

# Chemical Composition and Biological Activities of Essential Oils of *Phagnalon sordidum* (L.) Rchb. (Asteraceae) from Algeria

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## Summary

The essential oil constituents of aerial parts of *Phagnalon sordidum* (L.) Rchb. were analyzed by a combination of capillary gas chromatographic-flame ionization detector (GC-FID) and chromatography-mass spectrometry (GC-MS). A total of 125 constituents comprising 97.6 % of the total oil were identified. The volatile fraction was characterized by monoterpene hydrocarbons (51.4 %), oxygenated monoterpenes (10.4 %), sesquiterpene hydrocarbons (18.0 %), oxygenated sesquiterpenes (6.0 %) and non-terpenic components (11.8 %). The predominant constituents identified were  $\beta$ -pinene (26.0 %), (E)- $\beta$ -caryophyllene (10.0 %), limonene (8.5 %), myrcene (4.7 %), decanal (4.5 %), thymol (3.9 %), germacrene-D (3.8 %), and *p*-cymene (3.4 %). The antimicrobial activities of the essential oil evaluated against eleven bacteria and *Staphylococcus aureus*, *Klebsiella pneumoniae* and *Salmonella typhimurium* were the most susceptible microorganisms with a minimum inhibitory concentrations (MICs) 0.01, 0.04, 0.04 mg/mL respectively. Additionally, the essential oil showed moderate radical scavenging and electron donating activity.

## Key words

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*Phagnalon sordidum*, Essential oil, GC-MS, Antimicrobial activity, Antioxidant activity

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## Introduction

The genus *Phagnalon* belongs to the tribe *Gnaphalieae* (Asteraceae) and is distributed from Macaronesia in the west to the Himalayas in the east, from Southern Europe in the north to Ethiopia in the south; it comprises about 36 species (Qaiser and Abid, 2003). *Phagnalon* species are suffruticose shrubs or subshrubs and grow in a variety of habitats ranging from rocky crevices in high mountains to sandy soils in coastal plains (Montes Moreno et al., 2013). Many hybrids have been described (Quézel and Santa, 1963) to possess genetic stability, which reinforces their taxonomic values.

In Algeria this genus includes four species: *Phagnalon sordidum* (L.) Rchb., *Phagnalon garamantum* Maire, *Phagnalon saxatile* (L.) Cass., *Phagnalon rupestre* (L.) DC. (Quézel and Santa, 1963). The *Phagnalon* species are known locally as “*Foddia*” or “*Arfedj*”, and have been used in folk medicine for treating asthma, headache and as anesthetic for toothache (Ali-Shtayeh et al., 1998). The literature logs their antimicrobial and skin clearing activities, anti-allergic, anti-inflammatory and anticholinesterase properties (Ali-Shtayeh et al., 1998; Conforti and Rigano, 2010; Erdogan Orhan et al., 2013; Góngora et al., 2002; Hausen and Schulz, 1977; Olmos et al., 2005).

Algeria with its large surface and therefore its immense biodiversity provides a great potential for the discovery of new flavors and products. However, there is still a lack of chemical and biological activities information associated with plant species due to modest support in research work in this field.

This study aims to evaluate the chemical composition and *in vitro* antimicrobial and antioxidant activities of the essential oil of *Phagnalon sordidum* from Algeria.

## Material and Methods

### Collection of Plant Material

The specimens of *Phagnalon sordidum* were collected at the flowering stage (April to May 2012) from ten locations (PS1–10, PS1: Ain Douze, PS2: Mansourah 1, PS3: Oucheba 1, PS4: Dzarifette1, PS5: Mansourah 2, PS6: *Misserghin* 1, PS7: Oucheba 2, PS8: *Misserghin* 2, PS9: Ain Fetah, PS10: Dzarifette 2) in the north west of Algeria. The plants collected were identified by Prof. Noury Benabadi of the University of Tlemcen. A voucher specimen was deposited in the Laboratory of Ecology and Ecosystem Management, University of Tlemcen (Algeria). Plant samples were dried in the shade and conserved for future use.

### Obtaining Essential Oil

Essential oils were obtained from dried aerial parts (400 g of plant per sample) by hydrodistillation for 5 h, using a *Clevenger*-type apparatus according to the method recommended in the *European Pharmacopoeia* (1996), yielding 0.02–0.06 % of yellow essential oils. The oils were dried over anhydrous sodium sulfate and then stored in sealed glass vials at +4 °C prior to analysis.

### Oil Fractionation

A mixture of all oil samples (total oil: 550 mg) was submitted to flash chromatography [FC; silica gel 200–500 µm, elution

with *n*-hexane, then with diisopropyl ether]. Three fractions were obtained. An apolar fraction (FI: 400 mg) which contains hydrocarbon compounds was obtained by elution with *n*-hexane and two polar fractions containing oxygenated compounds (FII: 105 mg and FIII: 35 mg) were obtained by increasing elution of a mixture of *n*-hexane and diisopropyl ether. The yields of *Phagnalon sordidum* fractions FI, FII and FIII, expressed as a function of the total oil yield, were 72.7 %, 19.1 % and 6.4 % (w/w), respectively. The three fractions were subjected to analytical sequence GC and GC-MS.

### Gas Chromatography Analysis (GC)

GC analyses were carried out using a Perkin-Elmer (Waltham, MA, USA) Autosystem XL GC apparatus equipped with a dual flame ionization detection system and a fused-silica capillary columns (60 m x 0.22 mm I.D., film thickness 0.25 µm), Rtx-1 (polydimethylsiloxane). The oven temperature was programmed from 60 °C to 230 °C at 2 °C/min and then held isothermally at 230 °C for 35 min. Injector and detector temperatures were maintained at 280 °C. Samples were injected in the split mode (1/50), using helium as the carrier gas (1 mL/min); the injection volume was 0.2 µL. Retention indices (RI) of the compounds were determined relative to the retention times of the series of *n*-alkanes (C<sub>5</sub>–C<sub>30</sub>) with linear interpolation, using the Van den Dool and Kratz equation (1963) and software from Perkin-Elmer. Component relative concentrations were calculated based on GC peak areas without using correction factors.

### Gas Chromatography-Mass Spectrometry Analysis (GC-MS)

Samples were analyzed with a Perkin-Elmer Turbo mass detector (quadruple), coupled to a Perkin-Elmer Autosystem XL, equipped with the fused-silica capillary columns Rtx-1 (ion source temperature 150 °C; energy ionization 70 eV). EI mass spectra were acquired over the mass range 35–350 Da (scan time: 1 s). Other GC conditions were the same as described under GC except split 1/80.

### Component Identification

Identification of individual components was based (i) on comparison of calculated RI, on polar and apolar columns, with those of authentic compounds or literature data (König et al., 2001; NIST, 2008); and (ii) on computer matching with commercial mass spectral libraries (Adams, 2001; König et al., 2001; NIST, 1999) and comparison of mass spectra with those of our own library of authentic compounds or literature data (Adams, 2001; König et al., 2001).

