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Research Article

Chemical Composition, Antioxidant Potential, and Antibacterial Activity of Essential Oil Cones of Tunisian *Cupressus sempervirens*

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The extraction yield of the essential oil (EO) extracted by hydrodistillation from the cones of Tunisian *Cupressus sempervirens* L. was of 0.518%. The chemical composition was analyzed by GC-MS. Results showed that this essential oil was mainly composed of monoterpene hydrocarbons (65%) with α-pinene as the major constituent (47.51%). Its antioxidant activity was ascertained by evaluating the total antioxidant capacity and also by evaluating its inhibitory effect against DPPH and ABTS radicals. In addition, it showed a strong antioxidant power against the DPPH (IC_{50} = 151 μg/mL) and ABTS (IC_{50} = 176.454 μg/mL) radicals scavenging. Moreover, its antibacterial activity was tested against different species of pathogenic bacteria (three Gram-positive and eight Gram-negative bacteria). The bacterial strains susceptible to the evaluated oil were *Bacillus subtilis*, *Escherichia coli*, *Klebsiella oxytoca*, *Morganella morganii*, *Shigella*, and *Vibrio cholerae*.

1. Introduction

Cupressus (Cupressaceae), comprising twelve species, is distributed in North America, the Mediterranean region, and subtropical Asia at high altitudes [1]. The geographic area of *Cupressus* genus is limited to the northern hemisphere and many species have been studied [2, 3]. In Tunisia, only one species of the genus *Cupressus*, *Cupressus sempervirens* L. [4], was native.

Cupressus sempervirens L. is a medicinal plant. The dried leaves of this plant are used as an emmenagogue and for stomach pain [5] as well as for diabetes [6]. Its dried fruit plant is used for inflammation treatment [7], toothache, and laryngitis [8] and also as a contraceptive [9], astringent, and antiphrastic drug [10]. The dried seed of this tree has been used for wounds, ulcers, bruises, sores, pimples, pustules, skin eruptions, and erysipelas [11]. *Cupressus sempervirens* essential oil

is used externally for headache, colds, cough, and bronchitis [12].

Studies on phytochemical compounds of *Cupressus sempervirens* L. revealed that it contains active constituents such as flavonoids (cupressuflavone, amentoflavone, rutin, quercitrin, quercetin, and myricitrin), phenolic compounds (anthocyanidin, catechins flavones, flavonols and isoflavones, tannins, and catechol), and essential oils (EO) [13, 14]. It has been demonstrated that principals active from *Cupressus sempervirens* L. display antiseptic, aromatherapeutic, astringent, balsamic, and anti-inflammatory activities [15]. *Cupressus sempervirens* L. antimicrobial activity has been reported in several studies [14, 16].

Even studies have focused on chemistry and biological activities of *Cupressus sempervirens* L. leaves originating from different areas in the world [14, 16, 17]; there is no report concerning this species (cones) in Tunisia. The aim of this study is to ascertain the chemical composition of the EO of Tunisian *Cupressus sempervirens* L. cones and to evaluate its antioxidant and antibacterial activities.

2. Materials and Methods

2.1. Plant Material. Cones (the aerial parts) of *Cupressus sempervirens* L. were collected from Sidi Thabet (North of Tunisia) in March 2014. The botanical identification was achieved by Pr. Mohammed Chaeib from the Faculty of Sciences of Sfax-Tunisia. Female cones were dried at room temperature for 7 days and used for analyses. Voucher specimens of the plants were deposited in the Herbarium of this laboratory.

2.2. Extraction of the Essential Oil. *Cupressus sempervirens* cones were dried at room temperature. After that, 100 g of dry matter was used for essential oil extraction by hydrodistillation in a Clevenger apparatus for four hours. The resulting essential oil recovered is dried by anhydrous sodium sulphate and then stored at 4°C for further analysis.

