



## Chemical composition and antibacterial activity of essential oil from leaves and twigs of *Pistacia lentiscus* growing in Mostaganem Province (Algeria)

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### Abstract

This study was designed to examine the chemical composition and the antibacterial activity of essential oil of *Pistacia lentiscus* (Anacardiaceae) from Mostaganem province (northwest of Algeria). Oil was obtained using the hydrodistillation method (Clevenger type) and subsequently analyzed by Gas Chromatography–Mass Spectrometry (GC–MS). The *in vitro* antimicrobial activity against some clinical pathogens was evaluated using the agar diffusion method, the minimum inhibitory concentrations (MIC) were also determined against the same microorganisms using the microdilution method. Among the 50 constituents identified (representing 99.9% of the oil composition) of which the monoterpene hydrocarbons are the dominated (72.43%);  $\alpha$ -pinene (42.13%), sabinene (6.46%),  $\gamma$ -terpinene (6.21%) et  $\alpha$ -terpinolene (2.18%) being the main components. Antimicrobial activity revealed that the essential oil had promising anti-microbial effects against several multiresistant bacteria, giving satisfactory zone diameter values (40.00, 24.71, 24.60, 23.54, 16.07, 14.58 and 12.86 mm) and MIC values (0.05, 0.1, 0.1, 0.2, 0.2, 0.2 and 0.1%) for gram-negative bacteria: *Helicobacter pylori*, *Escherichia coli*, *Morganella morganii*, *Enterobacter cancerogenus* and *Serratia fonticola* and gram-positive bacteria: *Staphylococcus aureus* and *Enterococcus faecalis*, respectively.

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## Introduction

Medicinal plants have been used in traditional medicine since ages. Currently, despite the multiplicity of curative substances, the search on medicinal plants have led to discover novel drug against diverse diseases (Derwich *et al.* 2010). The World Health Organization (WHO) has also indicated that herbal medicines serve the health needs of about 80 percent of the world's population ; especially for millions of people in the vast rural areas of developing countries (Hosseinzadeh *et al.*, 2015). Essential oils, also called volatile odoriferous oil, are aromatic oily liquids extracted from different parts of plants :Leaves, peels, barks, flowers, buds, seeds, and so on (Tongnuanchan and Benjakul, 2014). There are at least 150 types of essential oils that have been traded in the international market (Kusuma and Mahfud, 2017). *Pistacia lentiscus* (Anacardiaceae) is known by its long utilisation in folk medicine since the ancient Greeks. It is an evergreen shrub or small tree from 1 to 8 meters tall (Iauk *et al.*, 1996). *P. lentiscus* is very common in the mediterranean basin, it is found in the wild, in scrub and scrubland in all types of soil, although it prefers siliceous soils (More and White, 2005). The essential oil and gum of the cited plant have been widely used as food and drink additives in the Mediterranean region, without any toxicity reported (Loutrari *et al.*, 2006; Ghalem and Mohamed, 2009).

The essential oil of *Pistacia lentiscus* is also used in cosmetics, perfumery and as a flavoring agent in food preparations (Daferera *et al.*, 2002). In Algeria, the leaves of *Pistacia spp.* were used to purify water and to increase the time of conservation of dry figs and sun-dried tomatoes; they are also used as natural preservatives for fish and meat products (Djenane *et al.*, 2011). Scientific findings also revealed the wide pharmacological activities from various parts of *Pistacia*, such as antioxidant, antimicrobial, antiviral, anticholinesterase, anti-inflammatory, antinociceptive, antidiabetic, antitumor, antihyperlipidemic, antiatherosclerotic, and hepatoprotective activities and also their beneficial effects in gastrointestinal disorders (Bozorgiet

*al.*, 2013; Remila *et al.*, 2015; Della *et al.*, 2013). According to Van der Berg (1998), the essential oil of this plant has an anti-*Helicobacter pylori* activity and can be beneficial in the treatment of peptic ulcer.

Therefore, due to the importance of this plant, we conducted the present study to investigate the chemical composition of essential oil of *Pistacia lentiscus* harvested from Mostaganem region (Algeria) and to evaluate its antibacterial potential against many of pathogens.

