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Chemical composition, antioxidant and antimicrobial activities of the essential oil of *Santolina chamaecyparissus* L. of Algeria

Nouasri Ahmed[1], Dob Tahar[1*], Krimats Soumia[1], Dahmane Dahmane[1], Toumi Mohamed[1], Lynda Lamari[2], Chelgoume Chabane[3], Racheme Farida[4]

[1]*Bioactive Molecules Laboratory and Valorisation of Biomass, Ecole Normale Supérieure, B. P.92, 16500 Kouba, Algers, Algeria*

[2]*Microbiology Laboratory, Ecole Normale Supérieure, B. P.92, 16500 Kouba, Algers, Algeria*

[3]*Chromatography Laboratory, USTHB, Algers, Algeria*

[4]*Microbiology Laboratory, CRD Saidal El Mohamdia, Alges, Algeria*

PEER REVIEW

ABSTRACT

Objective: To investigate the chemical composition, antioxidant and antimicrobial activities of essential oil of *Santolina chamaecyparissus* L. cultivated in Algeria.
Methods: The chemical composition of hydrodistilled essential oil from flowering aerial parts has been analyzed by gas chromatograph and gas chromatograph-mass spectrometer techniques. The antioxidant activities conducted by 2,2-diphenyl-1-picrylhydrazyl scavenging test and β-carotene bleaching test. The antimicrobial activity was realised by agar disc diffusion method and cornell medical index was determined by agar dilution method in solid medium.
Results: About 36 components accounting more than 82% of the total oil were identified. Oxygenated monoterpenes was the main fraction (54.66%) and was represented by artemisia ketone (40.33%) as major component of this oil. The other major constituents were (Z)-thujone (9.82%), (2Z,6E)-farnesol (7.30%) and limonene (6.87%) and concerning antiradical scavenging test (2,2-diphenyl-1-picrylhydrazyl) had demonstrated a weak activity obtained with an EC_{50} of (43.01±8.04) and moderate activity for β-carotene bleaching test (47.00±3.13) at 120 min. In the other hand, this oil was found effective against all tested strains except *Pseudomonas aeruginosa* and *Aspergillus* species. This activity was ranging from 10 mm to 24 mm with the lowest minimum inhibitory concentration value between under 0.070 µg/mL to upper 9 µg/mL.
Conclusions: The results provided evidence that the studied plant might indeed be potential sources of natural antioxidant and antimicrobial agents.

1. Introduction

Many species of aromatic plants belonging to the Asteraceae family grow wild in the Mediterranean basin, in which *Santolina* was a small genus belonging to the Anthemideae tribe. Most of the *Santolina* species are pleasant by smelling evergreen shrubs or herbs and widely growing in Mediterranean regions. Members of this genus have been of interest due to their excellent medecinal value such as antiinflammatory, antispasmodic, anthelminthic[1], vermifuge and parasite repellent[2]. It is commonly known that these species are rich with essential oils[2-5]. However, different classes of natural products have been isolated from these species including flavonoids[1,6], coumarins[1,7,8], sesquiterpene lactones[9] and triterpenoids[10].

The preservative effect of many plant spices and herbs suggests the presence of antioxidants and antimicrobial constituents in their tissues[11]. The excessive production of free radicals and the

*Corresponding author: Dob Tahar, Bioactive Molecules Laboratory and Valorisation of Biomass, Ecole Normale Supérieure, B. P.92, 16500 Kouba, Algers, Algeria.
Tel: 00213555631955
E-mail: t_dob2010@yahoo.co.uk

unbalanced mechanisms of antioxidant protection results in the onset of numerous diseases and accelerate ageing; oxidation mediated by free radical reaction is also responsible for the rancidity of unpreserved food rich in unsaturated fatty acids and many synthetic antioxidant components [butylated hydroxyanisole and butylated hydroxytoluene (BHT)] have showed toxic and/or mutagenic effect[12]. Therefore, plant antioxidants are suggested as an interesting alternative[13]. In the other hand, the development of bacterial resistance to presently available antibiotics has necessitated the search for new antibacterial agents[14]. The revival of interest in herbal medication is due to the perception that there is a lower incidence of adverse reaction to plant preparation compared to synthetic pharmaceuticals[15].