2.3. Analysis of the Essential Oil. *Cupressus sempervirens* L. essential oil composition was investigated by GC and GC/MS. The analytical GC was carried out on an HP5890-series II gas chromatograph (Agilent Technologies, California, USA) equipped with Flame Ionization Detectors (FID) under the following conditions: the fused silica capillary column, apolar HP-5, and polar HP Innowax (30 m × 0.25 mm ID, film thickness of 0.25 mm). The oven temperature was held at 50°C for 1 min, then programmed at rate of 5°C/min to 240°C, and held isothermal for 4 min. The carrier gas was nitrogen at a flow rate of 1.2 mL/min; injector temperature: 250°C; detector: 280°C; the volume injected: 0.1 mL of 1% solution (diluted in hexane). The percentages of the constituents were calculated by electronic integration of FID peak areas without the use of response factor correction. GC/MS was performed in a Hewlett-Packard 5972 MSD System. An HP-5 MS capillary column (30 m × 0.25 mm ID, film thickness of 0.25 mm) was directly coupled to the mass spectrometry. The carrier gas was helium, with a flow rate of 1.2 mL/min. Oven temperature was programmed (50°C for 1 min, then 50–240°C at 5°C/min) and subsequently held isothermal for 4 min; injector port: 250°C; detector: 280°C; split ratio: 1 : 50; volume injected: 0.1 mL of 1% solution (diluted in hexane); mass spectrometer: HP5972 recording at 70 ev scan time: 1.5 s; mass range: 40–300 amu. Software adopted to handle mass spectra and chromatograms was ChemStation. The identification of the compounds was based on mass spectra (compared with Wiley 275.L, 6th edition mass spectral library). Further confirmation was done from Retention Index data generated from a series of alkanes retention indices (relative to C9–C28 on the HP-5 and HP-20M columns) [18].

2.4. Antioxidant Activities

2.4.1. Evaluation of Total Antioxidant Capacity. The total antioxidant capacity is based on the reduction of Mo (VI) to Mo (V) by the oil and the formation of a phosphate subsequent green/Mo (V) complex at acid pH [19]. In a reaction volume of 1 mL was added to different concentrations of tested oil and standard sulfuric acid (0.6 M), sodium phosphate (28 mM), and ammonium molybdate (4 mM). The solutions were then incubated in a water bath at 95°C for 1 hour. After cooling to room temperature, the optical density is measured at 695 nm. Each fraction was analyzed in triplicate.

2.4.2. DPPH Radical-Scavenging Activity. The ability of our essential oil to reduce the DPPH was measured according to the method described by Tuberoso [20]. For each concentration, one milliliter was added to 0.25 mL of ethanolic solution of DPPH. The mixture was stirred vigorously and then incubated at room temperature for 30 min in the dark. The absorbance illustrating the power of the extract to reduce the free radical DPPH to the yellow-colored diphenylpicrylhydrazine was measured at 520 nm. So, antiradical activity is expressed as IC_{50} ($\mu g\,mL^{-1}$), the extract dose required to induce a 50% inhibition. The ability to scavenge the DPPH radical was calculated using the following equation: DPPH scavenging effect = $[(A_0 - A_1)/A_0] \times 100$, where A_0 and A_1 were the absorbance of the control and the sample after 30 min, respectively. Each experiment was analyzed in triplicate.

2.4.3. ABTS Assay. For ABTS assay, we used the method described by Hayouni et al. [21]. The stock solutions included 7 mM $ABTS^{\bullet+}$ and 2.45 mM potassium persulfate solution. The working solution was then prepared by mixing the two stock solutions in equal quantities and allowing them to react for 12 h at room temperature in the dark. The solution was then diluted by mixing 1 mL $ABTS^{\bullet+}$ solution with 50 mL methanol to obtain an absorbance of 0.7 ± 0.02 units at 734 nm using the spectrophotometer 6305 UV-VIS. Fresh $ABTS^{\bullet+}$ solution was prepared for each assay. 50 μL of each concentration of the essential oil is added with 950 μL of $ABTS^{\bullet+}$ solution, allowed to react in the dark and then we follow the kinetics of this mixture every 5 min for 30 min. Then the absorbance was measured at 734 nm. Antiradical activity is expressed as IC_{50} ($\mu g\,mL^{-1}$), the extract dose required to induce a 50% inhibition. A low IC_{50} value corresponds to