## Material and methods

### Study area and plant material

The samples of aerial parts (leaves and twigs) of *Pistacia lentiscus* (Fig. 1), were harvested in March 2016 from the Mostaganem province located at 35°55'52" N, 0°05'21" E, at an altitude of 85 m and with an area of 2269 km<sup>2</sup> large. The region is characterized by a Mediterranean climate and showed relatively abundant populations of *Pistacia*. The average yearly temperatures and total precipitation amounts are 17.9°C and 347mm (www.fr.climate-data.org).

### Essential oil extraction

A sample of 250 g of whole fresh leaves and twigs of *Pistacia lentiscus* were subjected to hydrodistillation in a Clevenger apparatus (Fig. 2) for 3 h with 2000 ml of distilled water according to Duru *et al.* (2003). The essential oil obtained was dried over anhydrous sodium sulfate (Na<sub>2</sub>SO<sub>4</sub>). The oil was finally stored in obscurity at 4°C until further analysis (Gardeli *et al.*, 2008). The yield of essential oil was calculated using the equation below (Kusuma and Mahfud, 2017).

$$y = \frac{V}{w} \times 100$$

Where: y is the oil yield (% w/w), V is the weight of extracted oil (g), and W is the weight of fresh plant parts (g).

### Essential oil analysis

The analysis of the essential oil was performed using a Shimadzu GC-2010 chromatograph connected to an MS-QP2010 SE mass spectrometer at Organic and Macromolecular Chemistry Research Unit (URCOM),

University of Le Havre, France.

Essential oil was initially diluted to 1/100 (v/v) in ethanol 96°. The separation of compounds was carried out on a ZB-5MS capillary column (5% phenyl, 95% dimethylsiloxane, 30 m × 0.25 mm, 0.25 µm film thickness). The carrier gas used was Helium (He) at a flow rate of 1 ml/min. The volume of injections was 1 µL of an ethanol-solution oil, injected in split mode (ratio split 1/50). The column was programmed initially at 50°C for 3 min, increased gradually to 270°C with a 5 °C/min heating ramp and subsequently maintained for 15 min. The mass spectrometer was operated in electron impact mode with ionization energy of 70 eV, the analyzer is carried out in the scanning range of 35-300 m/z. Oils components were identified by co-injection with standards (wherever possible) and confirmed with National Institute of Standards and Technology (NIST) V.2.0 GC-MS library. The relative concentration of each compound in the essential oil was expressed as percentage by peak area normalization (Babushok *et al.*, 2011).

#### *Antibacterial activity of essential oil*

The essential oil of *Pistacia lentiscus* was screened against seven bacterial species relatively resistant to the antibiotics usually used in therapy: five Gram-negative (*Helicobacter pylori*, *E. coli*, *Morganella morganii*, *Enterobacter cancerogenus* and *Serratia fonticola*) and two gram positive (*Staphylococcus aureus* and *Enterococcus faecalis*). *Helicobacter pylori* was isolated from gastric biopsies of a patient suffering from a gastric ulcer at the Pasteur Institute, Algiers, while the other bacteria were isolated from the stools of 20 patients hospitalized in Ain Tedeles district, Mostaganem. All bacteria were identified by studying their cell morphology and by biochemical tests using API system (API 20 E, API 20 NE, STAPH and API CAMPY bioMérieux Marcy-l'Etoile, France).

The chromatogram is a lab test that allows phytotherapists to analyze in vitro the antibacterial activity of essential oils and to more accurately select those essential oils best able to suppress or destroy

the targeted germs (Peter and Kate Damian, 1995). Different types of chromatograms, in solid, liquid, are exploitable. However, in everyday practice, the solid medium is the simplest and most easily reproducible (Pibiri, 2005).

The antibacterial activity of essential oils was determined by the agar diffusion method (Hazzit *et al.*, 2009). Hense, Petri dishes (90 mm) were prepared by pouring 20 ml of Muller Hinton Agar (MHA) medium and allowed to solidify and to dry for 30 min. McFarland density of bacterial culture was adjusted in normal saline (85%, v/v) using densitometer to achieve the final concentration of ~10<sup>6</sup> UFC/ml of each test bacteria individually (Mohapatra *et al.*, 2011) and 0.1 ml of standardized inoculum suspension (0.5 McF ~10<sup>6</sup> UFC/ml) was poured and uniformly extended and the inoculum was allowed to dry for 5 min.