Several studies concerning the chemical composition of essential oils[16-22] and few work for biological activity[23-25] of the essential oils from *Santolina chamaecyparissus* (*S. chamaecyparissus*) have been reported.

The present study attempts to examine the essential oil composition and determination of chemotype as well as the antioxidant and antimicrobial activities of essential oil from *S. chamaecyparissus* L. of Algeria.

2. Materials and methods

2.1. Plant Materials

The flowering aerial parts were collected in May 2003 from *S. chamaecyparissus* cultivated in National Experimental Garden, El Hamma, Algiers (Algeria). The plant material was authenticated by Beloued botanist in Department of Botany from Agronomic National Institute, Algiers-Algeria, where voucher specimens were deposited at the Herbarium (HINA/FA/no. P61). The plant material was shade air-dried, finely pulverized until experiments.

2.2. Extraction of the essential oil

Essential oil was extracted from air-dried flowering aerial parts of *S. chamaecyparissus* by hydrodistillation for 3 h using a Clevenger-apparatus type. Isolated oil was dried over anhydrous sodium sulfate, filtered and stored in a dark glass vial at 4 ℃ until chemical analysis and biological assays. The yield of the obtained oil was 1.67%, based on dry weight of samples.

2.3. Physico-chemical properties

The physico-chemical indices of the oil were determined following the International Standards Organization (ISO) regulations. ISO 280: 1976 for the refractive index, ISO 279: 1981 for the specific gravity, ISO 592: 1981 for the optical rotation, ISO 709: 1980 for the ester value, ISO 1242: 1973 for the acid value.

2.4. Gas chromatography (GC)

Analytical gas chromatography was performed using Thermoquest-Finnigan Trace GC chromatograph CE system fitted with a DB-1 fused silica capillary column (30 m×0.25 mm, film thickness 0.25 μm). Oven temperature was kept at 60 ℃ for 3 min and programmed to reach 240 ℃ at the rate of 3 ℃/min; using nitrogen as the carrier gas at a flow rate of 1 mL/min; split mode ratio of 1:20. The samples were injected (0.2 μL of pentane) at the injector temperature of 250 ℃; detector temperature of flame ionization detector: 260 ℃.

Quantitative data were obtained from electronic integration of GC peak area percent data on DB-1 column, without the use of correction factors.

2.5. Gas chromatography-mass spectrometry (GC-MS)

A Thermoquest-Finnigan Trace GC-MS system, equipped with a DB-1 fused silica capillary column (30 m×0.25 mm i.d., film thickness 0.25 μm) and operating under the GC conditions: programmed heating at 60 ℃ for 3 min to 240 ℃ for 3 ℃/min. The injector temperature was 250 ℃. Helium was the GC carrier gas at 1 mL/min; the sample (1 μL) was injected in the split mode (1:20). The MS conditions were as following: ionization energy, 70 eV; electronic impact ion source temperature, 200 ℃; scan rate, 1.6 scan/s; mass range m/z, 40-500 Da.

2.6. Identification of essential oil components

The retention indices (RI) were calculated by comparing the retention times of the eluting peaks with those of a series of n-alkanes (C5-C28) using linear interpolation[26]. The components were identified by comparing RI and mass spectra with those stored in the Mississippi electronic library (NIST) and with those reported in the literature[27].

2.7. Antioxydant activity

2.7.1. 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity assay

The method of Braca *et al*. with a little modification was used for determination of scavenging activity of DPPH free radical[28]. Essential oil was prepared in methanol of 400 μg/mL. Different methanolic dilutions of essential oil (10 μg/mL to 100 μg/mL) were mixed with equal volumes of freshly prepared DPPH methanol solution (0.004% w/v). The reaction mixture was vortexed thoroughly and then left to stand at room temperature in the dark for 30 min. The absorbance was read at λ=517 nm using a blank containing the same concentration of extracts without DPPH. Ascorbic acid and BHT were taken as standards. Inhibition of the DPPH free radical in percent (I%) was calculated based on control reading, which contain equal volumes of DPPH solution and methanol without any test compound using the following equation:

$$\% \text{ inhibition} = \frac{A_C - A_S}{A_C} \times 100$$

Where A_C is the absorbance of control reaction and A_S is the absorbance of the sample.

