advantage due to the gradual discovery of the applications of essential oils in their health care and their uses in other areas of economic interest. Their many uses do they know demand increasingly strong global markets.

With the isolation of the first active ingredients with advances in chemistry to 18th and 19th centuries, the history of the therapeutic role of plants is as long as the history of humanity itself [3]. Currently, about 25% of modern medicines are developed from plants [4,5].

At present, approximately 25-30% of all the drugs available for the treatment of diseases are derived from natural products (plants, animals, bacteria and fungi).

Today, many work, conducted in the field of ethnopharmacology, show that the plants used in traditional medicine and have been tested, are often effective and almost all free of toxic plants.

The enhancement of natural resources is a concern that is becoming increasingly important in many countries as herbal medicines have more advantages compared to synthetic drugs.

The African continent has a rich biodiversity among plants in the world, with a very high number of plants used as herbs, such as natural foods and for therapeutic purposes. Many different natural substances have been identified and many of them are used in traditional medicine for treatment of diseases.

Despite the heterogeneous nature of immense biodiversity of the African continent in general and Algeria in particular, there has been little effort devoted to the development of therapeutic agents for these plants. That is why we are interested in studying peppermint, frequently used in the Mediterranean.

MATERIALS AND METHODS

Botanical description of Peppermint (Mentha piperita)

Peppermint is native to the Middle East. It results from a hybridization between water mint (*Mentha aquatica*) and spearmint (*Mentha spicata*). It grows on rich soil cool and moist humus up to 1,800 m altitude. Common plant throughout the temperate regions of the world, especially in Central and Southern Europe, where it is widely cultivated [6].

This is a perennial plant rhizome long, creeping, tracing, chevelu. The rod 50 to 80 centimeters, erect or ascending, is divided in opposite branches. Its leaves are 4 to 10 cm long, are oval, opposite, short-stalked, lanceolate, acute, toothed, are of a beautiful green and are tinged with reddish shades in the sun and copper red in the shade, they are covered with large secretory rounded bristles wherein the fragrant volatiles accumulate [6] [7].

Classification

Reign (Kingdom): Plantae
Division: Magnoliophyta
Class: Magnoliopsida
Order: Lamiales
Family: Lamiaceae
Genre: *Mentha*
Species: *Mentha pepirita*

Figure 12: Peppermint

Peppermint is sometimes regarded as "the world's oldest medicine ', with archaeological evidence placing its use at least from ten thousandths years ago. [6] In therapy, mint is used against fever, weakness, cough, nausea, stomach pain, melancholy, hysteria, sight disorders, it also has medicinal properties, as an example we quote: stimulating the nervous system, tonic, stomachic, antiseptic, analgesic and worming. It is also used against parasites, stems and flowers of mint are burned to drive away fleas mattresses and pets, you can also place the mint sachets with sacks of grain and cheese to hunt rodents . [8] In the food industry include the accreditation requirements, creams, chocolates, candies, chewing gum, desserts, etc. [9] Peppermint can also be found in some shampoos and soaps, that give hair a mint flavor and produce a cooling sensation on the skin. [6]

Extraction of the essential oil

The mint was harvested in the region of Ain El Beida province of Ouargla in the south of Algeria during the period from the January 2015. The plant leaves are then dried in the shade in a dry and ventilated area protect from light. This work was carried out within the process engineering research laboratory (Faculty of Science and Technology and Material Sciences) from the University kasdi merbah Ouargla. The laboratory is responsible for the extraction, characterization and analysis of essential oils and plant extracts of aromatic plants.

Study of the Antibacterial activity of Essential Oil

Five bacteria (Proteus, *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Antirobactère*) were chosen for their high frequency in human infections. Bacterial strains are lots of ATCC (American Type Culture Collection). They are identified and confirmed in the laboratory of the hospital Bodiaf Ouargla Mohamed (Algeria).

The aromatogram is based on a technique used in medical bacteriology, called antibiogram [10], [11]. It has the advantage of being very flexible in the choice of products to test and apply to many bacterial species [12], [13]. In this method, we use filter paper discs of 6mm in diameter, impregnated in different concentrations of essential oil diluted in DMSO at 25%, 50% and 75%. These discs we deposit on the surface of an agar medium inoculated with the surface of a bacterial suspension. The incubation was carried out in an oven at 35°C for 24h for bacteria and at 25°C for 5 days for yeasts. The absence of microbial growth resulting in a translucent halo around the disc whose diameter is measured and expressed in millimeters.