To prepare the sample stock solution, a volume of pure essential oil was dissolved at 10% (v/v) in dimethylsulfoxide (DMSO) (Sigma Aldrich-Química, S.A.). Then, sterile filter paper discs of 6 mm diameter (Filter LAB ANOIA, Barcelona, Spain) were impregnated with 5 µL of essential oil using a micropipette. The dishes were left for 15 min at room temperature to allow diffusion of the essential oil and then incubated at 37 °C for 24 hours. A negative control was carried out by deposition of 5 µL of DMSO on disks stored on a previously inoculated medium of the tested bacterium. Each assay in this study was replicated 3 times.

Laboratory growth of *Helicobacter pylori* is difficult and can be achieved by using complex media containing serum, blood, or blood derivatives (Olivieri *et al.*, 1993). For the antimicrobial test against *Helicobacter pylori*, the inoculated plates were incubated in MHA plus 10% horse blood for 48 h at 37°C under microaerophilic environment, obtained with a GENbox Microaer paper sachet (Biomérieux) inside an anaerobic jar (oxygen concentration, 5%; CO<sub>2</sub> concentration, 10%) and incubated for 48-72 h at 37°C (Medouakh, 2010). After incubation, the

diameter of the clear zone around the disc was measured using a slurry foot and expressed in millimeters (mm) as antimicrobial activity. The sensitivity of the tested bacteria to the essential oil is classified according to the halos of inhibition diameters :  $\varnothing < 8$  mm: resistant bacteria;  $9 \text{ mm} < \varnothing < 14$  mm: sensitive bacteria;  $15 \text{ mm} < \varnothing < 19$  mm: very sensitive bacteria and  $\varnothing > 20$  mm: extremely sensitive bacteria (Ponce *et al.*, 2003).

The antibacterial tests and the determination of the minimum inhibitory concentration (MIC) are carried out according to the method reported by Remmal *et al.* (1993) and Farah *et al.* (2001). The essential oil is emulsified in 10% (DMSO) in order to disperse the compounds and to improve their contact with the microorganisms tested. Dilutions were prepared at 1/10, 1/25, 1/50, 1/100, 1/200, 1/300, 1/400 and 1/500 in DMSO solution. A volume of 1.5 ml of each of the dilutions is added to test tubes containing 13.5

ml of the nutrient agar MHA previously sterilized for 20 min at 120°C, cooled to 45°C and poured into Petri dishes. The final concentrations of essential oil are : 1/100, 1/250, 1/500, 1/1000, 1/2000, 1/3000, 1/4000 and 1/5000 (v/v). Negative Controls containing culture medium and DMSO alone were also prepared.

## Results and discussion

### *Chemical composition of Pistacia lentiscus essential oil*

The average yield of the essential oil extracted from leaves and twigs of *Pistacia lentiscus* was 0,39 %. This finding agrees with results of Arab *et al.* (2014) in the Boumerdes province, Algeria, Zrira *et al.* (2003) in Morocco and Congiu *et al.* (2002) in Sardinia. Our values were however highest than those reported in Tunisia (Amri *et al.*, 2012) and Grec (Tsokou *et al.*, 2007).

**Table 1.** Chemical composition of volatile oil isolated by hydrodistillation from *Pistacia lentiscus* from Mostaganem region, Algeria.

