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Chemical composition and analyses of enantiomers of essential oil obtained by steam distillation of *Juniperus oxycedrus* L. growing in Algeria

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Abstract
Essential oil of *Juniperus oxycedrus* growing in Algeria was obtained by steam distillation extraction method. The oil was analysed by capillary columns (non-polar DB5 and HP-chiral-20B) using gas chromatography with flame ionization and mass spectrometric detections. The compounds were identified according to their retention indices and mass spectra (EI, 70 eV). The oil contained α-pinene (36.7%), δ-3-carene (10.6%), limonene (5.8%), myrcene (4.9%) as major constituents. In addition to these components bornyl acetate (6.0%) and camphor (4.1%) were also present. In oil steam distilled 12 chiral compounds were detected. The enantiomeric distribution of chiral compounds revealed that for α-pinene and bornyl acetate, the (+)-enantiomeric forms were present with more than 16.7% and 8.03% respectively, in the total peak area of the essential oil characterized.

Keywords: *Juniperus oxycedrus*, Cupressaceae, essential oil composition, steam distillation, enantioselective-GC-FID, GC/MS- chiral.

1. Introduction
The genus *Juniperus* (Cupressaceae) consists of approximately 75 species, all of which grow in the northern hemisphere. The genus is divided into three sections: *Caryocedrus* (one species, *J. drupacea* Labill.); *Juniperus* (= *Oxycedrus*, 14 species) and *Sabina* (60 species). *Juniperus oxycedrus* L. is 1 of 14 species in the section Juniperus (= Oxycedrus) genus *Juniperus* throughout the world [1] , although, this shrub or small tree is chiefly a Mediterranean species that grows from Morocco, Algeria, and Tunisia in north Africa into Portugal, Spain, France, Italy, Greece, the Balkans, Turkey, Lebanon and eastward into Iran [2].

Various extraction methods to obtain essential oil components from plant materials such as; steam distillation (SD), hydrodistillation (HD), supercritical fluid extraction (SFE), microwave extraction (MW), and solvent extraction (SE). Among these methods, SD has been the most common approach to extract the essential oils from the medicinal herbs/plants. The proportion of different essential oils extracted by steam distillation (SD) is 93% and the remaining 7% is extracted by the other methods [3]. The botanical material is placed in a still and steam is forced over the material. The steam extraction is the traditional process to obtain essential oils from leaves and the aromatic industry uses this method because of its low cost compared to technologically advanced methods such as supercritical fluid extraction [4].

Several studies on the chemical composition of the hydrodistilled oil leaves from *J. oxycedrus* subspecies (or varieties) have been reported in the literature, These studies concerned plants from various origins all around the Mediterranean basin; Portugal [5-9], Spain [5, 7, 10-12], France [7, 13-15], Italy [7, 16, 17], Croatia [18] and Greece [7, 10, 19], and Turkey [7, 14, 15, 20] and North African; Tunisia [21-25], Algeria [26, 27] and Morocco [7, 28-30]. Few reports have been investigated on the chemical composition of *J. oxycedrus* oil isolated by steam distillation [5, 7, 10, 14, 31]. Two studies have been reported the chemical composition of *J. oxycedrus* is obtained by supercritical carbon dioxide [21, 32].

The essential oils of *J. oxycedrus* is obtained by hydrodistillation of leaves are usually characterized by a high content of α-pinene, whatever the subspecies, the origin and the extractions process are responsible for the variation in the chemical composition of the essential oils [13].

Recently, in Bouira city of Algreia (National Park of Djurdjura), Foudil-Cherif and Yassaa studied enantiomeric and non-enatiomeric distribution of monoterpenes in the headspace of *J. oxycedrus* using HS-SPME and chiral-GC/MS [27].

In previous work, the chemical composition of the essential oil hydrodistilled of *J. oxycedrus* from Djelfa city of Algeria has been compared of the percent total oil for leaf essential oils for *J. oxycedrus* (subspecies and varieties) of other European countries [26].

To the best of our knowledge, there is no previous report on the most complete identification of essential oil of *J. oxycedrus* extracted by steam distillation from Algeria. Consequently, in continuation with our work on the characterization of aromatic and medicinal plants from Algeria [33-34].

Therefore, the objectives of this study were to (a) to investigate the chemical composition of essential oil from aerial parts of *J. oxycedrus* from Algeria obtained by steam distillation (SD), (b) Determination of the enantiomeric ratio of the terpenes components present in *J. oxycedrus* steam distilled oil using chiral gas chromatography with flame ionization detection (enantio GC-FID) and chiral GC/MS.

2. Materials and methods

2.1. Plant material

The aerial parts (twigs with needles) of *Juniperus oxycedrus* was collected in May 2011 from a single location in a reserved garden of Lakhdaria city-Bouira (69 Km South-East of Algiers). The plant was authenticated in the botanical department, National Institute of Agronomic (NIA, Algeria), where a voucher specimen of the plant has been placed in the Herbarium of this school (HNIA/FA/N°: P105bis).

2.2. Standards

All Standards used were of analytical reagent grade. GC grade standards of enantiomeric and non-enantiomeric terpenes were obtained from Sigma-Aldrich-Fluka (Germany). These pure standards were used to: optimize the separation conditions; to determine the elution order of enantiomer pairs and to provide positive identification of the terpens present in the plant species. The chiral compounds were diluted 1:100 (v/v) in *n*-hexane prior to analysis in all applications.