The extract concentration providing 50% inhibition (IC_{50}) was calculated from the graph of scavenging effect percentage against extracts concentrations.

2.7.2. β-carotene/linoleic acid bleaching assay

This test was carried out according to a described procedure based on the aptitude of various extracts to decrease the oxidative discoloration of β-carotene in an emulsion[29]. About 2 mg of β-carotene was dissolved in 10 mL of chloroform. About 1 mL of this solution was pipetted into a round-bottom flask containing 20 mg of linoleic acid and 200 mg of Tween 40. Chloroform was completely evaporated using a vacuum evaporator. Then, 50 mL of distilled water was added slowly to the residue and the solution was vigorously agitated to form a stable emulsion. A total of 4.8 mL of the obtained emulsion were transferred into different test tubes containing 0.2 mL of essential oil. The mixture was then gently mixed and placed in a water bath at 50 ℃ for 120 min. Absorbance at 470 nm was measured every 30 min for 120 min. Blank solution was prepared in a similar way except that addition of β-carotene was omitted. Ascorbic acid and α-tocopheroland BHT were used as standards. The bleaching rate (R) of β-carotene was calculated according to first-order kinetics, as described in Al-Saikhan *et al*[30].

$$R = \ln \frac{A_{t=0}/A_{t=t}}{t}$$

Where, In=natural log; t is the time in minutes; $A_{t=0}$ is the initial absorbance of the emulsion immediately after sample preparation (t=0 min) and $A_{t=t}$ is the absorbance at time t (30, 60, 90 and 120 min). The percent of antioxidant activity (AA) was calculated using the equation:

$$AA = \frac{R_{control} - R_{sample}}{R_{control}} \times 100$$

Where, $R_{control}$ and R_{sample} are average bleaching rates of the negative control and the antioxidant (plant extract, ascorbic acid and BHT), respectively.

2.8. Antimicrobial activity

2.8.1. Test microorganism

In this study, nine bacteria strain were used five Gram-positive and four Gram-negative. All of them are ATCC: American Type Culture Collection, provided from CRD Saidal Algeria. Some of them are pathogenic, concerning moulds four fungus used in this study are from Microbiology laboratory ENS, Kouba, Algeria.

2.8.2. Disc diffusion method

This assay was carried out using the disc agar diffusion method with a little modification[31]. Tested strains grown on Müller-Hinton agar at 37 ℃ for 18 h for bacteria and Sabouroud agar of one weak for fungus were suspended in a saline solution (0.9% NaCl) and adjusted to a turbidity of 0.5 MacFarland standards (10^8 CFU/mL). The suspension was used to inoculate 90 mm diameter Petri plates containing medium cited above. Sterile paper discs No. 1 (6 mm diameter) were impregnated with 50 μL of essential oil after sterilisation the disc laded on the surface of agar plates. Before incubation, the Petri dishes were stored in the dark at 4° for 1 h to enable the diffusion of essential oil. The incubation conditions were at 37 ℃ for 24 h for bacteria and 25 ℃ for up than 2 d for fungus. Antimicrobial activities were evaluated by measuring the inhibition zone diameters.

The work was achieved in aseptically conditions. neomycin 25 μL/disc and amoxicillin 25 μL/disc were used as positive control to determine the sensitivity of Gram-negative and Gram-positive bacteria respectively[32] and nystatin (1 mg/mL, 10 μL/disc) was used[33] as standard antifungal. All tests were performed in triplicate for each microorganisms strain and the final results of inhibition zone measured in millimetre were presented as the average.

2.8.3. The minimal inhibition concentration (MIC) quantification

MIC of essential oils was determined using agar dilution method[34]. Appropriate amount of essential oil (450 mg) were added aseptically to medium, containing Tween 80 as emulsifier (0.5%, V/V) to produce concentrations range of 9 mg/mL to 0.07 mg/mL in Müller-Hinton agar and Sabouroud agar. The resulting agar solutions were immediately mixed and poured into Petri dishes. The plates were spot inoculated with 1 μL of microorganism. At the end of incubation period, the plates were evaluated for the presence or absence of growth. The MIC was defined as the lowest concentration of the essential oil. To compare the MIC of the essential oils with referential products, the neomycin trisulfate hydrate and amoxicillin for bacteria and nystatin for Fungus were used as positive control.