### Antimicrobial Assay

The microbiological activity of essential oils was studied in the Microbiology Laboratory of the Faculty of Sciences, University of Tlemcen. It used eleven pathogenic bacteria, four Gram-positives: *Staphylococcus aureus* (ATCC 25923), *Bacillus cereus* (ATCC 10876), *Listeria monocytogenes* (ATCC 15313) and *Enterococcus faecalis* (ATCC 49452), and seven Gram-negatives: *Klebsiella pneumoniae* (ATCC 70063), *Salmonella typhimurium* (ATCC 13311), *Pseudomonas aeruginosa* (ATCC 27853), *Escherichia coli*

(ATCC 25922), *Citrobacter freundii* (ATCC 8090), *Proteus mirabilis* (ATCC 35659) and *Acinetobacter baumannii* (ATCC 19606), being performed according to the rules and procedures of Clinical and Laboratory Standards Institute (CLSI, 2006). The technique of the dilution agar method was used to test the antimicrobial activity and the minimum inhibitory concentration (MIC) was determined as the lowest concentration with no turbidity. Gentamicin was used as positive antibiotic control and DMSO was used as negative control.

### Dilution-agar Method

A dilution agar method was used to determine the MIC<sub>5</sub>. Stock solutions were obtained by dissolving total essential oil in dimethylsulfoxide (DMSO 1%). Serial dilutions of the total oil were made to obtain concentrations ranging from 0 to 5mg/mL. Each mixture was added to Mueller-Hinton agar (Lennette and Balows, 1985; Cowan, 1999). The Petri dishes contained a sterile solution of DMSO and the culture medium, respectively, after incubation at 37 °C for 24h. The experiments were performed in triplicate.

### Antioxidant Activity

#### DPPH Assay

The free radical scavenging activity of essential oils depends on the ability of the antioxidant compounds to donate their hydrogen atoms. The hydrogen-donating abilities of essential oils were examined on the basis of the method described by Molyneux (2004) with some modifications. Used as reagent, 2,2-diphenyl-1-picrylhydrazyl hydrate (DPPH) obviously offers a convenient and accurate method for titrating the oxidizable groups of natural antioxidants. About 1 mL of DPPH solution (0.005 %) in methanol was thoroughly mixed with an equal volume of test extracts at various concentrations (40 to 200 µg/mL) and kept in the dark for 60 min. The absorbance was read at 520 nm using methanol as blank. 1 mL of DPPH solution mixed with 1 mL of methanol was used as control. The inhibition of DPPH radical was calculated using the following equation:

$$\text{DPPH scavenging effect (\%)} = ((A_0 - A_s)/A_0) \times 100$$

where:

A<sub>0</sub>: absorbance of the control (containing all reagents except the test compound),

A<sub>s</sub>: absorbance of the tested sample.

The actual decrease in absorbance induced by the tested sample (change of colour from deep-violet to light yellow) was compared to that of the positive control ascorbic acid. The IC<sub>50</sub> value represented the concentration of extract that causes 50% inhibition was determined. Experiments were carried out in triplicate and the mean value was recorded.

#### FRAP Assay

The total antioxidant potential of the essential oil was carried out according to the method of Benzie and Strain (1996) with some modifications. The *Ferric Reducing Antioxidant Power* (FRAP) assay measures the change in absorbance at 593 nm

owing to the formation of a blue colored Fe(II)-tripirydyltriazine compound from the colorless oxidized Fe(III) form by the action of electron donating antioxidants. 1 mL of the sample at various concentrations (100, 250 and 750 µg/mL) was mixed with phosphate buffer (2.5 mL, 0.2 M, pH 6.6) and potassium ferricyanide (2.5 mL, 1 % (v/w), K<sub>3</sub>Fe(CN)<sub>6</sub>), and the mixture was incubated at 50 °C for 20 min. Next, 2.5 mL of trichloroacetic acid (10 % (w/v)) was added to stop the reaction. After centrifugation at 3000 rpm for 10 min, the supernatant (2.5 mL) was mixed with distilled water (2.5 mL) and ferricchloride (0.5 mL, 1 % (w/v), FeCl<sub>3</sub>·6H<sub>2</sub>O), and the absorbance was measured at 700 nm using water as blank. Ascorbic acid was used as a standard. A stronger absorbance will indicate increased reducing power (Karagözler et al., 2008).

### Statistical Analysis

All the measurements were replicated three times for each assay and the results are presented as mean ± SD. Data was analyzed using PAST3.03 program.

### Results

#### Yields and Chemical Compositions of *Phagnalon sordidum* Essential Oils

Aerial parts of *Phagnalon sordidum* growing wild in west northern Algeria was analyzed for its essential oil composition. The essential oils of *Phagnalon sordidum* gathered from different stations were isolated by hydrodistillation and analyzed by GC-MS. The oil yields, on a dry weight basis, ranged between 0.02 and 0.06 % (w/w). All components were identified by comparison of their EI-MS and GC-retention indices with those of our laboratory-produced 'Arômes' library, with the exception of six components that were identified by comparison with spectral data and retention indices from the literature. The composition of the obtained essential oils from *Phagnalon sordidum* aerial parts are presented in Table 1. The components are listed in order of their elution on the column. Overall, one hundred twenty-five components were characterized, representing 97.6 % of the total oil.

Data analysis of the chemical composition revealed five main classes of components: seventeen monoterpene hydrocarbons, thirty-one oxygenated monoterpenes, fifteen sesquiterpene hydrocarbons, fifteen oxygenated sesquiterpenes and forty six non-terpenic components. Diterpene is present in small amount. Monoterpene hydrocarbons were the most abundant fraction in the investigated oil (51.4 %), followed by sesquiterpene hydrocarbon (18.0 %) (Table 1). Non-terpenic components and oxygenated monoterpenes were present in much smaller amounts (11.8 and 10.4 %, respectively) as well as the oxygenated sesquiterpenes (6.0 %). The analysis of the total oil showed the presence of two main monoterpene hydrocarbons in the sample from *Phagnalon sordidum*: β-pinene (26.0 %), limonene (8.5 %). Additionally, myrcene and p-cymene were identified with 4.7 % and 3.4 %, respectively, while thymol was the most abundant oxygenated monoterpenes (3.9 %). Concerning the sesquiterpene fraction, (E)-β-caryophyllene (10.0 %), germacrene D (3.8 %) and caryophyllene oxide (3.0 %) were the main components.

Table 1. Chemical composition of *Phagnalon sordidum* oils.