Compound <sup>a</sup>	RT (min)	RI	Area	%
Tricyclene	7.475	921	601719	0.24
$\alpha$ -Thujene	7.6	925	594140	0.24
$\alpha$ -Pinene	7.87	934	104477800	42.13
Camphen	8.34	949	2910112	1.17
Sabinene	9.07	972	16021996	6.46
$\beta$ -Pinene	9.21	976	4370203	1.76
$\beta$ -Myrcene	9.575	988	3367891	1.36
$\alpha$ -phellandrene	10.11	1005	1521771	0.61
$\alpha$ -Terpinene	10.45	1016	10162820	4.1
<i>o</i> -Cymene	10.69	1023	1792977	0.72
Limonene	10.84	1028	4471214	1.8
$\beta$ -Phellandrene	10.89	1030	4329380	1.75
E- $\beta$ -Ocimene	11.05	1035	502755	0.2
Z- $\beta$ -Ocimene	11.385	1045	3715100	1.5
Butyrate iso amyl	11.705	1055	1205552	0.49
$\gamma$ -Terpinene	11.77	1057	15413257	6.21
$\alpha$ -Terpinolene	12.615	1084	5417549	2.18
2-Nonanone	12.77	1089	1987035	0.8
Linalool	13.065	1098	297633	0.12
Nonanol	13.125	1100	1046866	0.42
iso-Amyl isovalerate	13.275	1105	426275	0.17

Terpinen-4-ol	15.575	1180	15434061	6.22
$\alpha$ -Terpineol	16	1194	7313787	2.95
Isopentyl hexanoate	17.565	1248	915591	0.37
Isopamyl hexanoate	17.625	1250	509819	0.21
Bornyl acetate	18.565	1282	2562876	1.03
2-Undecanone	18.77	1289	2615408	1.05
2-Tridecanol	19.05	1299	243635	0.1
$\beta$ -Elemene	21.435	1388	316135	0.13
Charyophelene	22.265	1420	10991141	4.43
Isoamyl benzoate	22.685	1437	655542	0.26
$\alpha$ -Humulene	23.18	1456	1810825	0.73
$\beta$ -Cadinene	23.575	1471	234555	0.09
$\gamma$ -Muurolene	23.645	1474	597657	0.24
Germaacrene D	23.82	1481	3357121	1.35
Valencene	24.18	1495	309789	0.12
$\alpha$ -Muurolene	24.23	1497	563572	0.23
$\gamma$ -Cadinene	24.6	1512	391191	0.16
Cubebol	24.715	1517	2397832	0.97
Spathulenol	26.155	1576	423032	0.17
Caryophyllene oxide	26.305	1582	250775	0.1
Globulol	26.38	1585	328341	0.13
Cubenol	27.255	1621	280067	0.11
T-Muurolol	27.345	1625	586024	0.24
Epi-Cadinol	27.705	1639	3847662	1.55
Muurolol	27.77	1642	833174	0.34
$\alpha$ -Cadinol	27.975	1651	4920965	1.98
Bisabolol	28.07	1654	237741	0.1
Benzyl Benzoate	30.42	1751	279968	0.11
Manool oxyde	35.105	1943	255446	0.1
<i>Monoterpenes</i>				
Hydrocarbons				72.43
Oxygenated				9.29
<i>Sesquiterpenes</i>				
Hydrocarbons				7.48
Oxygenated				5.69
Ketones				
Alcohols				
Esters				
Unknown				
Total identified				99.9

<sup>a</sup> Compounds listed in order of elution from an ZB-5MS capillary column.

RT: Retention time obtained by chromatogram (Fig. 3).

RI: Retention index.

The oil yield of *Pistacia lentiscus* seems to depend on the nature of plant parts used, the extraction method and geographical origin. According to Okoh *et al.* (2007), the yield of essential oil showed a maximum at the full flowering stage (0.97%) and a minimum during the pre-flowering stage (0.13%).

The constituents of leaves and twigs essential oil of *Pistacia lentiscus* are listed in order of their elution on the ZB-5MS capillary column (Fig. 3).

The GC-MS analysis of the *Pistacia lentiscus* essential oil resulted in the detection of 50 components

comprising 99.9% of the oil (Table 1). The essential oil was characterized by a high percentage of monoterpene hydrocarbons (72.43%), followed by oxygenated monoterpenes (9.29%) and sesquiterpene hydrocarbon (7.48%), while the oxygenated sesquiterpenoid fraction was 5.69%. Similar findings have been reported in the literature (Dob *et al.*, 2006). Different compounds have been observed in several studies of the chemical composition of essential oils of *pistacia lentiscus* in the Mediterranean countries (Castola *et al.*, 2000; Ben Douissa *et al.*, 2005; Derwich *et al.*, 2010).

