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Chemical composition, antioxidant and antibacterial activities of the essential oil of *Mentha spicata* L. from Algeria

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Abstract
Hydrodistilled essential oil of *Mentha spicata* L., cultivated at Ghardaïa (Algerian Septentrional Sahara), was analyzed by GC and GC/MS. 29 components representing 96.43% of the essential oil were detected with *cis*-carvone oxide (44.06%), 1,8-cineole (15.32%), *cis*-dihydrocarvone (8.85%) and limonene (5.80%) as the major components. Antioxidant activity was investigated using β-carotene/linoleic acid, DPPH•, ABTS cation radical decolorization and Metal chelating assays. Antibacterial activity was performed according to disc diffusion and minimum inhibitory concentration (MIC) methods.

Keywords: Mentha spicata L.; Lamiaceae; essential oil; Antioxidant; Antibacterial

1. Introduction

The genus *Mentha*, one of the most important members of the Lamiaceae family, is represented by 19 species and 13 natural hybrids. 15 *Mentha* species are distributed in Algeria [1] and most of them are used in folk medicine. *Mentha* species have been found to possess significant biological activities, including antimicrobial, anti-stomach, anti-vomitive, antiseptic, anti-infective, vermifuge, antitussive, digestive and diuretic [2–5]. *Mentha spicata* (sperarmint) is the most common and popular mint which is widely grown in temperate areas of the world, particularly Africa, temperate Asia, and Europe, and is cultivated throughout all regions of the world [6]. It is a perennial, rhizomatous and glabrous herb that has a strong aromatic odor. It is used against cough and cold and as a diuretic and a spasmolytic [7]. It has shown analgesic, antipyretic and anti-inflammatory effects as well [8]. It is applied as breath freshener, antiseptic mouth rinse, and toothpaste [9]. The fresh and dried iranian spearmint essential oils have been used as a flavoring agent in various food products, including cheese and doogh (Iranian yoghurt drink), chocolate, beverages, jellies, syrups, candies, ice creams, and chewing gum [10;11].
The essential oils extracted from *Mentha spicata*, worlwide distributed, were characterized by the chemotype couple carvone/limonene (15.3-78.76%)/ (5.3-22.31%) respectively [12-31]. 1,8-Cineole (6.84-17.0%) [12,14-15], menthone (18.6-21.9%) [19;25], piperitenone oxide (24.0-79.2%) [14;22;30], *cis*-carveol (21.3-24.3%) [21;31], piperitone (22.17-28.16%) [15], pulegone (26.71-72.1%) [15;25], *trans*-β-caryophyllene (5.23-8.01%) [15] and α-Humulene (0.1-29.9%) [22] have also been reported as main components of *Mentha spicata* essential oils. In continuation of our works on Lamiaceae essential oils [32-35], we report here the chemical composition, the antioxidant and antibacterial activities of the freshly hydrodistilled essential oil of *Mentha spicata*, cultivated at the Algerian Sahara. Litterature data on reported *Mentha spicata* essential oils is given here.

2. Experimental

2.2. Plant material

The aerial parts of cultivated *Mentha spicata* L., were collected in May 2012, from Ghardaïa (Algerian Septentrional Sahara). A voucher specimen was deposited at the herbarium of the University of Constantine 1, Algeria (LOST Ms.05.12).

2.2. Extraction

The hydrodistillation in a Clenvenger-type apparatus of fresh aerial parts (100 g) of *Mentha spicata*, for 3hours, yielded 2.1% of a pale yellowish essential oil.

2.3. Gas chromatography

GC analyses were performed using a Varian CP3800 gas chromatograph equipped with a cross-linked VF5-MS column (30 m × 0.25 mm, film thickness 0.25 μm). The oven temperature was programmed as isothermal at 60°C for 5 min, then raised to 275°C at 5°C/min and held at this temperature for 5 min. Helium was used as the carrier gas at a rate of 1 ml/min.

2.4. Gas chromatography-Mass spectrometry

GC-MS was performed using a Saturn 2200 mass selective detector. Operating conditions were the same as for the analytical GC. The MS operating parameters were as follows: 0.1 mL of crude oil was mixed with diethyl ether (40 %); ionization potential, 70 ev; ionization current, 2 A; ion source temperature, 200°C; resolution, 1000. scan time, 5 s; scan mass range, 40-400 u; split ratio, 1:50; linear velocity, 30.0 cm/sec. Relative percentage amounts were calculated from peak area without the use of correction factors.