a high antioxidant activity of plant extract. Results are expressed in μg Trolox equivalents (TE)/mg dry mass. The percentage inhibition of the ABTS cation radical by the samples was calculated according to the following formula: Inhibition percentage (% inhibition) = $(A_0 - A_t)/(A_0 \times 100)$, where A_0 is absorbance of control sample ($t = 0$ h) and A_t is absorbance of a tested sample in 5 or 30 min.

2.5. Antimicrobial Activities

2.5.1. Microorganisms. The tested microorganisms include Gram-positive bacteria, *Staphylococcus aureus ATCC 29213*, *Bacillus subtilis ATCC 6633*, and *Bacillus cereus ATCC 11778*, and Gram-negative bacteria: *Escherichia coli ATCC 8739*, *Klebsiella oxytoca CECT 8207*, *Salmonella salamae* ATCC *43972*, *Salmonella typhi ATCC 25241*, *Morganella morganii ATCC 25830*, *Salmonella anatum* ATCC *9270*, *Vibrio cholerae CECT 8265*, and *Shigella ATCC 29930*.

2.5.2. Disk-Diffusion Assay. Antibacterial activity was evaluated using the method described by Choi et al. (2006) [22]. The principle of this method is to use Whatman paper discs of 6 mm in diameter. The discs were impregnated with essential oil diluted in hexane. A disc soaked in hexane was used as negative control. These discs are then deposited on the surface of a middle swab with a bacterial suspension to an optical density of 0.5 McFarlend standard. We used the bacterial strains for the culture medium Muller-Hinton. At the end of the incubation, 24 hours at 37°C, the diameters of the zone of inhibition were measured.

2.5.3. Minimum Inhibitory Concentration. A broth microdilution was used to determine the minimum inhibitory concentration (MIC). The tests were performed in Muller-Hinton Broth. The essential oil was dissolved in Tween 80. A serial doubling of tested oil was prepared in a 96-well microtiter plate over the range 78 μg/mL–1 mg/mL. In each well containing a concentration was added 100 μL of bacterial suspension of 10 6 (0.5 MacFarlen standard). We incubated plates overnight at 37°C and then measured the optical density at 620 nm.

3. Results and Discussion

3.1. Chemical Composition. The extraction yield of the EO from the cones of Tunisian *Cupressus sempervirens* L. was of 0.518%. The coupling analysis of GC/MS and GC-FID/KI revealed 67 compounds (Figure 1). The major compound was α-pinene (47.51%). It was followed by δ-3-carene, α-terpinyl acetate, β-caryophyllene, and α-cedrol whose proportions were 7.40, 4.11, 4.53, and 4.99%, respectively (Table 1). These results are in accordance with those reported by Boukhris et al. (2012) [17] for *Cupressus sempervirens* L. collected from the random gardens in Sfax, Tunisia, and characterized by α-pinene (37.14%), δ-3-carene (19.67%), limonene (5.43%), and α-terpinolene (4.69%) as the most abundant volatiles. Furthermore, Riahi et al. (2012) [23] reported a yield of 0.92% for Tunisian *Cupressus sempervirens* L. It is higher than