Study of the Antifungal Activity of Essential Oil

For the realization of the antifungal activity was adopted method of direct contact, 03 species within the genus Fusarium are usedin this study (*Fusarium sporotrichioides Fusarium graminearum and Fusarium langsethiae*). To prepare the different concentrations were taken different concentrations of essential oil of *Mentha pepirita* (50, 10, 5, 2.5, 1.25, µl) and adjust to 20 ml PDA then stirred for 5 minutes to homogenize the medium PDA with essential oil.

Chromatographic analysis by GC-MS

Once extracted essential oil of peppermint is obtained, allows the analysis to identify and quantify the products composing it. The essential oil analysis is performed by GC / MS Brucker brand Scion SQ 436 (Agilent Technology)
Column BR-5 Fused Silica 0,25 mm l=15m
gradient method 60 ° C (6 min) to 240 ° C at 3 ° C / min
T injector: 250 ° C (Mode split / spliteless)
Detector: MS SQ (EI)
The vector gas: He
Gas flow 1.7 ml / min
Scan time 18min
T injection: 250 ° C
Oven temperature: 180 ° C
Temperature Detector: 250 ° C
Injected Quantity: 0,5µl

RESULTS AND DISCUSSION

Antibacterial activity

The results obtained with the experiment showing that the essential oil of *Mentha pepirita* has a different effect on the resistance of germs: for *Pseudomonnas aeroginosa* and *antirobactère* are moderately sensitive strains, against *Escherichia coli* and *Proteus* are strains that are highly sensitive.

Antifungal activity

Antifungal activity is revealed by the absence or presence of mycelial growth. The results of antifungal activity diameter of the essential oil of *Mentha pepirita* is presented in the following photos. photo N° 1 shows mycelial growth (mm) of *Fusarium sporotrichioides Fusarium graminearum and Fusarium langsethiae* according to incubation time and the concentration of essential oil of *Mentha pepirita*

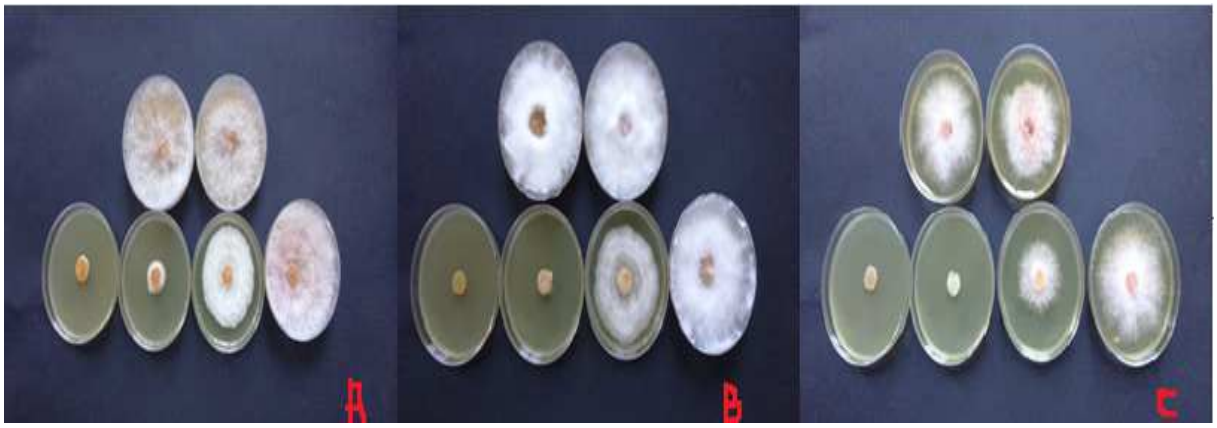

Photo . 01 : Antifungal Activity of Fusarium sporotrichioides Fusarium *graminearum* and Fusarium langsethiae of Mentha pepirita

With different concentrations of essential oil extracted from *Mentha pepirita photo* A, we observe that mycelial growth of *Fusarium sporotrichioides* is remarkable after 72 hours for the control and different concentrations of essential oil of *Mentha pepirita:* 0.0125, 0.025 and 0,05µl. According to the photo B which represent the antifonguique activity of *Fusarium graminearum* depending on the incubation time and the concentration of essential oil of *Mentha pepirita* one notices that there is an increase of mycelial growth with the incubation time. In accordance with the photo C representing Antifungal activity of *Fusarium langsethiae* With different concentrations of essential oil extracted from *Mentha pepirita*, we observe that mycelial growth is remarkable after 72 hours for the control and the different essential oil concentrations the plant 0 0062 and 0.0125 µl by cons 0.025 and 0.05 µl we observed no mycelial growth of *Fusarium langsethiae* in *Mentha pepirita*

Chromatographic analysis by GC-MS

Gas chromatography–mass spectrometry analysis of essential oil

The results of the analysis by gas chromatography-mass spectrometry of the chemical composition of the HE are presented in Table 1.