N°	Compounds <sup>a</sup>	RI <sub>lit</sub> <sup>b</sup>	RI <sub>a</sub> <sup>c</sup>	RI <sub>p</sub> <sup>d</sup>	Total Oil <sup>e</sup>	FI	FII	FIII	PS1	PS2	PS3	PS4	PS5	PS6	PS7	PS8	PS9	PS10	Min	Max	Identification <sup>f</sup>	
1	Hexanal	780	775	1076	0.1	-	0.1	-	tr	0.1	0.1	0.2	0.1	tr	0.1	0.1	0.1	0.2	0.1	0.2	RI, MS	
2	Octane	800	800	800	tr	0.1	-	-	tr	-	tr	tr	-	-	-	-	-	tr	0.1	-	RI, MS	
3	(E)-2-Hexenal	832	832	1213	0.1	-	0.1	-	0.1	tr	0.3	tr	tr	tr	0.1	0.1	0.1	0.2	0.1	0.3	RI, MS	
4	(Z)-3-Hexen-1-ol	851	844	1342	0.5	-	-	1.3	0.3	0.7	3.6	0.8	-	tr	0.9	0.1	0.5	0.4	0.1	3.6	RI, MS	
5	1-Hexanol	855	849	1340	0.2	-	-	0.8	0.1	0.1	0.6	0.2	tr	-	0.1	0.1	0.1	0.2	0.1	0.6	RI, MS Ref	
6	Heptanal	882	877	1179	0.1	-	0.1	-	tr	0.1	tr	tr	tr	0.1	0.1	tr	-	0.1	0.1	0.1	RI, MS	
7	Nonane	900	900	900	0.1	0.1	-	-	tr	0.1	0.2	0.1	0.1	0.1	0.2	0.1	0.1	0.2	0.1	0.2	RI, MS	
8	$\alpha$ -Thujene	932	926	1023	0.1	0.2	-	-	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.2	0.1	0.1	0.1	0.2	RI, MS	
9	$\alpha$ -Pinene	936	930	1024	2.1	2.2	-	-	0.1	1.2	1.6	1.6	2.1	2.1	1.4	2.7	1.2	2.9	0.1	2.9	RI, MS	
10	(E)-2-Heptenal	931	932	1340	tr	-	0.1	-	Tr	-	-	tr	-	-	-	-	-	tr	-	-	RI, MS	
11	$\alpha$ -Fenchene	941	941	1047	tr	0.1	-	-	0.1	-	0.1	tr	tr	tr	0.1	tr	tr	tr	0.1	0.1	0.1	RI, MS
12	Camphene	950	942	1068	0.1	0.1	-	-	tr	tr	-	0.1	-	-	-	0.1	0.1	0.2	0.1	0.2	RI, MS	
13	Sabinene	973	962	1121	2.5	3.2	-	-	1.5	1.9	0.1	2.1	3.2	3.1	1.8	3.6	1.9	3	0.1	3.6	RI, MS	
14	$\beta$ -Pinene	978	969	1113	26	23.8	-	-	16.7	18.3	20.6	20.1	24.3	24	21	25.7	19.6	27	16.7	27	RI, MS	
15	6-methyl-5-hepten-2-one	978	974	1330	0.2	-	0.2	-	tr	0.2	0.1	0.2	0.2	0.2	-	0.3	-	0.3	0.1	0.3	RI, MS	
16	Myrcene	987	980	1162	4.7	6.8	-	-	4	3.6	4.2	4.4	6.1	6	3.5	7.3	4.8	6.3	3.5	7.3	RI, MS	
17	Octanal	980	981	1284	0.1	-	0.7	-	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.2	0.2	0.1	0.1	0.2	RI, MS	
18	(Z)-Hex-3-enyl acetate	987	987	1311	tr	-	0.1	-	0.1	tr	-	-	-	-	-	-	-	-	0.1	0.1	RI, MS	
19	$\alpha$ -Phellandrene	1002	994	1167	1.5	2.3	-	-	2.3	0.7	1	2.8	2.8	2.8	1.3	2.4	0.8	2.4	0.7	2.8	RI, MS	
20	Decane	1000	999	1000	0.2	0.1	-	-	tr	tr	0.1	tr	tr	0.1	0.1	0.1	0.1	-	0.1	0.1	RI, MS	
21	$\alpha$ -Terpinene	1013	1005	1178	0.2	0.3	-	-	0.3	0.1	0.3	0.2	0.2	0.2	0.1	0.2	0.2	0.2	0.1	0.3	RI, MS	
22	<i>p</i> -Cymene	1015	1009	1269	3.4	5.6	-	-	3.4	2.3	1.9	4	3	3	2.2	3.6	2.8	4.3	1.9	4.3	RI, MS	
23	Phenylacetaldehyde	1012	1010	1607	tr	-	0.1	-	tr	-	-	-	-	-	-	-	-	-	-	-	RI, MS	
24	Limone	1025	1019	1203	8.5	13.7	-	-	8.3	7.1	7.1	8.2	10	9.8	6.6	12.1	9.9	10.6	6.6	12.1	RI, MS	
25	$\beta$ -Phellandrene	1023	1021	1211	1.1	1.2	-	-	1.1	0.8	0.9	0.8	1.3	1.3	0.8	1.2	0.8	1.1	0.8	1.3	RI, MS	
26	1,8-Cineole	1024	1022	1199	tr	-	0.1	-	tr	tr	-	tr	-	-	-	-	-	0.1	0.1	0.1	RI, MS	
27	(Z)- $\beta$ -Ocimene	1029	1027	1231	0.2	0.3	-	-	tr	tr	tr	0.3	tr	tr	0.2	0.1	tr	0.2	0.1	0.3	RI, MS	
28	(E)-2-Octenal	1034	1032	1421	0.2	-	0.5	-	0.2	0.5	0.1	0.2	tr	tr	0.2	0.2	0.2	0.2	0.1	0.5	RI, MS	
29	(E)- $\beta$ -Ocimene	1041	1034	1248	0.1	0.2	-	-	0.3	0.2	0.2	0.2	0.3	0.3	0.1	0.4	0.2	0.2	0.1	0.4	RI, MS	
30	(E)-Sabinene hydrate	1053	1047	1457	0.4	-	-	1.7	0.1	0.1	-	0.1	0.1	tr	-	0.1	0.2	-	0.1	0.2	RI, MS	
31	$\gamma$ -Terpinene	1051	1048	1244	0.1	0.6	-	-	0.6	0.2	0.4	0.4	0.3	0.3	0.4	0.4	0.5	0.4	0.2	0.6	RI, MS	
32	(E)-Linalool oxide THF	1058	1057	1429	tr	-	-	0.3	0.1	0.2	0.1	tr	tr	0.1	0.2	0.1	0.1	0.1	0.1	0.2	RI, MS	
33	<i>p</i> -Cymene	1075	1072	1430	0.1	0.1	-	-	0.1	tr	0.1	tr	-	tr	-	0.1	0.1	0.1	0.1	0.1	RI, MS	