2.9. Statistical analysis

All experiments were carried out in triplicate. Data were expressed as means±SD. Differences were evaluated by One-way ANOVA test completed by a student's *t*-test. The correlations between methods were determined using ANOVA and quantified in terms of the correlation factor. Differences were considered significant at $P < 0.05$.

3. Results

3.1. Essential oil yield

Air-dried aerial parts of cultivated *S. chamaecyparissus* were subjected to hydrodistillation using a Clevenger-type apparatus. Liquid, gilded yellow and penetrating strong odour essential oil was obtained with a yield of 1.67% (w/w), based on dry weight of the plants.

3.2. Physicochemical properties

Physicochemicals properties of the oil were also determined: specific gravity $[d]_D^{20} = 0.953$; refractive index $[n]_D^{20} = 1.4887$; optical rotation $[\alpha]_D^{20} = 0°$; acid value: 3.4575; ester value: 14.304; carbonyl value: 192.036.

3.3. Chemical composition of S. camaecyparissus L.

Component identification along with their percent composition and RI values are summarized in Table 1 where the constituents are arranged in order of their elution on DB-1 type phase capillary column and their identification has been carried out by means of together GC and GC/MS techniques. The chemical group distribution

of the oil components was also reported. In total, 36 compounds were identified, amounting to 82.75% of the whole oil. Overall, oxygenated monoterpenes represented 15 of the 36 compounds, corresponding to 54.66% of the total oil which were found as the dominating group of compounds represented by artemisia ketone (40.33%), Z-thujone (9.82%) and yomogi alcohol (1.05%). Monoterpene hydrocarbons mixture amount to 13.28%. Limonene (6.87%), camphene (3.14%) and β-pinene (2.67%) become the major components of this fraction. The oil contains a lower percentage of sesquiterpene mixture (12.44% of the whole oil, 13 derivatives), in which oxygenated ones form 8.73% represented by (2Z,6E)-farnesol (7.30%) and very low hydrocarbon fraction form 3.71% represented by γ-curjunene (2.31%).

Table 1

Chemical composition of the essential oil from *S. chamaecyparissus L.* aerial parts.

Compound[a]	Peak area (%)[b]	RI[c]
Santolina triene	T	908
Tricyclene	0.14	918
α-Pinene	0.89	929
Camphene	3.14	955
β-Pinene	2.67	975
Yomogi alcohol	1.05	983
Limonene	6.87	1008
Artemisia ketone	40.33	1044
4-Dycene	0.60	1066
P-Cymenene	1.32	1070
Linalool	0.19	1095
Z-Thyjone	9.82	1107
Cis-p-Menth-2-en-1-ol	0.10	1114
Z-Pinocarveol	0.18	1128
Camphor	0.81	1137
Trans-Pinocamphone	0.69	1143
α-Terpineol	0.84	1152
Borneol	0.25	1158
M-Cymene-8-ol	0.30	1213
Thymol methyl ether	T	1213
Bornyl acetate	T	1259
Neo-isopelegol acetate	T	1307
Cis-Piperitol acetate	T	1320
β-Longipinene	0.82	1372
Z-Caryophyllene	0.22	1396
α-Santalene	T	1407
Cis-Thujospene	T	1419
Z-β-Farnesene	T	1430
α-Curjunene	2.31	1456
Germacrene A	0.19	1479
Z-α-Bisabolene	T	1488
Z-Nerolidol	0.51	1522
1-Nor-bourbounanone	0.50	1547
Longiborneol	0.42	1578
Z-α-Santalol	0.17	1646
(2Z,6E)-Farnesol	7.30	1655
Grouped components		
Number of constituents	36	
Monoterpene hydrocarbons	15.65	
Oxygen containing monoterpenes	54.66	
Sesquiterpene hydrocarbons	3.71	
Oxygen containing sesquiterpenes	8.73	
Total identified	82.75	

Compounds[a] listed in order of elution on DB-1 column; [b] t<0.1%; RI[c] measured relative to *n*-alkanes on the DB-1 column.