2.5. Identification of components

Essential oil components were identified based on their retention indices (determined with reference to a homologous series of *n*-alkanes (C_6-C_{24}), and by comparison of their mass spectral fragmentation patterns with those reported in the literature [36;37] and with authentic compounds for major components.

2.6. Antioxidant activity

2.6.1. β-Carotene/linoleic acid assay

The total antioxidant activity was evaluated using the β-carotene-linoleic acid model test system, as previously described [38;39]. Ethanol was used as a control while BHA and α-tocopherol were used as antioxidant standards. The results were given as 50% inhibition concentration (IC_{50}). The sample concentration providing 50% antioxidant activity was calculated from the graph of antioxidant activity percentage against sample concentration.

2.6.2. DPPH free radical-scavenging assay

The free radical-scavenging activity was determined spectrophotometrically by the DPPH˙ assay [40]. Ethanol was used as a control while α-Tocopherol and BHA were used as antioxidant standards for comparison of the activity. The scavenging capability of DPPH radical was calculated using the following equation:

$$\text{DPPH scavenging effect (\%)} = [(A_{Control} - A_{Sample})/ A_{Control}] \text{ X } 100$$

Where: $A_{Control}$ is the initial concentration of the DPPH˙ and A_{Sample} is the absorbance of the remaining concentration of DPPH˙ in the presence of the extract and positive controls.

The results were recorded as 50% inhibition concentration (IC_{50}). The sample concentration providing 50% DPPH˙ scavenging effect was calculated from the graph of DPPH˙ scavenging effect percentage against sample concentration.

2.6.3. $ABTS^{\bullet+}$ cation radical decolorization assay

The spectrophotometric analysis of $ABTS^{\bullet+}$-scavenging activity was determined according to the method of Re et al., [41]. The $ABTS^{\bullet+}$ was produced by the reaction between 7 mM ABTS in water and 2.45 mM potassium persulfate, stored in the dark at room temperature for 12 h. Before usage, the $ABTS^{\bullet+}$ solution was diluted to get an absorbance of 0.703 ± 0.025 at 734 nm with ethanol. Ethanol was used as a control, while α-tocopherol and BHA were used as antioxidant standards. The results were given as 50% inhibition concentration (IC_{50}). The sample concentration providing 50% $ABTS^{\bullet+}$ scavenging effect (IC_{50}) was calculated from the graph of $ABTS^{\bullet+}$ scavenging effect percentage against sample concentration.

2.6.4. Metal chelating activity assay

The chelating activity of the oil on Fe^{2+} was measured spectrophotometrically according to the method described by Decker & Welch, [42], and calculated using the following equation.

$$\text{Metal chelating activity (\%)} = [(A_{Control} - A_{Sample})/ A_{Control}] \text{ X } 100$$

2.7. Antibacterial activity

The essential oil was individually used against a range of bacteria, namely *Escherichia coli* ATCC 25922, *Escherichia coli* (HS), *Staphylococcus aureus* ATCC 43300, *Staphylococcus aureus* (HS), *Pseudomonas aeruginosa* ATCC 27853, *Pseudomonas aeruginosa* (HS), *Enterobacter aerogenes* (HS), *Klebsiella pneumoniae* (HS), *Proteus mirabilis* (HS) and *Streptococcus sp.* (HS). The reference strains were obtained from the Pasteur Institute (Algiers). The hospital strains (HS) were obtained from the laboratory of bacteriology, Benbadis Hospital, Constantine, using conventional methods (clinical isolation). Susceptibility of the bacterial strains to the essential oil was investigated using the disk diffusion method and by comparing their antibiogram inhibition zones to those reported by the National Committee for Clinical Laboratory Standards (NCCLS). Freshly cultured bacterial suspensions in Mueller Hinton Broth were standardized to a cell density of $1.5x10^8$/mL (McFarland No. 0.5). Empty sterilized discs were impregnated by the essential oil which was diluted with 20 mL of DMSO used for antibacterial activity assays. The same volume (20 mL) of DMSO was used as control. The diameters of inhibition zones were measured and compared with those suggested by NCCLS (sensitive P≥15 mm). Ampicillin (10µg/mL) and Gentamicin (10µg/mL) were used as a positive reference standard to determine the sensitivity of each bacterial species tested. The inoculated plates were incubated at 37 ºC for 24 h. The susceptibility of the strains to the essential oil was further evaluated by agar dilution method; different concentrations of the essential oil were included in Mueller-Hinton agar plates (sensitive MIC ≤ 32 µg/mL). The minimum inhibitory concentration (MIC) was defined as the concentration at which no colony was observed after incubation [43]. The agar plates were prepared and inoculated with bacterial suspension. After incubation at 37°C for 18–24h, the inhibition zones were measured and averaged. The essays were performed in triplicate. MICs of the essential oils were also determined by an agar dilution method.