N°	Compounds <sup>a</sup>	RI <sub>lit</sub> <sup>b</sup>	RI <sub>a</sub> <sup>c</sup>	RI <sub>p</sub> <sup>d</sup>	Total Oil <sup>e</sup>	FI	FII	FIII	PS1	PS2	PS3	PS4	PS5	PS6	PS7	PS8	PS9	PS10	Min	Max	Identification <sup>f</sup>
34	Terpinolene	1082	1075	1282	0.7	1.2	–	–	1.2	0.7	0.8	0.8	1	–	0.7	0.6	1	0.7	0.6	1.2	RI, MS
35	Nonanal	1086	1078	1387	1.4	–	4.4	1	1.3	1.7	0.9	1.4	0.5	0.5	1.7	0.6	1.9	1.3	0.5	1.9	RI, MS
36	Linalool	1087	1079	1541	1	–	3.1	2	0.3	0.6	1.7	0.8	0.2	0.2	0.6	0.6	1	1.4	0.2	1.7	RI, MS
37	(Z)-p-Menth-2-en-1-ol	1108	1110	1561	tr	–	–	0.3	0.2	0.2	0.3	0.2	0.1	0.1	0.2	0.1	0.2	0.1	0.1	0.3	RI, MS
38	(Z)-p-Menth-2,8-dien-1-ol	1125	1118	1669	tr	–	–	0.1	tr	0.1	tr	0.1	tr	tr	–	–	–	0.3	0.1	0.3	RI, MS
39	Camphor	1123	1120	1496	0.3	–	3.3	–	0.3	0.2	0.3	0.3	0.2	0.2	0.2	0.2	0.4	0.2	0.2	0.4	RI, MS
40	(E)-Pinocarveol	1126	1126	1646	tr	–	0.3	–	0.1	0.1	–	0.1	–	0.1	0.2	0.1	0.1	0.2	0.1	0.2	RI, MS
41	(Z)-Verbenol	1132	1130	1618	0.2	–	–	0.4	tr	0.1	0.1	0.2	0.1	0.1	–	0.2	0.2	–	0.1	0.2	RI, MS
42	(E)-2-Nonenal	1139	1136	1527	tr	–	0.6	–	0.3	0.2	0.3	0.2	0.1	0.1	0.1	0.1	0.2	0.1	0.1	0.3	RI, MS
43	Pinocarvone	1137	1140	1551	0.3	–	0.9	–	0.1	–	–	–	0.1	0.1	–	–	0.2	–	0.1	0.2	RI, MS
44	Umbellulone	1151	1152	1612	0.1	–	0.2	–	0.1	tr	0.1	–	0.1	–	0.1	–	tr	0.2	0.1	0.2	RI, MS
45	Cryptone	1160	1154	1667	0.2	–	0.1	–	tr	0.2	0.1	0.2	tr	0.1	0.2	0.1	0.1	0.1	0.1	0.2	RI, MS
46	Terpinen-4-ol	1164	1158	1595	1	–	3.1	4.9	1.3	0.7	1	1.1	0.6	0.6	0.8	1	1.2	1.3	0.6	1.3	RI, MS
47	p-Cymen-8-ol	1169	1162	1834	tr	–	–	0.1	tr	–	tr	tr	–	–	–	–	tr	–	–	–	RI, MS
48	Myrtenal	1172	1669	1620	0.3	–	1	–	0.2	0.1	0.2	0.2	0.3	0.1	0.1	0.2	0.3	0.2	0.1	0.3	RI, MS
49	Myrtenol	1178	1170	1781	0.6	–	–	1.7	0.6	0.5	0.7	0.7	0.1	0.3	0.6	0.5	0.8	0.9	0.1	0.9	RI, MS
50	Safranal	1182	1171	1627	tr	–	0.1	–	0.2	–	–	–	–	–	–	–	tr	tr	0.2	0.2	RI, MS
51	α-Terpineol	1176	1176	1690	0.3	–	–	10.9	0.1	0.2	0.3	0.3	0.1	–	0.2	0.2	0.4	0.3	0.1	0.4	RI, MS
52	Decanal	1180	1183	1497	4.5	–	15	–	3.2	6.4	5.9	5.3	2.9	2.9	8.1	2.4	6.8	2.9	2.4	8.1	RI, MS
53	β-Cyclocitral	1195	1196	1601	0.1	–	0.4	–	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.2	RI, MS
54	Dodecane	1200	1200	1200	0.1	0.1	–	–	0.1	0.1	0.1	0.1	0.1	0.1	–	–	0.1	–	0.1	0.1	RI, MS
55	Nerol	1210	1212	1790	0.3	–	–	1.6	tr	tr	–	–	–	tr	0.1	–	0.1	–	0.1	0.1	RI, MS
56	Thymyl methyl oxide	1215	1213	1581	0.1	–	–	–	tr	0.1	–	0.1	0.1	0.1	0.2	0.1	0.2	0.1	0.1	0.2	RI, MS
57	Cuminaldehyde	1215	1214	1765	tr	–	0.2	–	0.1	0.3	0.2	0.2	0.1	tr	0.2	0.1	tr	0.2	0.1	0.3	RI, MS
58	(Z)-hex-3-enyl 2-methylbutyrate	1220	1215	1460	tr	–	0.4	–	tr	0.1	0.2	0.2	–	–	–	0.2	0.2	0.3	0.1	0.3	RI, MS
59	Geraniol	1235	1234	1838	0.4	–	–	5	tr	0.2	0.1	0.2	tr	0.1	0.2	0.2	0.1	0.2	0.1	0.2	RI, MS
60	(E)-2-decenal	1240	1236	1647	0.1	–	0.7	–	tr	tr	–	–	tr	tr	0.2	tr	0.2	tr	0.2	0.2	RI, MS
61	Thymol	1267	1269	2167	3.9	–	–	–	7.3	7	11	2.5	4.5	4.5	6.8	1.8	3.1	1.3	1.3	11	RI, MS
62	Bornyl acetate	1270	1271	1545	tr	–	–	–	0.1	tr	–	0.2	–	0.1	–	0.1	–	0.2	0.1	0.2	RI, MS
63	2-Undecanone	1273	1272	1595	0.3	–	–	–	tr	0.1	0.4	0.1	0.1	–	0.5	0.2	0.5	–	0.1	0.5	RI, MS
64	(E,E)-2,4-Decadienal	1291	1288	1820	0.1	–	1	–	0.2	0.3	0.1	0.5	0.1	0.1	0.2	0.3	0.1	0.1	0.1	0.5	RI, MS
65	(Z)-3-Hexenyl tiglate	1291	1295	1653	0.4	–	1.5	–	0.3	0.4	0.2	0.7	0.2	0.2	0.5	0.3	0.7	0.4	0.2	0.7	RI, MS
66	Tridecane	1300	1298	1300	0.1	0.1	–	–	0.1	tr	0.1	0.1	–	0.1	0.1	0.1	0.1	0.1	0.1	0.1	RI, MS
67	neo-Dihydro carveol acetate	1338	1336	1715	tr	–	0.3	–	0.1	0.1	–	0.2	0.1	0.1	–	0.1	0.2	tr	0.1	0.2	RI, MS