3.4. AA

The essential oil was subjected to screening for the possible AA by two complementary test systems namely DPPH free radical and β-carotene/linoleic acid system and the results are given in Table 2 and Figures 1 and 2. As can be seen from the Table 2 and Figure 1, *S. chamaecyparissus* L. essential oil exhibited weak antioxidant abilities for reduce DPPH radicals (IC$_{50}$ was determined as 43.010 mg/mL). When compared to BHT 0.072±0.001 and ascorbic acid 0.004±0.001, oil has been found significantly less effective than these antioxidant agents (*P*>0.05).
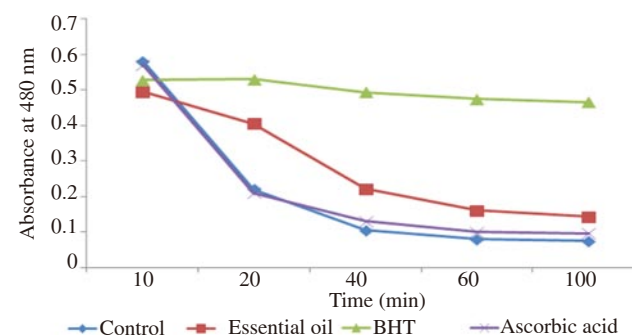
Table 2

AA of essential oil from *Santolina* sp. and standards.

Plant extracts	DPPH[b]	β-carotene/linoleic acid (%)
Essential oil	43.010±8.040	47.00±3.13
Ascorbic acid	0.004±0.001	11.05±1.43
BHT	0.072±0.001	96.92±0.51

Each value is presented as mean±SD (*n*=3); [b]IC$_{50}$ in mg/mL.
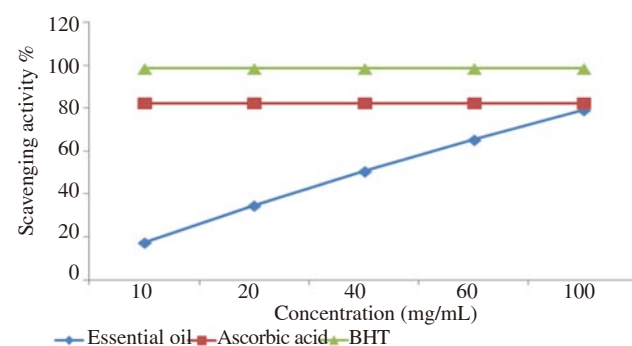
Figure 1. The DPPH radical scavenging activities of essential oil of *S. chamaecyparissus* and standards.

Figure 2. Inhibition of bleaching of β-carotene/linoleic acid emulsion by the essential oil of *S. chamaecyparissus* and standards.

For the second system, *S. chamaechyparissus* L. essential oil exhibited moderate inhibition against linoleic acid oxidation 47% (Table 2 and Figure 2). But the positive control acid ascorbic (11.05%) were not as active as BHT that demonstrated the highest activity in the β-carotene/linoleic with 96.92%.

3.5. Antimicrobial activity

3.5.1. Antibacterial activity

Determination of the inhibition zones by means of the disc

Table 3

Inhibition zone diameter and MIC of essential oil and positive control.

Bacteria strains		Inhibition diameter zone (mm)			MIC (mg/mL)		
		EO	Positive control 1	Positive control 2	EO	Positive control 1	Positive control 2
Gram-negative bacteria	*P. aeruginosa* ATCC 9027	0.00±0.00	12.00±0.00	0.00±0.00	>9.0000	9.000	>9.000
	Escherichia coli ATCC 4157	13.00±1.15	33.00±4.16	35.00±4.36	0.5600	2.250	4.500
	Bordetilla pronchiseptica ATCC 4617	14.00±1.53	21.00±0.58	27.00±4.04	>9.0000	0.560	0.281
	K. pnomoniae ATCC 4352	16.00±1.53	31.00±0.58	31.00±0.00	>9.0000	No defined	0.281
Gram-positive bacteria	*Bacillus cereus* ATCC 10876	16.00±3.61	25.00±1.15	15.00±0.58	0.2810	No defined	<0.070
	Bacillus subtilus ATCC 9372	14.00±0.58	28.00±1.53	17.00±1.73	0.5600	<0.070	1.125
	Micrococcus luteus ATCC 533	22.00±2.89	19.00±0.58	39.00±1.00	0.1400	<0.140	<0.070
	Staphylococcus aureus ATCC 6538	17.00±3.61	32.00±2.00	19.00±0.58	1.1250	<0.140	<0.070
	Enterococcus faecium ATCC 6569	18.00±2.65	33.00±4.16	33.00±4.16	<0.0175	0.140	0.281