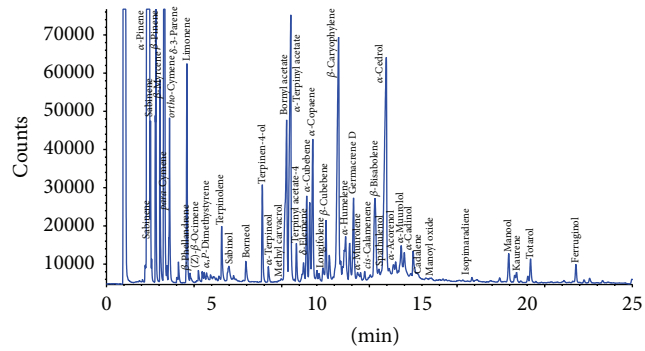

Figure 1: Essential oil chromatogram from cones of *Cupressus sempervirens*.

that obtained for Algerian *C. sempervirens* (0.26%) [14] and comparable to that of the Cameroon species (1%) [24].

The major components of Tunisian *Cupressus sempervirens* L. as reported by Riahi et al. (2012) [23] were α-terpinolene (24.44%), 3-carene (18.60%), α-limonene (11.61%), terpinen-4-ol (10.56%), β-myrcene (4.89%), camphor (4.62%), and β-linalool (4.23%). For essential oils of most *Cupressus* species, α-pinene and 3-carene were cited as the major compounds [25]. However in our sample α-pinene component is absent. Results by Tapondjou et al. (2005) revealed that the oil from *Cupressus sempervirens* L. in Cameroon mainly consists of α-pinene (9.9%), terpinen-4-ol (11.2%), and sabinene (14.8%).

Moreover, α-pinene and γ-terpinene accounted, respectively, for 39.5 and 11.56% of the whole essential oil of *Cupressus sempervirens* L. cones originating from Greece [26]. The cone essential oil of Egyptian *Cupressus sempervirens* L. showed antibacterial activity [27]. It is in contradiction with the results reported by Chéraif et al. (2005) [28]. Indeed, α-pinene is present in the essential oil of *Cupressus sempervirens* L. leaves at a low rate (20%) compared with that of cone essential oil. In addition, the proportion of δ-3-carene is important (22.9%) in the essential oil branches of *Cupressus sempervirens* L. by comparison with that of cones (7.40%). Limonene is of the order of 5.1% in the essential oils branches of *Cupressus sempervirens* L. Its rate is lower in cones essential oil (1.75%). The same results have been reported for α-terpinyl acetate whose rates were of 7.5% and 4.11%, respectively, in branches and cones. Furthermore, β-caryophllene was detected as trace in twigs whereas its rate was of 4.53% in cones. Among volatiles, α-terpinolene was present in the branches essential oil with an amount of 9.4%. However, it was absent in cones essential oil.

Loukis et al. (1991) [26] reported that α-pinene and γ-terpinene accounted, respectively, for 39.5 and 11.56% of the whole essential oil of *Cupressus sempervirens* L. cones originating from Greece.

Emami et al. (2004) [29] detected 42 compounds in the essential oil of cones *Cupressus sempervirens* L. originating from Egypt. They reported a yield of 0.26% for leaves essential oil. Tognolini et al. (2006) [30] reported that the α-pinene is the major compound. This is the second monoterpene hydrocarbon predominant compound according to the results of

Table 1: Essential oil composition from cones of *Cupressus sempervirens*.