N°	Compounds <sup>a</sup>	RI <sub>lit</sub> <sup>b</sup>	RI <sub>a</sub> <sup>c</sup>	RI <sub>p</sub> <sup>d</sup>	Total Oil <sup>e</sup>	FI	FII	FIII	PS1	PS2	PS3	PS4	PS5	PS6	PS7	PS8	PS9	PS10	Min	Max	Identification <sup>f</sup>	
68	(E)-2-Undecenal	1345	1338	1702	0.2	-	0.3	-	tr	-	-	tr	-	-	-	-	-	0.2	0.2	0.2	RI, MS	
69	Neryl acetate	1342	1341	1718	0.1	-	0.7	-	0.2	0.1	0.1	0.2	0.1	0.1	0.2	0.1	0.2	0.1	0.1	0.1	0.2	RI, MS
70	Geranyl acetate	1362	1360	1750	0.9	-	3.2	-	1	1	0.8	1.2	1	1	0.3	1.2	1.9	0.8	0.3	1.9	0.3	RI, MS
71	$\alpha$ -Copaene	1379	1375	1491	0.4	0.9	-	-	0.7	0.4	0.5	0.5	0.4	0.4	0.6	0.3	0.5	0.3	0.3	0.3	0.7	RI, MS
72	(Z)-Jasmone	1378	1378	1917	tr	-	-	0.1	0.1	tr	-	0.1	tr	-	-	-	tr	tr	tr	0.1	0.1	RI, MS
73	$\beta$ -Bourbonene	1386	1380	1516	0.1	0.2	-	-	0.2	0.1	0.1	0.1	-	tr	0.2	tr	0.1	0.1	0.1	0.1	0.2	RI, MS
74	$\beta$ -Elemene	1389	1382	1589	0.4	0.6	-	-	0.5	0.4	0.3	0.5	0.1	0.3	0.6	0.1	0.5	0.4	0.1	0.1	0.6	RI, MS
75	Dodecanal	1385	1388	1690	0.1	-	0.3	-	0.1	0.1	0.1	0.2	0.2	0.2	0.2	0.1	tr	0.1	0.1	0.1	0.2	RI, MS
76	$\alpha$ -Gurjunene	1413	1411	1524	0.3	0.5	-	-	0.4	0.1	0.3	0.3	0.3	0.3	0.3	0.2	0.3	0.3	0.1	0.1	0.4	RI, MS
77	(E)- $\beta$ -Caryophyllene	1421	1418	1595	10	20.3	-	-	15.8	14	11.3	11.5	14.7	14.8	13	10.6	10.8	9.6	9.6	9.6	15.8	RI, MS
78	$\beta$ -Copaene	1430	1424	1579	0.1	0.1	-	-	0.2	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.2	RI, MS
79	Geranyl acetone	1430	1428	1845	0.2	-	0.8	-	0.1	0.1	0.1	0.4	0.1	0.1	0.3	0.2	0.4	0.3	0.1	0.1	0.4	RI, MS
80	$\alpha$ -Humulene	1455	1448	1665	0.5	1.1	-	-	0.8	0.7	0.6	0.6	0.7	0.8	0.7	0.6	0.6	0.5	0.5	0.5	0.8	RI, MS
81	Thymyl isobutyrate	1462	1458	1875	0.1	-	0.2	-	0.3	0.4	0.2	0.1	0.2	0.1	0.3	0.1	0.2	-	-	0.1	0.4	RI, MS
82	Allo-Aromadendrene	1462	1465	1642	0.6	1.2	-	-	0.9	0.4	0.7	0.7	0.6	0.6	0.9	0.5	0.7	0.5	0.4	0.4	0.9	RI, MS
83	$\beta$ -Ionone	1464	1466	1929	0.2	-	0.5	-	0.2	0.3	0.2	0.2	-	0.2	0.4	0.3	0.1	0.2	0.1	0.1	0.4	RI, MS
84	$\gamma$ -Gurjunene	1472	1469	1648	0.1	0.1	-	-	0.3	0.1	0.3	0.2	0.2	tr	0.2	0.1	0.2	0.1	0.1	0.1	0.3	RI, MS
85	$\gamma$ -Muurolene	1474	1470	1671	0.1	0.1	-	-	0.1	tr	0.1	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.2	RI, MS
86	Tridecan-2-one	1477	1479	1809	0.2	-	0.3	-	0.1	-	-	tr	tr	-	0.1	0.2	0.1	0.2	0.1	0.2	0.2	RI, MS
87	Germacrene D	1479	1473	1707	3.8	8.2	-	-	7.6	5.8	4.4	4.3	6	6.1	6.5	3.6	4.1	3.1	3.1	3.1	7.6	RI, MS
88	Viridiflorene	1491	1488	1688	0.6	0.4	-	-	0.1	0.2	-	0.1	-	0.2	-	-	0.1	-	-	0.1	0.2	RI, MS
89	Bicyclogermacrene	1494	1490	1737	0.5	0.3	-	-	0.2	0.2	0.3	0.7	0.2	0.2	0.3	0.1	0.5	0.6	0.1	0.1	0.7	RI, MS
90	4- <i>epi</i> -Cubebol	1490	1492	1878	0.1	-	-	1.4	tr	0.5	0.1	0.2	1	1	0.2	0.5	0.2	0.1	0.1	0.1	1	RI, MS
91	Pentadecane	1500	1499	1500	tr	0.1	-	-	0.1	tr	tr	tr	tr	-	-	-	tr	tr	-	0.1	0.1	RI, MS
92	$\gamma$ -Cadinene	1507	1505	1749	tr	0.5	-	-	tr	0.6	0.3	0.5	0.5	0.5	0.7	0.4	0.4	0.3	0.3	0.3	0.7	RI, MS
93	Cubebol	1514	1509	1929	0.4	-	-	1.2	0.6	0.1	-	0.1	-	-	-	0.4	tr	0.1	0.1	0.1	0.6	RI, MS
94	$\tau$ -Cadinene	1520	1514	1752	0.5	1	-	-	0.8	0.5	0.5	0.6	0.4	0.4	0.7	0.4	0.6	0.4	0.4	0.4	0.8	RI, MS
95	(Z)-Nerolidol	1522	1518	1910	tr	-	-	0.1	tr	tr	-	tr	tr	-	-	-	-	-	-	-	-	RI, MS
96	(E)-Nerolidol	1553	1548	2041	0.1	-	0.2	-	0.1	tr	tr	0.1	0.1	-	-	0.1	0.2	0.1	0.1	0.1	0.2	RI, MS
97	cis-3-Hexenyl benzoate	1551	1555	2093	0.1	-	0.4	-	0.2	0.1	0.4	0.2	tr	tr	0.1	0.2	0.1	0.1	0.1	0.1	0.4	RI, MS
98	1,5-Epoxy-salvial(4)14-en	1554	1559	1910	0.2	-	0.2	-	0.2	0.3	0.1	0.1	0.2	tr	-	tr	0.2	0.1	0.1	0.1	0.3	RI, MS
99	Palustrol	1569	1564	1915	0.1	-	0.6	0.2	0.5	0.7	0.1	0.2	0.6	0.6	0.5	0.2	0.1	0.2	0.1	0.2	0.7	RI, MS
100	Spathulenol	1572	1567	2107	tr	-	-	0.2	tr	-	0.1	0.1	-	-	0.2	0.2	0.1	0.1	0.1	0.1	0.2	RI, MS
101	Germacrene-4-ol	1571	1572	2037	0.1	-	-	0.1	0.1	0.1	tr	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	RI, MS