**Table 2.** Antibacterial activity of essential oil of *Pistacia lentiscus* collected in the region of Mostaganem, Algeria against seven pathogenic bacterial strains.

Strain	Oil concentrations								NC
	1/100	1/250	1/500	1/1000	1/2000	1/3000	1/4000	1/5000	
<i>Hp</i>	-	-	-	-	+	+	+	+	+
<i>Ec</i>	-	-	-	+	+	+	+	+	+
<i>Mm</i>	-	-	-	+	+	+	+	+	+
<i>Enc</i>	-	-	+	+	+	+	+	+	+
<i>Sf</i>	-	-	+	+	+	+	+	+	+
<i>Sa</i>	-	-	-	+	+	+	+	+	+
<i>Ef</i>	-	-	+	+	+	+	+	+	+

- : inhibition ; + : growth ; *Hp* : *Helicobacter pylori* ; *Ec* : *Escherichia coli* ; *Mm* : *Morganella morganii* ; *Enc* : *Enterobacter cancerogenus* ; *Sf* : *Serratia fonticola* ; *Sa* : *Staphylococcus aureus* ; *Ef* : *Enterococcus faecalis*, NC = negative control.

In this study  $\alpha$ -pinene (42.13%) was the major compound of the essential oil. This compound was also abundant in the samples from Oran, Algeria (19.0%) (Dob *et al.*, 2006), France (31.9%) (Castola *et al.*, 2000), Spain (13.0%) (Ana Fernández *et al.*, 2000) and Morocco (16.1% - 38.5%) (Zrira *et al.*, 2003). The other main constituents of the oil were sabinene (6.46%), terpinen-4-ol (6.22%),  $\gamma$ -terpinene (6.21%), charyophellene (4.43%),  $\alpha$ -terpinene (4.1%),  $\alpha$ -terpineol (2.95%) and  $\alpha$ -terpinolene (2.18%). Furthermore, this majority is also observed in the chemical composition of the essential oil of *pistacia lentiscus* in Spain, whose main compounds were  $\alpha$ -pinene (24.9%) followed by terpinen-4-ol (6.8%), sabinene (4.6%),  $\gamma$ -terpinene (3.3%),  $\alpha$ -terpineol

(2.5%),  $\alpha$ -terpinene (2.2%) and trans-caryophyllene (2.0%) (Ana Fernández *et al.*, 2000). However, the oils obtained from fresh leaves of *Pistacia lentiscus* collected in Tunisia (Aissiet *et al.*, 2016) displayed a different profile than that detected in the present study since it consisted mainly of germacrene D (11.9%),  $\alpha$ -pinene (9.9%), limonene (8.5%),  $\delta$ -cadinene (8.5%),  $\beta$ -caryophyllene (8.2%) and terpinen-4-ol (5.1%). Similarly, comparing our results with those reported by Kivçak *et al.* (2004), the major compounds in Turkey were terpinene-4-ol (29.2%),  $\beta$ -caryophyllene (29.2%) and p-cymene (7.1%). According to Olga Tzakou *et al.*, (2017) the common feature for the samples of *Pistacia atlantica* studied, as well as for the most studied *Pistacia* species, is the biosynthesis of monoterpenoids as the

main class of compounds in their essential oils, irrespective of the observed variability of the terpene composition of the oils.

The chemical composition of essential oils can vary among species and among the different plant parts in the same species. Boelens and Jimenez (1991) reported that the main constituents of the gum oil were: 79%  $\alpha$ -pinene and 3%  $\beta$ -myrcene; of the leaf oil: 11%  $\alpha$ -pinene and 19%  $\beta$ -myrcene; of the unripe-fruit

oil: 22%  $\alpha$ -pinene and 54%  $\beta$ -myrcene, and of the ripe-fruit oil: 11%  $\alpha$ -pinene and 72%  $\beta$ -myrcene. Other factors can affect the oil chemical composition such as climate, soil quality, harvest season, genetics (Cunha *et al.*, 2013) and nutrients (Djenane *et al.*, 2011). Otherwise, yield and composition of the oils were correlated with herbivores, weather parameters (day length, temperature and humidity) and to the attack of fungal pathogens, particularly in the months of rainfall (Hassiotis *et al.*, 2010).