2.8. Statistical analyses

All data on both antibacterial and antioxidant activity tests were the averages of triplicate analyses. The data were recorded as means ± standard error meaning (S.E.M.). Significant differences between means were determined by student's-t test; *p* values <0.05 were regarded as significant.

3. Results and Discussion

3.1. Chemical composition

29 components were identified in the essential oil, representing 96.43% of the total oil content. The major constituents of the essential oil were found to be *cis*-carvone oxide (44.06%), 1,8-cineole (15.32%), *cis*-dihydrocarvone (8.85%) and limonene (5.80%) (Table 1).

This composition has been compared to those previously reported and from which material data is given in table 2.

The compositions of previously reported essentials oils of *Mentha spicata* are given in tables 3-4. which show shared chemotypes depending to the geographical distribution of the investigated essential oils.

From table 3, exhibiting the major components (≥5%) of the studied essential oils of *M. spicata*, growing in the Mediterranean area, it appears that the couple carvone/limonene is a chemotype of *M. spicata* collected from Tunisia (**Me1**), Greece (**Me2**), Portugal (**Me6**) and Egypt (**Me7**) [12-13; 17- 18]; the highest content (68.58, 20.8%) is found in **Me7** and **Me1** [18;12], respectively whereas the lowest content (40.8, 14.3%) was found in plants collected from Tunisia and Greece, respectively [12;13]. The essential oil of the species collected from Morocco (**Me5**) is mainly characterized by carvone (29.0%) and *trans*-carveol (14.0%) [16]. Piperitenone oxide (35.7%), and 1,8-cineole (14.5%) were mainly found in the essential oil of **Me3**, collected from Greece [14]. Pulegone (26.71-29.56%), piperitone (22.17-28.16%) dominated the composition of **Me4** essential oil of *M. spicata* from Turkey followed by 1,8-cineole (6.84-7.76%) and *trans*-β-caryophyllene (5.23-8.01%) [21].

Table 1: Chemical composition of the essential oil of *Mentha spicata* cultivated at Ghardaïa

N°	Compounds	RI [a]	(%)
1	α-Pipene	939	1.30
2	Camphene	954	0.56
3	β-Pinene	979	3.23
4	Myrcene	991	4.11
5	Isoamyl 2-methylbutyrate	1023	0.24
6	Limonene	1029	**5.80**
7	1,8-cineole	1031	**15.32**
8	β-phellandrene	1032	1.96
9	Borneol	1132	0.19
10	neo-Verbanol	1145	0.58
11	*trans*-Limonene oxide	1147	0.18
12	(E)-2-Nonenol	1163	0.26
13	Terpinen-4-ol	1177	0.25
14	α-Terpineol	1186	0.16
15	1-Terpineol	1189	0.12
16	*cis*-Dihydrocarvone	1193	**8.85**
17	*trans*-Dihydrocarvone	1201	0.41
18	*trans*-Carveol	1217	0.50
19	*cis*-Carvone oxide	1263	**44.06**
20	*trans*-Carvone oxide	1276	0.24
21	*endo*-bornyl acetate	1279	0.22
22	neo-dihydro carveol acetate	1307	0.63
23	β-Bourbonene	1390	0.15
24	*trans*-Caryophyllene	1419	1.53
25	δ-Cadinene	1531	0.45
26	[3.2.1]Propellane	1537	0.62
27	Caryophellene oxide	1599	0.88
28	1,10-di-epi-Cubenol	1619	0.41
29	*t*-Cadinol	1640	3.28
	Total (%)		**96.43**

[a] RI (retention index) measured relative to n-alkanes (C_6-C_{24}) using a DB5 MS column

Table 2: Material plant data of reported essential oils of fresh aerial parts of *Mentha spicata* L.