Number	Volatile compound	RI[a]	RI[b]	Amount (% of whole EO)	Methods of identification
1	Tricyclene	924	1015	0.14	MS, RI
2	α-Pinene	939	1032	47.51	MS, RI, Co-GLC
3	α-Fenchene	953	1044	0.74	MS, RI
4	Camphene	954	1076	0.15	MS, RI, Co-GLC
5	Sabinene	976	1132	0.6	MS, RI, Co-GLC
6	β-Pinene	980	1118	1.53	MS, RI, Co-GLC
7	β-Myrcene	994	1174	1.28	MS, RI, Co-GLC
8	δ-3-Carene	1011	1159	7.4	MS, RI, Co-GLC
9	α-Terpinene	1018	1188	0.11	MS, RI, Co-GLC
10	p-Cymene	1026	1280	0.2	MS, RI, Co-GLC
11	m-Cymene	1023	1278	1.12	MS, RI, Co-GLC
12	Limonene	1030	1203	1.75	MS, RI, Co-GLC
13	β-Phellandrene	1006	1176	0.13	MS, RI
14	(Z)-b-Ocimene	1040	1246	0.14	MS, RI
15	(E)-b-Ocimene	1050	1266	0.12	MS, RI
16	g-Terpinene	1062	1266	0.12	MS, RI, Co-GLC
17	α,p-Dimethylstyrene	1080	1452	0.15	MS, RI
18	Fenchone	1087	1406	0.13	MS, RI
19	Terpinolene	1088	1290	0.6	MS, RI, Co-GLC
20	Sabinol	1210	1800	0.24	MS, RI
21	Borneol	1165	1719	0.31	MS, RI
22	Terpinen-4-ol	1018	1188	0.97	MS, RI, Co-GLC
23	α-Terpineol	1189	1706	0.17	MS, RI, Co-GLC
24	Carvacrol, methyl ether	1244	1586	0.07	MS, RI, Co-GLC
25	Bornyl acetate	1295	1597	2.75	MS, RI
26	α-Terpinyl acetate	1350	1677	4.11	MS, RI
27	Terpinyl-4 acetate	1333	1709	0.39	MS, RI
28	δ-Elemene	1337	1479	0.24	MS, RI
29	α-Cubebene	1352	1468	0.93	MS, RI
30	α-Ylangene	1372	1493	0.9	MS, RI
31	α-Copaene	1376	1497	1.72	MS, RI, Co-GLC
32	β-Bourbonene	1385	1535	0.15	MS, RI
33	β-Elemene	1391	1600	0.12	MS, RI
34	Longifolene	1413	1575	0.2	MS, RI
35	β-Cubebene	1348	1456	0.69	MS, RI
36	α-Gurjunene	1408	1529	0.37	MS, RI
37	β-Caryophyllene	1415	1612	4.53	MS, RI
38	β-Gurjunene	1432	1612	0.2	MS, RI
39	α-Cedrene	1411	1568	0.14	MS, RI
40	Aromadendrene	1439	1628	0.29	MS, RI
41	α-Humulene	1454	1687	0.64	MS, RI, Co-GLC
42	Alloaromadendrene	1474	1661	0.54	MS, RI
43	Germacrene D	1480	1696	1.14	MS, RI, Co-GLC
44	α-Muurolene	1477	1704	0.15	MS, RI
45	δ-Cadinene	1517	1773	0.11	MS, RI
46	*cis*-Calamenene	1543	1837	0.16	MS, RI
47	Cadina-1,4-diene	1552	1768	0.12	MS, RI
48	β-Bisabolene	1508	1740	1.73	MS, RI
49	Spathulenol	1576	2144	0.21	MS, RI
50	α-Cedrol	1597	2021	4.99	MS, RI, Co-GLC

Table 1: Continued.

Number	Volatile compound	RI[a]	RI[b]	Amount (% of whole EO)	Methods of identification
51	T-Cadinol	1642	2187	0.25	MS, RI
52	α-Acorenol	1630	2163	0.34	MS, RI
53	β-Acorenol	1637	2212	0.31	MS, RI
54	epi-α-Cadinol	1640	2170	0.11	MS, RI
55	α-Muurolol	1642	2209	0.73	MS, RI
56	β-Eudesmol	1650	2257	0.68	MS, RI
57	α-Eudesmol	1641	2216	0.17	MS, RI
58	α-Cadinol	1649	2255	0.55	MS, RI
59	Cadalene	1674	2200	0.09	MS, RI
60	Sandaracopimara-8(14),15-diene	1960	2255	0.04	MS, RI
61	Manoyl oxide	2010	2396	0.07	MS, RI
62	Isopimara-9-(11),15-diene	1906	2175	0.05	MS, RI
63	Abietadiene	2054	2530	0.05	MS, RI
64	Manool	1989	2376	0.38	MS, RI
65	Kaurene	2048	2399	0.13	MS, RI
66	Totarol	2280	2314	0.31	MS, RI
67	Ferruginol	2295	2330	0.23	MS, RI
				Total (%) 97.69	

RI: Retention Index; MS: mass spectrometry; Co-GLC: coinjection.
%: percentage calculated by GC-FID on nonpolar HP-5 capillary column.
[a]Apolar HP-5 MS column.
[b]Polar HP Innowax column.