EO: Crude essential oil of *S. chamaecyparissus* L.(50 µL/disc); Positive control 1: Neomycin treesulfat (2.77 mg/mL, 25 µL/disc); Positive control 2: Amoxicillin (2.77 mg/mL, 25 µL/disc). All values are expressed as mean±SD of three replicates.

diffusion method (Table 3) showed that the essential oil of *S. chamaecyparissus* L., exhibited an antibacterial effect against Gram-negative and Gram-positive nosocomial and animal pathogenic tested bacteria; expect *Pseudomonas aeruginosa* (*P. aeruginosa*) were no activity observed (against positive control 1 12.00±0.00). We see clearly that the effect antibacterial activity was moderate. For the others bacteria Gram-negative, the higher inhibition zone was noted for *Klebsiela pnomoniae* (*K. pnomoniae*) 16.00±1.53 against positive control 1 and 2 respectively (31.00±0.58, 31.00±0.00). The MIC of *K. pnomoniae* (Table 3) is upper 9 µg/mL, positive control 1 no identified, positive control 2: 0.281 µg/mL.

Concerning Gram-positive bacteria, the essential oil assayed for antibacterial activity showed a more pronounced effect. We noted that *Micrococcus luteus* are the big sensitive strain (Table 3).

Essential oils with inhibition zones (22.00±2.89) mm (against positive control 1 and positive control 2 respectively: 22.00±2.89, 39.00±1.00), and MIC of 0.140 µg/mL and for the positive control 1<0.140 µg/mL; positove control 2<0.070 µg/mL; for the MIC of *Enterococcus faecium*<0.0175 µg/mL, which is low than 0.140 µg/mL against positive control 1 and 0.281 µg/mL against positive control 2; and the inhibition zone (18.00±2.65) against positive control 1 and positive control 2 respectively: (33±4.16), (33±4.16).

3.5.2. Antifungal activity

Results of antifungal activity of essential oil from *S. chamaecyparissus* L. are recorded in Table 4. The oil inhibited *Penicillum* with inhibition zone of (15.00±2.31) *vs* (15.00±1.50) of Cont. and *Fusarium culmorum* (12.00±2.08) *vs* (19.00±1.73) of Cont. with the same MIC 10 mg/mL. The two spaces of *Aspergillus* no activity was recorded.

Table 4

Antifungal activities of *S. chamaecyparissus* L.

	Crude essential oil		Positive control (nystatin 30 mg/mL)	
	Inhibition zone (mm)	MIC (mg/mL)	Inhibition zone (mm)	MIC (µg/mL)
Aspergillus flavus	0.00±0.00	Not achieved	17.00±0.50	0.940
Penicillum sp.	15.00±2.31	10	15.00±1.50	0.230
Fusarium culmorum	12.00±2.08	10	19.00±1.73	0.007

All values are means of three replicates in mm and disc diameters are mean±SD.

4. Discussion

The yield of extraction obtained 1.67% (w/w) in this study, when it compared to other previously investigated samples reported in the literature of *S. chamaecyparissus* species, subspecies collected from other site in the world and other *Santolina* species[2,3]. We can say that essential oil of *S. chamaecyparissus* coming from the experimental garden of Algiers (Algeria) was considerably rich in essential oil, especially that the harvest was carried out after flowering stage, at which time the output is supposed to be the highest[35]. The different harvesting sites and development phase of plants can be partly responsible for this difference[36].