Code	Locality	Reference
Me1	Tunisia	[12]
Me2	Greece	[13]
Me3	Greece	[14]
Me4	Turkey	[15]
Me5	Morocco	[16]
Me6	Portugal	[17]
Me7	Egypt	[18]
Me8	Serbia	[19]
Me9	Iran	[20]
Me10	Tamilnadu, India	[21]
Me11	Uttarakhand, India	[22]
Me12	North-West Himalayan region, India	[23]
Me13	India	[24]
Me14	Karnataka, India	[25]
Me15	USA	[26]
Me16	China	[27]
Me17	Yemen	[28]
Me18	Bangladesh	[29]
Me19	Cuba	[30]
Me20	Pakistan	[31]

Table 3: Percentage of major components (≥ 5%) of essential oil of *Mentha spicata* growing in the Mediterranean area.

Compounds	**Me1**	**Me2**	**Me3**	**Me4**	**Me5**	**Me6**	**Me7**
Limonene	20.8	14.3	-	-		20.1	16.42
1,8-Cineole	17.0	-	14.5	6.84-7.76	-	-	-
trans-Carveol	-	-	-	-	14.0	-	-
Pulegone	-	-	-	26.71-29.56	-	-	-
Carvone	40.8	67.1	-	-	29.0	41.1	68.58
Piperitone	-	-	-	22.17-28.16	-	-	-
Piperitenone oxide	-	-	35.7	-	-		-
Copaene	-	-	-	-	-	5.6	-
trans-β-Caryophyllene	-	-	-	5.23-8.01	-	-	-

[a]: Order of elution is given from an apolar column (HP-5 MS).

Table 4, representing the major components of *M. spicata* growing in other countries, showed that the couple carvone/limonene is also a chemotype of most reported essential oils [20-21;23-24; 27-29;31], its highest yield was in **Me12** (76.65; 22.31%) growing in North-West Himalayan region, India [23]. However, β-bourbonene (11.23%), α-humulene (0.1-29.9%) and pulegone (72.1%), were mainly exclusive to **Me9**, **Me11**, **Me14** from Iran [20], Uttarakhand, India [22] and Karnataka, India [25] respectively. In addition, menthone (21.9%) was a chemotype of **Me8**, collected from Serbia [19]. In another part, piperitenone oxide (52.3%) was mainly exclusive to **Me19** from Cuba [30] whereas *cis*- carveol (21.3%) was the major compound of **Me10**, collected from Tamilnadu, India [21].

Table 4: Percentages of major components (≥5%) of essential oil of *M. spicata* growing in other countries

Compounds	**Me8**	**Me9**	**Me10**	**Me11**	**Me12**	**Me 13**	**Me 14**	**Me15**	**Me16**	**Me 17**	**Me18**	**Me 19**	**Me 20**
Limonene	-	11.50	11.3	-	9.57-22.31	10.1	-	-	18.19	18.2	7.59	-	5.3
Menthone	21.9	-	-	-	-	-	18.6	-	-	-	-	-	-
cis-Carveol	-	-	21.3	-	-	-	-	-	-	-	-	-	24.3
Pulegone	-	-	-	-	-	-	72.1	-	-	-	-	-	-
Carvone	69.5	78.76	48.6	15.3-68.5	49.62-76.65	57.1	-	40.12	65.33	65.3	73.29	-	51.7
Piperitenone oxide	-	-	-	24.0-79.2	-	-	-	-	-	-	-	52.3	-
β-Bourbonene	-	11.23	-	-	-	-	-	-	-	-	-	-	-
α-Humulene	-	-	-	0.1-29.9	-	-	-	-	-	-	-	-	-

[a]: Order of elution is given from an apolar column (HP-5 MS).

Compared with essential oils of *Mentha spicata* growing in the Mediterranean area and other countries, the present essential oil is similar, with the main content of 1,8-cineole (17,0%, 14.5%, 6.84-7.76%), to the essential oils of plants grown in Tunisia (**Me1**), Greece (**Me3**) and Turkey (**Me4**), respectively and is similar, with the main presence of limonene (20.8%, 14.3%, 20.1%, 16.42%), to the essential oils of plants collected from Tunisia (**Me1**), Greece (**Me2**), Portugal (**Me6**) and Egypt (**Me7**), respectively.