Sacchetti et al. (2005) [31]. Furthermore, the most abundant volatile compounds in the essential oil of Turkish *Cupressus sempervirens* L. are α-pinene and Δ-3-carene. Obviously, the composition of essential oils is significantly influenced by the organ. It is also influenced by many other factors including environmental factors such as rainfall, sunlight, soil, and climatic conditions and agronomic factors such as the date of harvest and the density of the culture.

3.2. Antioxidant Activities. Considering the many aspects of antioxidants and their reactivity, several tests are applied as antioxidants. Among them, the total antioxidant capacity, DPPH, and ABTS radicals scavenging tests are used to determine the antioxidant power of essential oil.

The chemical complexity of essential oils, often a mixture of dozens of compounds with different functional groups, polarity, and antioxidant activity, may lead to scattered results, depending on the test employed [31].

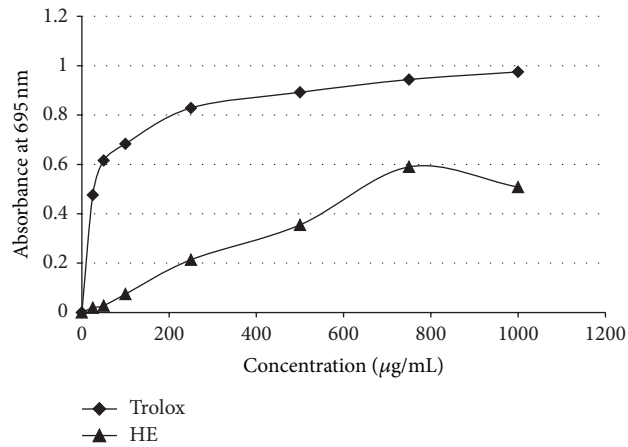

Figure 2: Total antioxidant capacity from cones essential oil of *Cupressus sempervirens*.

3.2.1. Total Antioxidant Capacity. The total antioxidant activity of *Cupressus sempervirens* L. is expressed as Trolox equivalent. The phosphomolybdenum method is based on the reduction of Mo (VI) with Mo (V) by the antioxidant compounds and the formation of a green phosphate/Mo complex (V). Our results showed that the antioxidant activity is proportional to the extract concentration (0.0434 μg ET/g DW) (Figure 2). Baykan Erel et al. (2012) [32] studied the antioxidant activity of essential oils of six species of Artemisia. It was noted that only essential oil of *Artemisia absinthium* and *Artemisia arborescens* has a total antioxidant activity with values, respectively, in the order of 2.89 mg ET/g DW and 3.39 mg ET/g DW.

3.2.2. DPPH Radical-Scavenging Activity. Free radical-scavenging activities of the tested oil and positive control (Trolox) are presented in Figure 3. In fact, *Cupressus sempervirens* L. essential oil remarkably reduced the concentration of DPPH free radical and transformed its stable, purple color into the yellow-colored DPPH-H with an efficiency $IC_{50} = 151$ μg/mL. The effect of antioxidant on DPPH radical scavenging was thought to be due to its hydrogen-donating ability. DPPH

Table 2: Antibacterial activity from essential oil cones of *Cupressus sempervirens*.