**Table 3.** Mean values of Diameter of Inhibition Zone (DIZ, mm) and Minimal Inhibitory Concentration (MIC) of essential oil of *Pistacia lentiscus* collected in the region of Mostaganem against seven pathogenic bacterial strains.

Strain	DIZ	MIC %	Oil sensitivity
<i>Helicobacter pylori</i>	40.00	0.05	ExS
<i>E.coli</i>	24.71	0.1	ExS
<i>Morganella morganii</i>	24.60	0.1	ExS
<i>Enterobacter cancerogenus</i>	16.07	0.2	VS
<i>Serratia fonticola</i>	14.58	0.2	S
<i>Staphylococcus aureus</i>	23.54	0.2	ExS
<i>Enterococcus faecalis</i>	12.86	0.1	S

ExS= Extra sensitive ; VS= Very sensitive ; S= Sensitive.

### Antimicrobial activity

Lately it has been targeted the interest for biologically active molecules, isolated from plant species to eradicate pathogenic microorganisms.

The *in vitro* antibacterial activity of *Pistacia lentiscus* essential oil were qualitatively and quantitatively assessed by the presence or absence of inhibition zones and MIC values, respectively.

As seen in tables 2 and 3, the essential oil of *Pistacia lentiscus* has displayed an important inhibitory activity against both gram positive and gram negative bacteria tested where the average diameter of inhibition zone ranged from 12.86 to 40.00 mm (table 3) as also reported in the literature (Magiatis *et al.*, 1999; Koutsoudaki *et al.*, 2005; Mharti *et al.*, 2011).

The diameters of the inhibition zones allowed us to classify the bacterial strains according to their

sensitivity to the essential oil tested according to the spectrum indicated above (Ponce *et al.*, 2003). For gram-negative bacteria, the largest zones of inhibition were obtained for *Helicobacter pylori*, *E. coli* and *Morganella morganii* (40.00, 24.71 and 24.60 mm, respectively). It was thus considered that these organisms were extra sensitive to the oil. In addition, *Enterobacter cancerogenus* was found to be more sensitive to the oil than *Serratia fonticola* (16.07 and 14.58 mm, respectively). In the case of gram positive bacteria, *Staphylococcus aureus* has proved to be more sensitive than *Enterococcus faecalis* with inhibition diameters of 23.54 and 12.86 mm, respectively.

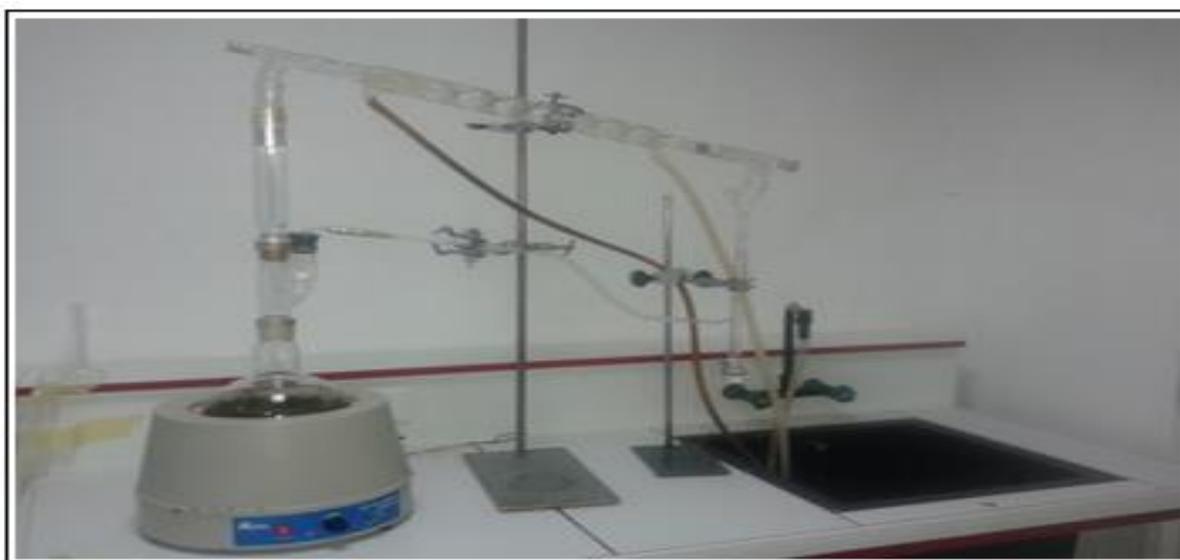
The Minimal Inhibitory Concentration (MIC) was defined as the lowest concentration of the test samples where the absence of growth was recorded (Ponce *et al.*, 2003). The MIC of essential oil of *Pistacia lentiscus* was tested at concentrations ranging from 1/100 to 1/5000 (v/v).