Table 1: Chemical composition of the essential oil of *Laurus nobilis* leaves from Algeria

compounds	Quantity (%)	formula
Limonen oxide, trans	23,3	$C_{10}H_{16}O$
7- Oxabicyclo[4,1,0]heptane,1-methyl-4-(methylethenyl)-	14,6	$C_{10}H_{16}O$
Cis-(-)-1,2-Epoxy-p-menth-8-ene	5,72	$C_{10}H_{16}O$
Bicyclo[2.2.1]heptane-2,5-diol,1,7,7-trimethyl-,(2-endo,5-exo)-	4,04	$C_{10}H_{18}O_2$
Isoneral	3,88	$C_{10}H_{16}O$
Cis-Verbenol	3,13	$C_{10}H_{16}O$
Verbenol	2,46	$C_{10}H_{16}O$
2-Cyclohexen-1-ol,1-methyl-4-(1-methylethenyl)-,trans-	2,26	$C_{10}H_{16}O$
3,6-Octadienal,3,7-dimethyl	1,82	$C_{10}H_{16}O$
trans –Verbenol	1,47	$C_{10}H_{16}O$
3-Oxatricyclo[4,1,1,0(2,4)]octane, 2,7,7-trimethyl-	1,47	$C_{10}H_{16}O$
Isogeranial	1,30	$C_{10}H_{16}O$
p-Mentha-1,8-dien-7-ol	1,15	$C_{10}H_{16}O$
trans -2-caren-4-ol	0,92	$C_{10}H_{16}O$
Bicyclo[3.1.1]hept-2-ene-2-methanol, 6,6-dimethyl-	0,81	$C_{10}H_{16}O$
Cis-p-Mentha-2,8-dien-1-ol	0,72	$C_{10}H_{16}O$
trans -p- Mentha-2,8-dienol	0,72	$C_{10}H_{16}O$
4-Cyclohexylidenebutyraldehyde	0,64	$C_{10}H_{16}O$
p-Mentha-1(7),8-dien-2-ol	0,61	$C_{10}H_{16}O$
o-Mentha-1(7),8-dien-3-ol	0,59	$C_{10}H_{16}O$
3-Cyclohexen-1-carboxaldehyde, 1-3-4-trimethyl	0,50	$C_{10}H_{16}O$
Naphth[2,3-b]oxirene,decahydro-	0,50	$C_{10}H_{16}O$

2,6-Octadiene-1,8-diol,2,6-dimethyl-	0,50	$C_{10}H_{18}O_2$
9,12,15-Octadecatrienal	0,40	$C_{10}H_{30}O$
2,6-Dimethyl-3,5,7-octatriene-2-ol,,E,E-	0,40	$C_{10}H_{16}O$
Bicyclo[3.3.1]non-2-en-9-ol,9- methyl-	0,37	$C_{10}H_{16}O$
Artemiseole	0,37	$C_{10}H_{16}O$
p-Mentha-1(7),8(10)-dien-9-ol	0,37	$C_{10}H_{16}O$
Carveol	0,35	$C_{10}H_{16}O$
(S)-(-)-(4-Isopropenyl-1-cyclohexenyl) methanol	0,34	$C_{10}H_{16}O$
Cis,cis,cis7,10,13-Hexadecatrienal	0,29	$C_{10}H_{26}O$
Cosmen-2-ol	0,29	$C_{10}H_{16}O$
4a,8a-Naphthalenediol,octahydro-,cis-	0,28	$C_{10}H_{18}O_2$
Limonene diepoxide	0,24	$C_{10}H_{16}O_2$

A total of Thirty four compounds were identified, corresponding to a percentage of 75,46% of all the isolated components. The Limonen oxide, trans to be the major constituent of the essential oil (23,3%), followed by 7-Oxabicyclo[4,1,0]heptane,1-methyl-4-(methylethenyl)- (14,6%), Cis-(-)-1,2-Epoxy-p-menth-8-ene (5,72%), and Bicyclo[2.2.1] heptane-2,5-diol,1,7,7-trimethyl-,(2-endo,5-exo)- (4,04%).
The major components Translated By the chromatographic profile (fig.1)