Microorganisms	Concentration (mg/mL)								Negative control
	1	0.5	0.25	0.125	0.0625	0.0312	0.0156	0.0078	
Bacillus subtilis ATCC 6633	−	−	−	+	+	+	+	+	+
Escherichia coli ATCC 8739	−	−	−	−	+	+	+	+	+
Klebsiella oxytoca CECT 8207	−	−	−	−	+	+	+	+	+
Morganella morganii ATCC 25830	−	−	+	+	+	+	+	+	+
Shigella ATCC 29930	−	−	−	−	+	+	+	+	+
Vibrio cholerae CECT 8265	−	−	−	−	−	+	+	+	+

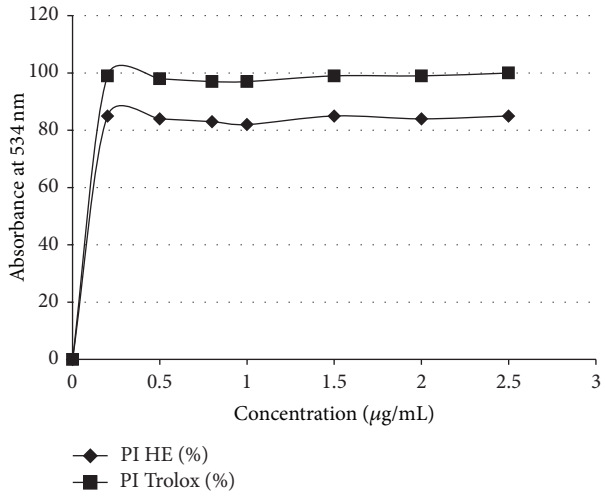

Figure 3: Scavenging of DPPH radical from cones essential oil of *Cupressus sempervirens*.

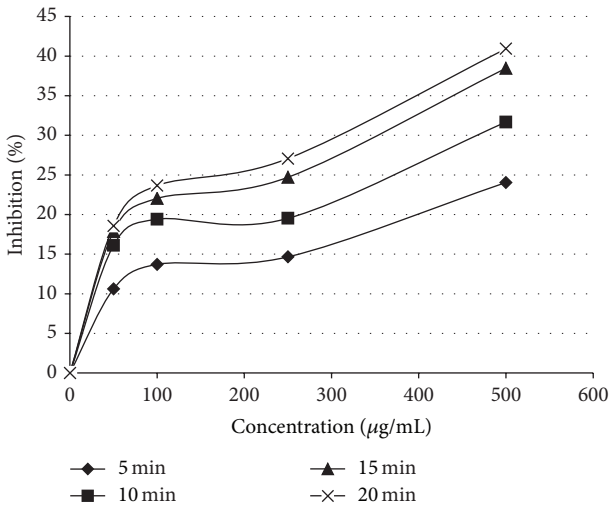

Figure 4: Scavenging of ABTS radical from cones essential oil of *Cupressus sempervirens*.

radical is a stable free radical and accepts an electron or hydrogen radical to become a stable diamagnetic molecule [33].

In two antioxidants screening studies *Cupressus sempervirens* L. was reported to display moderate radical-scavenging effect against DPPH [31, 34]. The leaf methanolic extract of Egyptian *Cupressus sempervirens* L. had strong DPPH radical-scavenging activity as reported by Ibrahim et al. (2009) [35].

Needless to say, potency of antioxidant activity of a substance is correlated with the applied method. In a study on some Iranian conifers the leaf and fruit MeOH extracts of *Cupressus sempervirens* L. were highly effective in ferric thiocyanate (FTC) and thiobarbituric acid (TBA) methods [36]. Nevertheless, activity of these extracts was also changeable according to the method.