N°	Compounds <sup>a</sup>	RI <sub>lit</sub> <sup>b</sup>	RI <sub>a</sub> <sup>c</sup>	RI <sub>p</sub> <sup>d</sup>	Total Oil <sup>e</sup>	FI	FII	FIII	PS1	PS2	PS3	PS4	PS5	PS6	PS7	PS8	PS9	PS10	Min	Max	Identification <sup>f</sup>
102	<b>Caryophyllene oxide</b>	1578	1572	1973	3	-	12	-	2.4	2.6	2	2.5	2	2	1.9	3.2	3.6	2.3	1.9	3.6	RI, MS
103	Tetradecanal	1596	1592	1851	0.3	-	0.4	-	tr	tr	tr	0.3	tr	0.1	0.1	tr	0.3	0.2	0.1	0.3	RI, MS
104	Ledol	1600	1596	2041	0.1	-	-	2.3	0.4	0.4	0.2	0.1	0.2	0.1	0.1	0.3	0.1	-	0.1	0.4	RI, MS
105	Hexadecane	1600	1601	1600	0.1	0.1	-	-	0.1	0.1	tr	tr	0.1	tr	-	0.1	0.1	tr	0.1	0.1	RI, MS
106	<i>epi</i> -Cubanol	1623	1618	2052	0.1	-	-	0.4	0.1	0.1	0.1	0.1	0.1	0.2	0.1	0.1	0.1	0.1	0.1	0.2	RI, MS
107	<i>r</i> -Cadinol	1633	1625	2158	0.6	-	1.8	4.6	0.7	0.8	0.5	0.7	0.6	0.7	0.9	0.7	0.6	0.5	0.5	0.9	RI, MS
108	<i>r</i> -Muurolol	1633	1628	2154	0.2	-	-	1	0.1	0.1	tr	0.1	-	0.1	0.1	0.1	0.1	0.1	0.1	0.1	RI, MS
109	<i>α</i> -Cadinol	1643	1638	2220	0.7	-	-	12.6	0.9	1	0.7	0.9	0.9	0.9	1.3	0.7	0.6	0.5	0.5	1.3	RI, MS
110	Eudesma-4 (15) 7-diene-1 $\beta$ -ol	1671	1668	2341	0.3	-	-	4.9	0.1	0.2	0.1	0.1	0.2	0.2	-	0.2	0.1	0.1	0.1	0.2	RI, MS
111	Pentadecanal	1693	1694	1990	0.2	-	0.7	-	0.1	0.4	0.4	0.3	0.2	0.2	0.2	0.2	0.2	0.2	0.1	0.4	RI, MS
112	Heptadecane	1700	1698	1700	tr	0.1	-	-	tr	tr	-	-	-	0.1	0.3	-	tr	-	0.1	0.3	RI, MS
113	Benzyl benzoate	1730	1732	2578	tr	-	-	0.3	0.1	tr	-	0.1	tr	-	0.1	-	0.1	-	0.1	0.1	RI, MS
114	Tetradecanoic acid	1761	1755	2641	0.2	-	-	0.2	0.1	tr	tr	0.1	0.1	tr	-	-	tr	-	0.1	0.1	RI, MS Ref
115	Hexadecanal	1795	1795	2113	tr	-	0.1	-	tr	tr	-	tr	-	-	-	-	tr	-	-	-	RI, MS Ref
116	Octadecane	1800	1798	1800	tr	tr	-	-	tr	tr	-	tr	-	-	-	tr	-	-	-	-	RI, MS Ref
117	Phytone	1823	1829	2121	0.7	-	2.5	-	0.3	0.7	0.2	0.6	0.1	0.1	0.6	0.2	0.5	0.4	0.1	0.7	RI, MS Ref
118	Nonadecane	1900	1900	1900	0.1	tr	-	-	tr	0.1	tr	0.1	tr	tr	0.1	tr	0.1	tr	0.1	0.1	RI, MS
119	Hexadecanoic acid	1962	1967	2671	tr	-	-	0.1	0.2	-	tr	-	-	-	-	-	-	-	0.2	0.2	RI, MS Ref
120	Eicosane	2000	1999	2000	tr	tr	-	-	-	0.1	-	tr	-	-	-	-	-	-	0.1	0.1	RI, MS
121	Heicosane	2100	2100	2100	0.1	0.1	-	-	tr	tr	0.2	-	-	-	0.1	-	-	-	0.1	0.1	RI, MS
122	( <i>E</i> )-Phytol	2114	2107	2606	0.2	-	-	0.9	0.1	0.4	-	0.7	0.1	0.1	0.4	tr	0.4	-	0.1	0.7	RI, MS
123	<i>n</i> -Tricosane	2300	2299	2300	tr	0.1	-	-	tr	0.1	tr	tr	tr	-	-	tr	-	tr	0.1	0.1	RI, MS
124	<i>n</i> -Tetracosane	2400	2401	2400	tr	0.1	-	-	tr	0.1	tr	-	-	tr	-	-	tr	-	0.1	0.1	RI, MS
125	<i>n</i> -Pentacosane	2500	2499	2500	0.3	0.5	-	-	0.2	0.6	0.3	0.5	0.4	0.4	0.6	0.2	0.4	0.2	0.2	0.6	RI, MS
Identification					97.6	98.9	63.9	62.7	96.4	89.7	95.9	95.9	99.0	97.7	99	98.9	95.9	98			
% Monoterpene hydrocarbons					51.4	61.9	0	0	40.1	37.2	39.4	46.1	54.7	53	40.3	60.7	44	59.7			
% Oxygenated monoterpenes					10.4	0	17.6	29.1	13.3	12.7	17.5	9.4	8.2	8.3	11.8	7.3	11.2	8.3			
% Sesquiterpene hydrocarbons					18.0	35.5	0	0	28.7	23.7	19.8	20.9	24.3	24.8	24.8	17.1	19.6	16.4			
% Oxygenated sesquiterpenes					6.0	0	14.8	29	6.2	6.9	4	5.4	6	5.9	5.4	6.8	6.1	4.4			
% Diterpene					0.2	0	0	0.9	0.1	0.4	0	0.7	0.1	0.1	0.4	0	0.4	0			
% Non-terpene oxygenated					10.5	0	30.7	3.7	7.4	7.4	14.1	12.1	4.9	4.6	14.5	6.2	13.2	8.3			
% Non-terpene hydrocarbons					1.3	1.5	0.8	0	0.6	1.4	1.1	1.3	0.8	1	1.8	0.8	1.4	0.9			

The main compounds are reported in bold.

<sup>a</sup>: Order of elution is given on apolar column (Rtx-1); <sup>b</sup>: Retention indices of literature on the apolar column (RI<sub>lit</sub>); <sup>c</sup>: Retention indices on the apolar Rtx-1 column (RI); <sup>d</sup>: Retention indices on the polar Rtx-Wax column (RI); <sup>e</sup>: Oil; total essential oil; <sup>f</sup>: RI; Retention Indices; MS: Mass Spectra in electronic impact mode; Ref: Compounds identified from literature (König et al., 2001; NIST, 1999) (5, 114, 115, 116, 117, 119); tr < 0.1.