The above results for chemical composition show that our oil was dominating by artemisia ketone (40.33%), followed by *Z*-thujone (9.82%), (2Z,6E)-farnesol (7.30%) and limonene (6.87%) while a notable amount of camphene (3.14%), β-pinene (2.67%) and γ-curjunene (2.31%) were detected.

Among the essential oils of the *S. chamaecyparissus* analyzed (Table 3), the artemisia ketone chemotype is clearly distinguishable; its content dominates in several cases throughout the world[17-23]. It was possible to distinguish other types of oils rich in camphor[16,21,24], borneol or copaenol[24] and β-phellandrene or longiverbenone[20]. The present oil was found to be similar to the artemisia ketone riche oil type, with a difference being that *Z*-thujone and (2Z,6E)-farnesol, which were respectively the second and third major constituents in the present oil. These two major components were present in much lower level for *Z*-thujone or absent for (2Z,6E)-farnesol in other oils. The absence of β-phellandrene in our sample is particularly surprising as this component was usually encountered in the oils of *S. chamaecyparissus*.

It is very quite clear from these remarks the existence of variation between these *S. chamaecyparissus* oils. This variation indicate that development stage of plant and ecological factors in which plants grow, strongly influence the chemical composition of oils.

In conclusion, the essential oil composition of Algerian cultivated *S. chamaecyparissus* was typical as artemisia ketone oil and was in conformity with artemisia ketone chemotype.

At the first of bioactivities tests, the DPPH assay has been widely

used to determine the free radical scavenging activity of various plant extract and pure compounds[36]. DPPH is a stable free radical generally used to determine the ability of compounds to scavenge free radicals. The method is based on the reduction of methanolic solution of DPPH in presence of hydrogen donating molecules. The reduction of DPPH solution is monitored by measurement of absorption at 517 nm. Color changes from purple to yellow and scavenging activity correspond inversely to the absorption value measured after incubation time[36] as related at results the essential oil has performed a lower potent to scavenging the DPPH as compared to references compounds. It's due to the particularity of the chemicals composition were the monoterpenic are abundance[37].We assessed also the lipid peroxidation inhibitory activity of the essential oil by the β-carotene/linoleic acid model systems. β-carotene undergoes rapid discoloration in the absence of an antioxidant, which results in a reduction in absorbance of the test solution with reaction time. This is due to the oxidation of linoleic acid that generates free radicals that attacks the highly unsaturated β-carotene molecules in an effort to reacquire a hydrogen atom. When this reaction occurs, the β-carotene molecule loses its conjugation and, as a consequence, the characteristic orange colour disappears. The presence of antioxidant avoids the destruction of the β-carotene conjugate system and the orange color is maintained[38]. Acid ascorbic, as well known polar antioxidant, remains in aqueous phase and is consequently less efficient in protecting linoleic acid. This fact has been previously reported by other investigators[37]. Overall, results were better than those provided by radical scavenging activity and better than acid ascorbic. Probably the high terpinic percentage were more effective as a consequence of higher specificity of the assay for lipophilic compounds[39].

Concerning results obtained from antibacterial activity show that essential oil of *S. chamaecyparissus* possess a promising antibacterial activity against those strain tested, of which some of them are pathogenic, expect *P. aeruginosa*, since it's known to have high level of intrinsic resistance to virtually all known antimicrobials and antibiotics[40]. The minor susceptibility of Gram-negative bacteria may be attributed to an outer membrane surrounding the cell wall which restricts diffusion of hydrophobic compounds through the lipo-polysaccharide. Moreover, the periplasmic space contains enzymes, which are able to break down foreign molecules introduced from outside[41]. Also from this study, it appear that Gram-positive bacteria are more sensitive to this oil than Gram-negative bacteria, likewise some strain tested have a big inhibition zone but their MIC is lower or the contrary as reported that the bacteria demonstrating the biggest inhibition zones by diffusion method are not always the one that present the lowest MIC[42]. Antibacterial activity was also reported with essential oils in plants belonging Asteraceae family[43]. A wide variety of essential oils are known to possess antimicrobial properties and in many cases, this activity is due to the presence of active constituents, mainly attributable to isoprenes