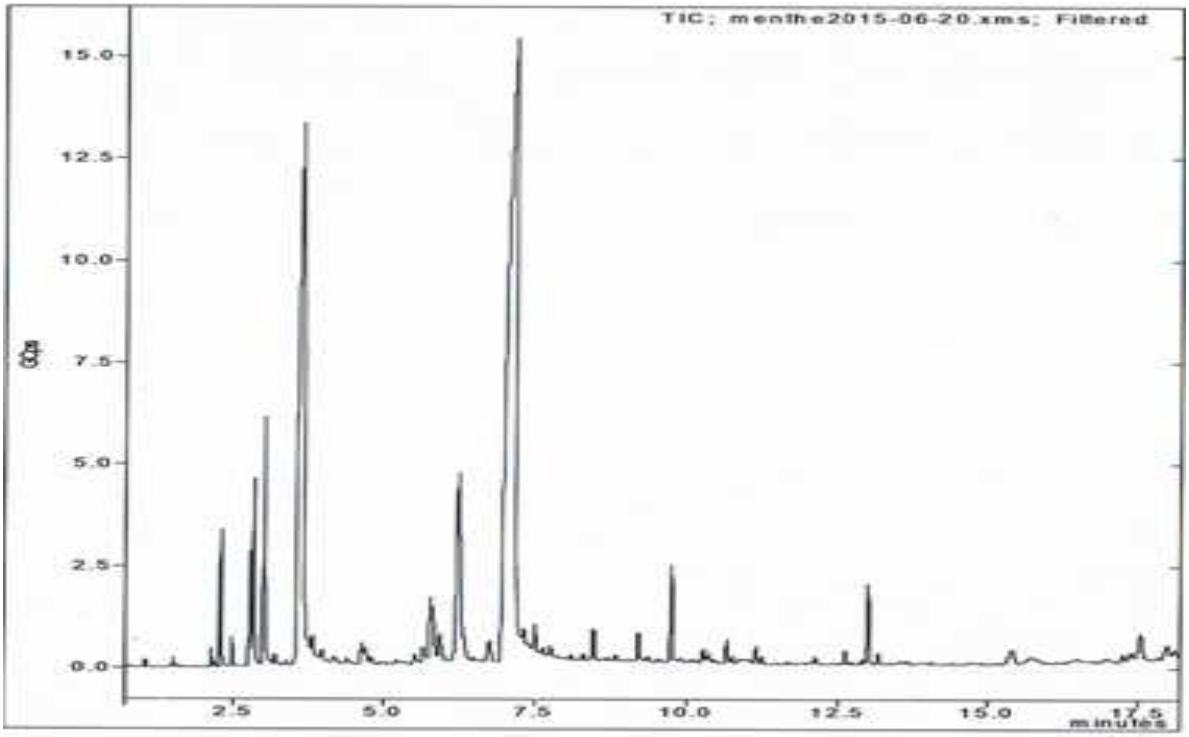

Figure 1- EO of Mint chromatogram analysis by GC -MS

The essential oil's content showed some variations in the same plant, harvested from different geographical origins, and from different parts of the tree. While comparing between the oil's composition of the *Mentha pepirita*, collected in different country, Our results are concomitant with other results found by [14]. [15,16] have found that carvone and D-limonene as a major compounds of this ET with values> 56%, in particular other [13] found the carvone (51.7%), cis-carveol (24.3%), limonene (5.3%), cisdihydrocarvone (2.2%) and germacrene D (1.5%). Sokovic and Griensven reported carvone (49.52%), menthone (21.92%), limonene (5.77%) and 1,8-cineole (3.06%) as oil major composed for Mentha spicata cultivated in Montenegro. Kofidis et al. reported that linalool is the major component for the EO Mentha spicata Greece.

[18] showed that essential oil extracted from the plant Mentha pepirita from the area of the ghardaia that is in south Algerian who has almost the same condition that the extrinsic poivérée mint harvested in the wilaya of Ouargla, is composed by trans carveol (58.98%) D-limonene (19.94%), carvone (2.07%), 4-terpineol (3.01%) α caryophyllene (1.56%), germacrene-D (1.42%) α-pinene (1.26%), β-pinene (2.42%)

we noticed that there are there is evidence that the chemical composition changes in the essential oils could be attributed to the geographical origin of the plant, the extraction technique, time of harvest and climate factors.

CONCLUSION

Mint is an aromatic herb highly sought by the pharmaceutical, aromatherapy, and several areas. Determining performance of the organoleptic characteristics and chemical compositions of the essential oils have been investigated.

The evaluation of the chemical composition of properties is an interesting and useful task, especially for new sources of natural chemical agents. In this context, we tried to evaluate the chemical composition of essential oil extracted from the poiverée mint grown in southern Algeria.

In the development of this study, the essential oil of peppermint from the Ouargla region, shows that he has gifted biological activities due to its wealth of bioactive compounds (Limonen oxide, trans) meter among the plants that it must be valued and discover its secrets. It would be interesting to conduct further study on this plant and other medicinal plants to isolate, purify and identify the active ingredients with biological activities.

Our results clearly demonstrate that the essential oils of *Mentha pepirita* can well present an interesting alternative natural, which it can be useful for food preservation pharmaceutical treatment and aromatherapy products.

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