**Fig. 1.** *Pistacia lentiscus* of Mostaganem province, Algeria.

As seen in table (3), the essential oil of *Pistacia lentiscus* revealed a strong inhibitory activity against all germs tested. Although, the microorganisms studied did not express the same sensitivity. The data indicated that *Helicobacter pylori* is inhibited at a concentration of 1/1000 (v/v), while *Escherichia coli*,

*Morganella morganii* and *Staphylococcus aureus* were inhibited at a concentration of 1/500 (v/v). However, a higher MIC value (1/250) (v/v) was obtained with *Enterobacter cancerogenus* and *Enterococcus faecalis*.



**Fig. 2.** The experimental setup for *Pistacia lentiscus* essential oil extraction by hydrodistillation (Clevenger type).

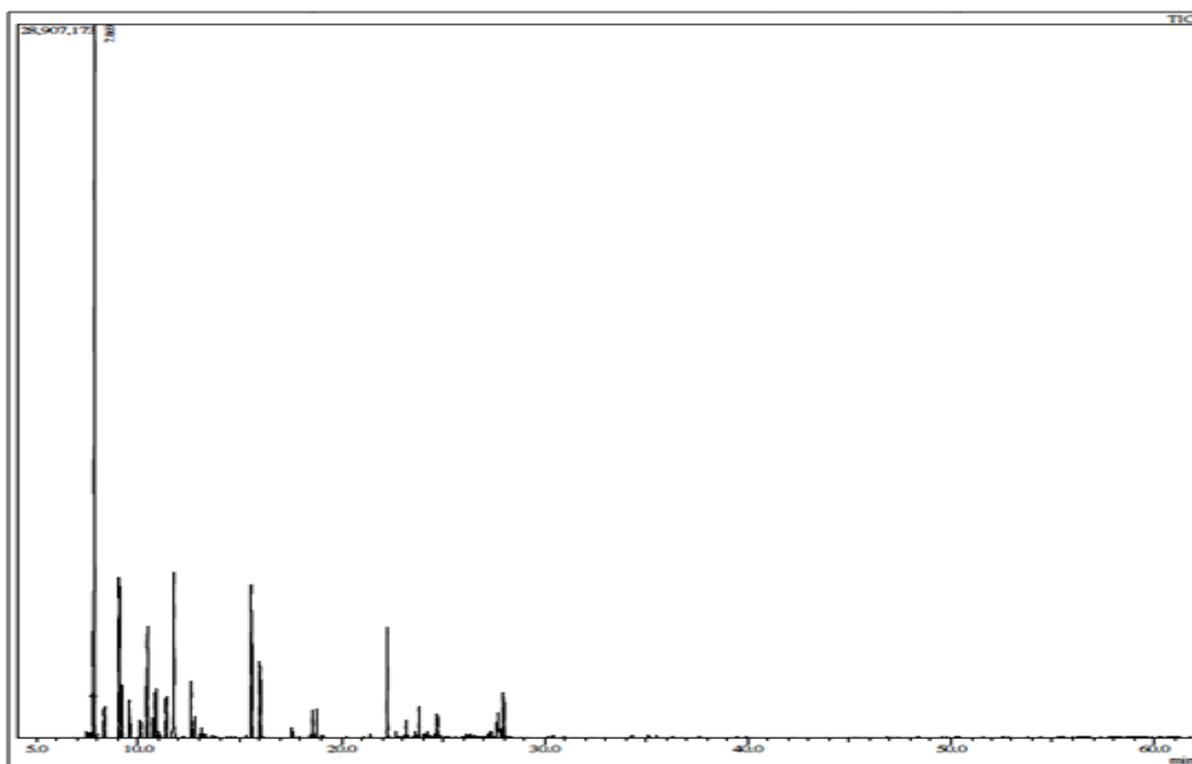
Differences in MIC values of bacteria may be related to differential susceptibility of bacterial cell wall, which is the functional barrier to minor differences present in the outer membrane in the cell wall composition (Zhao *et al.*, 2001). Gram-negative bacteria are surrounded by a thin peptidoglycan cell