PS: *Phagnalon sordidum*; **PS1**: Ain Douze, **PS2**: Misserghin 1, **PS3**: Ouicheba 1, **PS4**: Dzarrifette 1, **PS5**: Mansourah 2, **PS6**: Misserghin 2, **PS7**: Ouicheba 2, **PS8**: Misserghin 2, **PS9**: Ain Fetah, **PS10**: Dzarrifette 2.

These terpenoids were accompanied by non-terpenic components found at appreciable contents in this species with decanal representing the main constituent (4.5 %).

### Antimicrobial Activity

Hydrodistilled oil of *Phagnalon sordidum* was subjected to in vitro antimicrobial activity evaluation using a dilution agar method. Eleven common Gram-positive and -negative human pathogenic bacteria like *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Salmonella typhimurium* and methicillin-resistant *Staphylococcus aureus* (MRSA) etc. were tested against the oil obtained from aerial parts with antimicrobial standard agent for comparison (Table 2). According to the results of MICs determination (Table 2), essential oil of *Phagnalon sordidum* exhibited good antimicrobial activity. *Staphylococcus aureus* (ATCC 25923), *Salmonella typhimurium* (ATCC 13311), *Klebsiella pneumoniae* (ATCC 70063) and *Enterococcus faecalis* (ATCC 49452) were the most sensitive bacteria with the lowest MICs values of 0.01±0.00, 0.04±0.00, 0.04±0.00 and 3.25±0.278 mg/mL, respectively. *Pseudomonas aeruginosa* (ATCC 27853) bacteria are also more susceptible to *Phagnalon sordidum* oil, with the MIC value of 6.51±0.278 mg/mL, in comparison with gentamicin. On the other hand, *Escherichia coli* (ATCC 25922), *Listeria monocytogenes* (ATCC 15313), *Citrobacter freundii* (ATCC 8090), *Proteus mirabilis* (ATCC 35659), *Acinetobacter baumannii* (ATCC 19606) and *Bacillus cereus* (ATCC 10876) displayed high resistance to oil with MICs of >20 mg/mL.

**Table 2.** Minimum inhibitory concentrations of *Phagnalon sordidum* essential oil by agar dilution method.

Microorganisms	Essential oil	Antibiotic
	<i>Phagnalon sordidum</i> MIC (mg/mL) <sup>a</sup>	Gentamicin MIC (mg/mL) <sup>a</sup>
<b>Gram-positive</b>		
<i>Staphylococcus aureus</i>	0.01±0.00	0.01±0.00
<i>Bacillus cereus</i>	52.080±0.863	52.08±0.01
<i>Listeria monocytogenes</i>	104.156±0.528	3.12±0.00
<i>Enterococcus faecalis</i>	3.25±0.278	3.25±0.00
<b>Gram-negative</b>		
<i>Klebsiella pneumoniae</i>	0.04±0.00	3.12±0.00
<i>Pseudomonas aeruginosa</i>	6.51±0.278	<sup>b</sup>
<i>Escherichia coli</i>	104.156±0.528	0.39±0.00
<i>Citrobacter freundii</i>	52.08±0.863	0.19±0.00
<i>Acinetobacter baumannii</i>	26.04±0.341	0.78±0.00
<i>Proteus mirabilis</i>	26.04±0.341	0.19±0.00
<i>Salmonella typhimurium</i>	0.04±0.00	0.78±0.00

<sup>a</sup> Data are presented as mean values ± SD (n=3); MIC: Minimum Inhibitory Concentration expressed in mg/mL; ATCC: American Type Culture Collection.

<sup>b</sup> -: no inhibition in the concentration ranges tested.

### Antioxidant Activity

The antioxidant potential of the hydrodistilled oil obtained from *Phagnalon sordidum* was studied by analyzing the radical scavenging capacity and electron donating ability of the constituents in the essential oil. The DPPH radical scavenging activities of the essential oil and standard antioxidative compound (ascorbic acid) are shown in Table 3. The result showed a dose-dependent inhibition of DPPH radical by both, the oil and the standard and the scavenging capacity of the essential oil was lower than the one measured with the standard. The concentration at which the oil decreased DPPH radical by 50 % (IC<sub>50</sub> values) was 0.780±0.009 mg/mL. Correspondingly, IC<sub>50</sub> value for ascorbic acid, used as standard, was 0.048±0.003 mg/mL.

The reducing power of a sample was related to its electron transfer ability and might, therefore, serve as an indicator of its potential antioxidant activity. The reductive ability was determined by monitoring the Fe<sup>3+</sup>-Fe<sup>2+</sup> transformation in the presence of the hydrosols. Table 4 depicts the reducing power of the hydrodistilled oil of *Phagnalon sordidum*. All extracts showed the presence of reductive effects, which increased with an increase in concentration. However, *Phagnalon sordidum* oil has moderate reducing capacity (1.5 mg/mL) which remains smaller in relation to ascorbic acid. This moderate antioxidant activity of essential oil from the aerial parts of *Phagnalon sordidum* may be related to the sum of the effects of constituents in the essential oil.

### Discussion

β-pinene (26.0 %), (E)-β-caryophyllene (10.0 %), limonene (8.5 %), myrcene (4.7 %), decanal (4.5 %), thymol (3.9 %), germacrene-D (3.8 %), and *p*-cymene (3.4 %) were the main components of the essential oil of the Algerian *Phagnalon sordidum* (L.). The similarity in the composition of the aerial parts essential oil was found with the Corsican *Phagnalon sordidum* where (E)-β-caryophyllene (14.4 %), β-pinene (11.0 %) and thymol (9.0 %) have been predominant constituents followed by smaller amounts of hexadecanoic acid (5.3 %), limonene (4.3 %), *p*-cymene (3.5 %), caryophyllene oxide (3.1 %), linalool (3.1 %) and germacrene D (3.0 %) (Brunel et al., 2016). Senatore et al. (2005) reported similar composition for the essential oil obtained from aerial parts of *Phagnalon saxatile* (L.) Cass. from Italy, where β-pinene (5.4 %), (E)-β-caryophyllene (4.6 %) and limonene (3.5 %) were dominant components, followed by *p*-cymene, γ-cadinene, aromadendrene, δ-cadinene and spathulenol. Italian authors found that hydrodistilled oil from *Phagnalon saxatile* aerial parts was also rich in hexahydrofarnesyl acetone (4.3 %) and hexadecanoic acid, the latter amounting to 19.3 % of the oil. Algerian *Phagnalon sordidum* essential oils, Corsican *Phagnalon sordidum* (Brunel et al., 2016) and Italian *Phagnalon saxatile* (Senatore et al., 2005) were qualitatively similar but they differed by relative abundances of their main components. All the essential oils contained predominantly β-pinene, (E)-β-caryophyllene and limonene, while the Italian *Phagnalon saxatile* is dominated by hexadecanoic acid, which is present in trace amounts in *Phagnalon sordidum* essential oil from north-western Algeria.