However, the present essential oil is exclusively characterized by the main presence of *cis*-carvone oxide (44.06%) and *cis*-dihydrocarvone (8.85%) which has not been found in any reported essential oil of *Mentha spicata*.

The transformation the carvone to *cis*-carvone oxide and dihydrocarvone can be explained by the mechanisms illustrated in figure 1. There are three double bonds in carvone capable of reduction; the product of reduction depends on the reagents and conditions used. Catalytic hydrogenation of carvone (**1**) can afford either carvomenthol (**2**) or carvomenthone (**3**). Zinc and acetic acid reduce carvone leading to dihydrocarvone (**4**). Meerwein–Ponndorf–Verley (MPV) reduction using propan-2-ol and aluminium isopropoxide provides carveol (**5**); a combination of sodium borohydride and $CeCl_3$ (Luche reduction) is also effective. A Wolff-Kishner reduction with hydrazine and potassium hydroxide afford limonene (**6**) (Figure 1) [44].

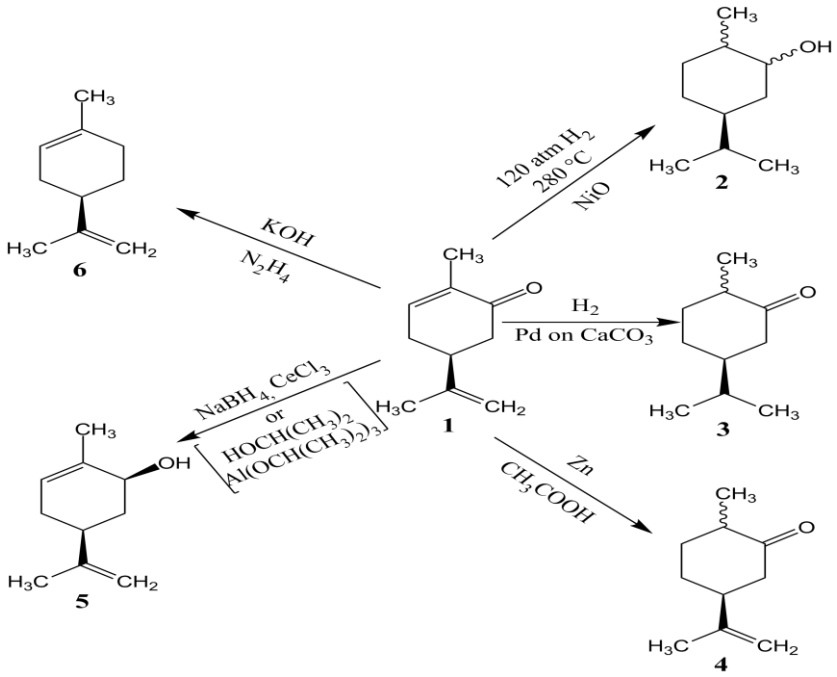

Figure 1. Various chemical reductions of carvone.

Various products can be ontained by the oxidation of carvone. diketone (**7**) is obtained by the oxidation of carvone, in the presence of an alkali such as $Ba(OH)_2$ whereas the epoxide (**8**) is formed by the use of hydrogen peroxide. Carvone may be cleaved using ozone followed by steam, leading to dilactone (**9**), while (**10**) is obtained in the presence of $KMnO_4$ (Figure 2) [44].