wall, which itself is surrounded by an outer membrane containing lipopolysaccharide which creates a barrier toward hydrophobic compounds such as those found in essential oils. While, gram-positive bacteria lack an outer membrane but are surrounded by layers of peptidoglycan many times

thicker than is found in the gram-negatives (Silhavy *et al.*, 2010).

Koutsoudaki *et al.* (2005) and Burt (2004) reported that plant extracts are more active against gram-positive than gram-negative bacteria. However, results of this study are supported by Zaika (1988) hypothesis who proposed that gram-positive bacteria are more resistant than gram-negative bacteria to the antibacterial properties of plant volatile oils. Indeed, several researchers have reported that there is a relationship between the most abundant volatile compounds in the essential oil tested and the antimicrobial activity (Ghalem and Mohamed, 2009; Koutsoukadi *et al.*, 2005). According to Rios and Recio (2005), extracts or oils from plant species with MIC values below 100 µg/ml are considered

promising as potential antimicrobial agents. Halouiet *al.* (2015) reported that leaves essential oil exhibit a higher antibacterial effect with MIC values of 0.015, 0.5, 1 and 4 % fold least compared to twigs essential oil with MIC values of 0.5, 4, 4 and 16% against *Bacillus subtilis*, *Staphylococcus aureus*, *E. coli* and *Pseudomonas aeruginosa*, respectively. According to Hafset *al.* (2017) the minimum inhibitory concentration for *Mycobacterium aurum*, *Bacillus sp.* and *Staphylococcus aureus* was 1/250 (v/v), while the most resistant strains were *Pseudomonas aeruginosa*, *Enterococcus faecalis* and *Salmonella sp.* with an MIC of 1/125 (v/v). In an other hand, Medjkane *et al.* (2016) reported a strong antimicrobial activity of *Pistacia lentiscus* essential oil against algerian clinical isolates of *Helicobacter pylori* with a MIC of 1/5000 (v/v).



**Fig. 3.** Chromatogram of *Pistacia lentiscus* from Mostaganem, Algeria.

In the present work,  $\alpha$ -pinene (42.13%) is the main compound of essential oil of *Pistacia lentiscus* studied. Several authors reported that essential oils rich in  $\alpha$ -pinene have demonstrated potential antibacterial activity. Naturally, the antibacterial efficacy of essential oil of *pistacia lentiscus* is due to a number of its components working synergistically

(Derwich *et al.*, 2010).

### Conclusion

This work was carried out to study the chemical composition of the essential oils of *Pistacia lentiscus* and to evaluate its antibacterial activity in vitro. Based on the results of composition analysis of oil,  $\alpha$ -

pinene was detected as the main compounds. The results of the antibacterial activity tests indicate that essential oil of *Pistacia lentiscus* exhibited high degree of inhibitory activity against most of the seven tested pathogens. Overall, this study further support the view that *Pistacia lentiscus* are promising as nature source with antibacterial activity and thus confirm its potential uses as antimicrobial agents for industrial applications such as pharmaceutical, perfumery and food preservation. Nonetheless, *in vivo* studies should be conducted to justify and evaluate the potential use of *Pistacia lentiscus* oil.

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