such as monoterpenes sesquiterpenes, and related alcohol, other hydrocarbons and phenols[44]. In this study, chemical composition of essential oil showed that the oxygen containing monoterpenes and sesquiterpenes are more abundant than hydrocarbons ones. The bioactivities of major component artemisia ketone 40.33% are unknown[45]. The other compounds have antimicrobial activity for example Z-thujone, camphor[41]. The antibacterial activity exhibited by the essential oil from *S. chamecyparissus* L. against all bacteria strains (expect *P. aeruginosa*) could, in part, be associated with the major monoterpens constituent Z-thujone and other active molecule component such as α-pinene, β-pinene, myrcene and cis-ocimene. α-pinene and β-pinene have been reported to display strong antibacterial effects[46]. It is not so clear whether the antibacterial effect may be caused by a single active component or by the synergy of many active constituents found in the essential oil[47]. An important characteristic of essential oils and their components is their hydrophobicity, which enables them to partition the lipids of the bacterial cell membrane and mitochondria, disturbing the cell structures and rendering them more permeable. Extensive leakage from bacterial cells or the exit of critical molecules and ions will lead to death[48]. Considering the large number of different group of chemical compounds present in essential oils. It's assumed that their antimicrobial properties are most likely not attributable to only one specific mechanism[44].

For antifungal activity, the essential oil of *S. chamaecyparissus* L. has no activity against *Aspergillus flavus* and moderate inhibition zone for *Fusarium* and *Penicillium* like wise the MIC was heigher. Our oil has a variety of phytochemicals that could be considerate as responsible for the significant part of the antimicrobial activity[49]. The inherent activity of essential oils can be expected to relate to the chemical configuration of the components and the proportions in which they are presenting and inter actions between them. An additive effect is observed when the combined effect is equal to the sum of the individual effects. Antagonism is observed when the effect of one or both compounds is less when they are applied together than as individually applied[46].

All the compounds of oil known for their antimicrobial activities are either trace or a low percentage. It's the explanation of the moderate bioactivities. But compared to the standards used, in some cases our oil's displayed a higher activities than those of standard. Its either antioxidant or antimicrobial, if the concentration of the actives compounds was higher, equal or pure product, might have a superior activity to the standard used[44].

In conclusion, the essential oil composition of Algerian cultivated *S. chamaecyparissus* was typical as artemisia ketone oil and was in conformity with artemisia ketone chemotype and the bioactivities was moderate both antioxidant or antimicrobial. But as reported above in some cases, its activities pass those of standard product likewise some pathogenic bacteria tested are sensitive to it. Hence, this medicinal aromatic plant *S. chamaecyparissus* L. will be a source of natural antioxidant antimicrobial products.

Conflict of interest statement

We declare that we have no conflict of interest.

Acknowledgements

Comments

Background

At present, most of the microbial pathogens are resistant to standard antibiotics. Therefore, researchers are searching alternative sources for drug discovery. Plants offer therapeutic potentials in the form of their secondary metabolites. Plant extracts and oils have been screened for their potential uses as alternative remedies for the treatment of many infectious. Essential oils are a rich source of biologically active compounds. There has been an increased interest in looking at antimicrobial properties of essential oils of aromatic plants.

Research frontiers

The present work investigates the chemical composition, antioxidant and antimicrobial activity of the essential oil of *S. chamaecyparissus* L. of Algeria.

Related reports

The chemical composition of essential oil of *S. chamaecyparissus* is reported in this paper. The antioxidant, antibacterial and antifungal activity of this oil described by authors.

Innovations and breakthroughs

The authors used *in vitro* assays for biological activity evaluation. The authors reported chemical composition of oil by GC-MS.

Applications

Essential oils have been shown to possess antibacterial, antifungal, antiviral insecticidal and antioxidant properties. Djeddi *et al.* 2012 published a paper on *in vitro* antimicrobial properties and chemical composition of *S. chamaecyparissus* essential oil from Algeria.

Peer review

This is a good area of research where we can explore the biological activity of plants in the form of extracts and oils. The authors reported chemical compositions of oil by GC-MS and biological activity of oil which is a good part of the paper.

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