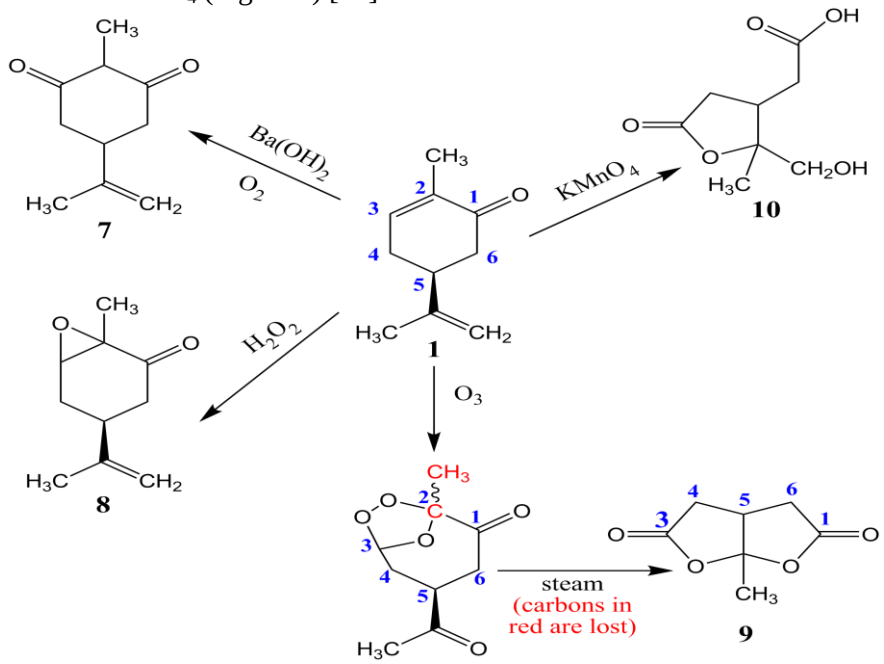

Figure 2. Various oxidations of carvone

3.2. Antioxidant activitiy

Four methods were selected to determine the antioxidant capacity of the essential oil namely; $DPPH^{\bullet}$ and $ABTS^{\bullet+}$ assays for radical-scavenging activity the β-Carotene-linoleic acid assay for lipid peroxidation activity. Since transition metals can accelerate the lipid peroxidation *via* the Fenton reaction, the metal chelating activity was selected to measure the binding capacity of iron of the essential oil. From Table 5, the essential oil exhibited a moderate antioxidant activity in all the tests compared with the used standards and the best

inhibition was observed with the DPPH assay where the oil's activity (IC_{50}: 93.45±1.42 µg/mL), was half lower than the activity of BHA (IC_{50}: 45.37±0.47 µg/mL), used as a standard. This selective activity exhibited in the DPPH assay may be explained by the different mode of action of one or more of the major components (synergic effect) of the tested oil.

Table 5. Antioxidant activities of *Mentha spicata* by the Metal chelating, $DPPH^{*}$, β-Carotene and $ABTS^{\cdot+}$ assays.[a]

Sample	Antioxidant activity			
	IC_{50}[a] (µg/mL)			
	$DPPH^{\cdot}$ assay	$ABTS^{\cdot+}$ assay	β-Carotene-linoleic acid assay	Fe^{2+}-Ferrin assay
M. spicata oil	93.45 ± 1.42	74.19 ± 2.58	42.24 ± 5.21	66.84 ± 0.30
α-tocopherol[b]	7.31±0.17	4.31 ± 0.10	2.10 ± 0.08	NT
BHA [b]	45.37±0.47	4.10 ± 0.06	1.34 ± 0.04	NT
EDTA[b]	NT[c]	NT	NT	6.50 ± 0.07

[a] IC_{50} values represent the means ± standard deviation of three parallel measurements ($p < 0.05$).
[b] Reference compounds: BHA: Butylatedhydroxyl anisole; EDTA: Ethylenediaminetetraacetic acid
[c] not tested.

Dhifi *et al.* [45] reported that the IC_{50} value of the *M. spicata* essential oil harvested from south of Tunisia (chemotype menthone/pulegone) was about 10 µg/mL. Some compounds such as monoterpenes and oxygenated sesquiterpenes are reported to possess antioxidant activity [46] which may explain this interesting antioxidant activity of the presently studied essential of *Mentha spicata*, cultivated at the Alerian Septentrional Sahara, which is found to be rich with 1,8-cineole [12], limonene [12,47], *cis*-carvone oxide [47;48] which have been already reported to possess a moderate to good antioxidant activities.

In the β-carotene/linoleic acid system, the oxidation of linoleic acid generates peroxyl free radicals due to the abstraction of a hydrogen atom from diallylic methylene groups of linoleic acid [49]. The free radical will then oxidize the highly unsaturated β-carotene. The presence of antioxidants in the essential oil will minimize the oxidation of β-carotene by hydroperoxides, formed in this system, which will be neutralized by the antioxidants present in the essential oil. Thus, the degradation rate of β-carotene depends on the antioxidant activity of the essential oil. Oxygenated monoterpenes and monoterpene hydrocarbons are mainly responsible for the antioxidant potential of essential oil [50]. However, we can not attribute the antioxidant effect of a total essential oil only to the major compounds, minor molecules may make significant contributions to the oil activity.