Nowadays, many antibiotics are accessible for treating several bacterial pathogens. However, increased multidrug resistance has led to severe diseases caused by bacterial pathogens. In addition,



**Table 3.** DPPH radical-scavenging of essential oil from *Phagnalon sordidum* measured at different concentrations.

SOURCE	Concentrations (mg/mL)					
	1.0	0.8	0.6	0.2	0.1	IC <sub>50</sub>
Essential oil	62.18±0.43	54.41±0.36	36.74±0.39	9.25±0.12	8.23±1.03	<b>0.780±0.009</b>
	0.2	0.08	0.06	0.05	0.04	IC <sub>50</sub>
Ascorbic acid	98.36±0.20	97.84±0.16	68.57±0.47	51.03±1.42	39.40±0.85	<b>0.048±0.003</b>

**Table 4.** Reducing power activities of *Phagnalon sordidum* essential oils.

SOURCE	FRAP (Å = 700 nm)			
	Concentrations (mg/mL)			
	1.5	1.0	0.75	0.5
Essential oil	0.231±0.011	0.208±0.009	0.055±0.004	0.015±0.001
Ascorbic acid	0.633±0.017	0.533±0.014	0.481±0.056	0.246±0.007

the use of various antimicrobial drugs at higher doses can be hazardous to human health. This has motivated researchers to explore alternative new key molecules against bacterial strains (Swamy et al., 2016). In this regard, plant essential oils and their major chemical constituents are potential candidates as antimicrobial agents (Andrade et al., 2014). The effect of antimicrobial activity of essential oils may inhibit or slow the growth of bacteria or destroy bacterial cells. The antimicrobial activity is more frequently measured as the minimum inhibitory concentration (MIC). The antimicrobial screening of essential oils is usually conducted using the agar diffusion method (CLSI, 2006). The effectiveness of essential oils differs from one type to another as well as against different target bacteria depending on their structure (Gram-positive and Gram-negative bacteria) (Swamy et al., 2016). For instance, oregano and thyme oils exhibit higher inhibitory activity against Gram-positive bacteria and they fail to inhibit Gram-negative bacterial strains (Nevás et al., 2004). However, the essential oil of *Achillea clavennae* exhibited strong antimicrobial activity against the Gram-negative *Pseudomonas aeruginosa* bacteria while Gram-negative *Streptococcus pyogenes* was the most resistant to the oil (Skocibusic et al., 2004). In general, Gram-negative bacterial strains were more resistant than the Gram-positive bacteria (Basak et al., 2016). In fact, a Gram-positive bacterium protects its cytoplasmic membrane with a thick cell wall (Nazzaro et al., 2013). The resistance of Gram-negative bacteria may be due to the character of their hydrophobic membrane which blocks the permeation of hydrophobic molecules (Nikaido, 2003).

The antimicrobial activity of *Phagnalon sordidum* essential oil would be related to its monoterpene components which constitute about 61.8 % of the oil. Indeed, it was shown that monoterpene hydrocarbons and oxygenated monoterpenes in essential oils are able to destroy cellular integrity resulting in respiration inhibition and permeability alteration (Cox et al., 2000). Besides, the most abundant component in essential oil of *Phagnalon sordidum*,  $\beta$ -pinene, has been reported to exhibit bacteriostatic activity against *Staphylococcus aureus* (Leite et al., 2007), and this

compound is a major constituent in a number of antimicrobial essential oils (Aligiannis et al., 2001; Rivas da Silva et al., 2012). Some components of *Phagnalon sordidum* essential oil, such as the monoterpenoids limonene, thymol, myrcene, *p*-cymene and sabinene, are known for their potential antimicrobial properties against both gram-positive and gram-negative bacteria (Nazzaro et al., 2013; Hosseinkhani et al., 2016). In addition, other minor component such as  $\alpha$ -pinene, has been known to exhibit antimicrobial activity against the bacterial strains (*Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*) (Wang et al., 2012). In fact, the biological activity of essential oil is related either to the main constituents or to the synergistic effect between the major and some minor components. (Langeveld et al., 2014).

Free radicals and other reactive oxygen species have long been implicated in oxidative damage inflicted on biomolecules, e.g. proteins, amino acids, lipids or DNA (Krumova and Cosa, 2016). Their overproduction is associated with numerous disorders (Namiki, 1990). Oxidative stress caused by the imbalance between excessive formation of free radicals and limited antioxidant defenses is connected to many pathologies including age-related disorders, diabetes, atherosclerosis, dyslipidemia, cancer, inflammatory, and neurodegenerative diseases (Lobo et al., 2010; Santo et al., 2016). Under such conditions supplementation with exogenous antioxidants is required to regain a balance between free radicals and antioxidants.

Actually, essential oils, as natural sources of phenolic components, attract researchers to evaluate their activity as antioxidants or free radical scavengers (Bakkali et al., 2008; Amorati et al., 2013) because some synthetic antioxidants, e.g. butylated hydroxyanisole (BHA), butylhydroxytoluene (BHT), propyl gallate or *tert*-butylhydroquinone (TBHQ), widely used as food additives, are now suspected to be potentially harmful to human health (Namiki, 1990; Amorati et al., 2013).

The antioxidant activity of essential oils cannot be attributed only to the presence of phenolic constituents; monoterpene alcohols, ketones, aldehydes, hydrocarbons and ethers also

contribute to the free radical scavenging activity of some essential oils. For instance, the essential oil isolated from *Hedychium forrestii* var. *palaniense*, was able to reduce DPPH radicals into the neutral DPPH-H form (Sinjumol and Mani, 2016). The most powerful scavenging constituents were found to be  $\beta$ -pinene,  $\beta$ -linalool, 1,8-cineole and 4-terpineol. Indeed, antioxidant properties of essential oils such as scavenging of free radicals and reducing power often result from their monoterpene hydrocarbons, oxygenated monoterpenes and sesquiterpenes (Loizzo et al., 2010).

## Conclusion

The current study indicates that chemical composition of *Phagnalon sordidum* oil is characterized by high content of terpenoids with  $\beta$ -pinene, (E)- $\beta$ -caryophyllene and limonene as dominant compounds. The oil expressed good antimicrobial activity compared to antioxidant results obtained for tested oil. We can report that *Phagnalon sordidum* essential oil exhibited high antimicrobial activity against various bacteria strains such as *Staphylococcus aureus*, *Klebsiella pneumoniae* and *Salmonella typhimurium*, which can be profitably explored. The biological properties manifested by *Phagnalon sordidum* essential oil in this study substantiate its use in numerous health problems and as natural additives to replace synthetic antimicrobial agents in food industry. However, further studies need to be conducted to obtain more information on the safety and toxicity of the oil.

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## Author contributions

A. H. designed research; C.I. and C.J. performed the research and analyzed the data; A. H., wrote and edited the article. All authors read and approved the final manuscript.

## Conflict of interest

The authors declare that there are no conflicts of interest.

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