3.2.3. ABTS Assay. Essential oil of *Cupressus sempervirens* L. had an antioxidant according to the DPPH test. The results relative to ABTS radical cation scavenging confirmed the previous result. In fact, IC_{50} obtained at the end of this test is 176.454 μg/mL (Figure 4). Ghazghazi et al. (2010) [37] obtained, by studying the essential oil samples of *R. canina* of Feija and Ain Draham (North of Tunisia) during the test of the ABTS, IC_{50} values of 159.0 μg/mL and 201.8 μg/mL, respectively. From these results we can conclude that the essential oil of *Cupressus sempervirens* L. has a strong antioxidant against ABTS cation. The antioxidant activity of essential oil could be assigned to the synergistic effects of two or more of its components. In this context, Lu and Foo (2001) [38] reported that most natural antioxidant compounds often work synergistically to produce a broad spectrum of antioxidant properties that create an effective defense system against free radicals. *Cupressus sempervirens* L. essential oil consists of a very complex mixture of various chemical classes (Table 1), which may produce either synergistic or antagonistic effects on the process of lipid oxidation [39].

3.3. Antibacterial Activity. The antibacterial test allowed us to determine the sensitive strains to the essential oil of *Cupressus sempervirens* L. The screening of this activity was firstly determined by the disk-diffusion method on agar for predicting the inhibitory activity of the oil on the growth of a bacterial culture. Our results showed that the evaluated oil had significant antibacterial effect. In fact, among twelve tested strains, six were sensitive to this oil (Table 2). Gram-negative bacteria (*Klebsiella oxytoca*, *Vibrio cholerae*, *Shigella*, and *E. coli*) were the most sensitive strains with the MIC values that do not exceed 125 μg/mL of essential oil (Table 2). This finding was in agreement with other findings. For example, the cone EO of Egyptian *Cupressus sempervirens* L. showed

antibacterial activity [27]. Chéraif et al. (2005) [28] tested the antibacterial activity of essential oil *Cupressus sempervirens* L. on Gram-positive and Gram-negative bacteria and found that this essential oil has a moderate activity against tested bacteria. It is important to mention that the activity was more pronounced for Gram-positive than Gram-negative bacteria. Similarly, Mazari et al. (2010) [14], when studying the biological activities of essential oils of *Cupressus sempervirens* L. and *Juniperus phoenicea*, reported that both oils constitute sources of antimicrobial agents. There are often large variations in the intensity of the antimicrobial activities against Gram-negative and Gram-positive bacteria.

The EO of *Cupressus sempervirens* L. presented antibacterial activity against Gram-positive and Gram-negative bacteria, showing the biggest inhibition with *B. subtilis* and *E. coli*. Additionally, the cypress essential oil was found to have moderate antimicrobial activity when compared to vancomycin (30 mcg) and erythromycin (15 mcg) as antibiotics [40].

The antibacterial activity could be affected by the solubility of the oil, the diffusion range in the agar and the evaporation [41, 42]. In addition, the antibacterial activities of the essential oils suggest their usefulness in the treatment of various infectious diseases caused by the tested bacteria.

It is well known that Gram-negative bacteria are more resistant to essential oil compound antibacterial properties than Gram-positive bacteria because of hydrophobic lipopolysaccharide in the outer membrane which provides protection against different agents [43]; however, the obtained results indicated that the evaluated oil possesses selective antibacterial activity and its effect was pronounced against Gram-negative bacteria compared to Gram-positive ones.

4. Conclusion

According to our results, the essential oil of cypress cones is very rich in α-pinene which is its major component. This compound is followed by Δ-3-carene, α-terpinyl acetate, β-caryophyllene, and cedrol. This composition is different from those found in other studies, and this difference may be due to climatic factors.

Furthermore, the evaluation of the antioxidant activity by chemical tests (total antioxidant capacity, DPPH, and ABTS) revealed a variable behavior of the essential oil against the used radicals. Moreover, IC_{50} values calculated are very low such that this reflects the high antioxidant power: they are of 176.45 and 151 μg/mL for ABTS and DPPH tests, respectively. The evaluation of the antibacterial activity of the tested essential oil showed that it possesses a significant activity. These promising results allow us to expand our studies to ascertain other activities of this essential oil such as antiparasitic, antifungal, and anticholinesterase activities.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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