3.3. Antibacterial activity

The antibacterial activity of the essential oil of *M. spicata* against all test microorganisms obtained by agar disk diffusion assay is shown in Table 6. The results of the current research revealed that the essential oil exhibited a moderate antibacterial effect against the tested microorganisms. In general, Gram-negative bacteria were more susceptible to *M. spicata* essential oil than Gram-positive bacteria. As shown in Table 6, *Escherichia coli* ATCC 25922, *Escherichia coli* (HS), *Enterobacter aerogenes*, and *Proteus mirabilis* were the most sensitive with 28 mm, 24 mm, 24 mm and 24 mm inhibition zone diameters, respectively. Accordingly, the essential oil inhibited the growth of most tested bacteria. MICs of the essential oils were also determined by an agar dilution method. The values were ranged from 40-80 µg/mL. From the literature, *M. spicata* essential oil showed an antibacterial effect on the growth of Gram negative and Gram-positive bacteria. Mahboubi and Haghi, reported that the essential oil of *M. spicata* exhibited a high antibacterial activity against *Staphylococcus aureus*, *Listeria monocytogenes*, *Bacillus cereus*, and *Escherichia coli* [51]. Recently, Dhifi *et al.* [45] reported that the *M. spicata* essential oil (chemotype menthone/pulegone) was active against Gram+ (*S. aureus* and *S. epidermidis*) and Gram- (*Salmonella* sp. and *E. coli*) bacteria, with 20 mm inhibition zone diameters against *Salmonella* sp. strain and 18 mm against *Escherichia coli*.

Generally, it is believed that the antibacterial activities of essential oils are likely related to the percentage of terpenes, and aldoketones [52–54]. 1,8-Cineole [12], limonene [12] and *cis*-carvone oxide [47;48] have been already reported to possess antibacterial activities which may explain the good antibacterial activity of the present essential oil which is rich with these three components.

Table 6: Antibacterial activity of the essential oil of *Mentha spicata* (Inhibition zone diameters and MICs: Minimum inhibitory Concentration)

Microorganisms	Inhibition zone (mm)			MIC (µg /mL)		
	Amp[a]	Genta[a]	oil[b]	Amp	Genta	oil
Escherichia coli ATCC 25922[c]	18	22	28	10	10	40
Escherichia coli (HS)[d]	-	-	24	-	-	40
Pseudomonas aeruginosa ATCC 27853[c]	-	12	18	-	-	80
Pseudomonas aeruginosa (HS)[d]	-	-	16	-	5	80
Enterobacter aerogenes (HS)[d]	14	22	24	-	-	40
Klebsiella pneumonia (HS)[d]		-	20	10	5	40
Proteus mirabilis (HS)[d]	30	16	24	-	-	40
Staphylococcus aureus ATCC 43300[c]	-	14	20	5	-	40
Staphylococcus aureus (HS)[d]	-	-	22	-	15	40
Streptococcus sp (HS)[d]	-	-	18	-	-	80

[a]: Control: Amp: Ampicillin (10 µg/mL); Genta: Gentamicin (10 µg/mL)
[b]: *Mentha spicata* oil (128 µg//mL)
[c]: Obtained from the Pasteur Institute (Algiers)
[d]: Clinical isolates from the laboratory of bacteriology (CHU Constantine, Algeria)

In addition, because of its main 1,8-cineole content [55], the present essential oil could be tested as a corrosion inhibitor.

Conclusion

The chemical composition of the hydrodistilled essential oil of *Mentha spicata*, cultivated at Ghardaia (Algerian Septentrional Sahara), was determined by GC and GC/MS. 29 components representing 96.43% of the essential oil were identified with *cis*-carvone oxide, 1,8-cineole, *cis*-dihydrocarvone, and limonene (5.80%) as the main components. Compared with essential oils of *Mentha spicata* growing in the Mediterranean area and other countries, the present essential oil is similar to the essential oils of *Mentha spicata* grown in Tunisia, Greece, and Turkey, with the main presence of 1,8-cineole. However, *cis*-carvone oxide and *cis*-dihydrocarvone are reported here, for the first time, as major components of *Mentha* essential oil.

Generally, the observed differences in chemical composition content of the essential oils of a species could be attributed to several factors including the method used for extraction of the essential oil, geographical conditions, climate and seasonal variations, the stage of the plant growth and processing of plant materials before extraction of the oil.

Due to its main components, the essential oil showed a moderate antioxidant and a good antibacterial activities which suggests to use it in food conservation.

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