2.3. Steam distillation apparatus and procedure

The fresh aerial parts of *J. oxycedrus* were steam distilled for 3h using a circulatory Clevenger type apparatus [35], and the oils were dried over anhydrous sodium sulfate and stored at 4°C in the dark. Extractions were performed at least 3 times, and the mean values were reported. The oil was diluted 1:10 in hexane prior to GC injection.

2.4. GC-FID Analysis

J. oxycedrus oil was injected into a Shimadzu GC-17A V.3 chromatograph using fused silica capillary column with stationary phase DB-5. The various parameters fixed for the DB-5 column are: 30 m x 0.32 mm, 0.25 µm film thickness. The temperature program was 60°C for 3 min then 3°C/min to 240°C for 3 min; injector 250°C; detector 250°C; N_2 was used as carrier gas at a flow rate 1 mL/min in the split mode 1:50, with an injection volume of 1 µL. Quantitative data was obtained from electronic integration of area percentages without the use of correction factors.

In order to determine retentions indices (RI), a series of n-alkane (C_5-C_{28}) mixtures were analyzed under the same operative conditions on DB-5 column, the samples indices were calculated following Van den Dool and Kratz [36].

2.5. Enantio-GC analysis

The GC chiral analyses were carried out using an Agilent Technologies a GC 7890A apparatus equipped with FID and fused silica capillary column HP-chiral-20B (30 m x0.32 mm, 0.25 µm film thickness). The oven temperature was programmed as follows: 40°C (5min), 40°C-130°C (1min) at 1°C/min, 130°C-200°C (3min) at 2°C/min. Inlet temperature (split: 1/100) was 250°C and detector temperature was 300°C. Carrier gas was helium (1mL/min). Injected volume was 0.1µL.

2.6. Gas Chromatography-Mass Spectrometry (GC/MS)

The GC/MS analysis was performed on a TRACE GC Ultra-DSQ II mass spectrometer using a DB-5 capillary column (30 m x 0.32 mm, 0.25 µm film thicknesses). It was programmed from 60°C (3min) to 240°C (3min) at 3°C/min with He carrier gas at a flow rate of 1 $mL.min^{-1}$ and injector heater 250°C. The MS conditions were EI source, electron energy 70 eV and source temperature 250°C. Acquisition mass range, m/z = 40-450.

The GC/MS chiral analysis was performed with a Hewlett Packard GC (HP5890 series II) /quadripole MS system (model HP MSD5971), equipped with an electronic impact source at 200°C , fitted with a fused silica-capillary column HP-chiral-20B (30 m x 0.32 mm, 0.25 µm film thickness). The chromatographic conditions were the same with GC chiral analysis, the electron impact spectra were recorded at an ion voltage of 70 eV over a scan range of 30-600 uma.

2.7. Component Identification

Identification of the individual components was based on comparison of their GC retention indices (RI) on apolar column, with those of authentic compounds or literature data, and by comparing their mass spectral data with those stored in the spectrometer databases using the Nist, Wiley mass spectral libraries and comparison of spectra with literature data [37-40].

3. Results and discussion

3.1. Chemical composition of the essential oil

The detailed qualitative and quantitative analytical data of the constituents of steam volatile obtained by steam distillation from the aerial parts (twigs with needles) of *J. oxycedrus*, and the percentage of each compound family are summarized in Table 1, and Figure 1 shows the corresponding gas chromatogram.
The compounds are arranged in order of GC elution on the non-polar column (DB-5), and their identification has been carried out by mean GC-FID, Co-GC retention time identical to authentic some compounds and GC/MS-EI analyses in combination with retention indices. The chromatographic analyses of the essential oil allowed the identification of 81 compound representing 98.5% of the total oil. Monoterpene hydrocarbons represented the most abundant constituents of the oil of the aerial parts of *J. oxycedrus* (64.6%).
The monoterpene hydrocarbons of the oil of *J. oxycedrus* was dominated by α-pinene (36.7%), δ-3-carene (10.6%), limonene (5.8%) and myrcene (4.9%), and lower amounts of β-pinene (1.7%), terpinolene (1.2%) and tricyclene (1.0%). The major oxygen-containing monoterpenes were found to be bornyl acetate (6.0%) and camphor (4.1%), and lower amounts α-terpenyl acetate (1.6%), linalool acetate (1.1%) and linalool (0.7%). The total oxygenated monoterpenes was (17.1%). The most important sesquiterpene components were shown to be α-gurjunene (1.8%), germacrene D (1.2%) and germacrene B (1.9%), (Z)-nerolidol (2.3%) and humulene epoxide II (1.4%). However the amounts of diterpenes were low in the analyzed oil, manoyle oxide and abietadiene were found as (0.5%) and (0.1%), respectively.
To our knowledge, There are only four reports on the phytochemical studies of the steam distilled leaf essential oil of *J. oxycedrus* growing in other parts of the Mediterranean regions (Morocco, Portugal, Spain, France, Italy and Greece) [5, 7, 10, 31]. A report indicated the presence of α-pinene (54.8%), limonene (17.11%) and germacrene D (6.85%) in the leaves steam distilled oil of *J. oxycedrus* ssp. *oxycedrus* growing in Eastern Athens of Greece [31]. A similar result was obtained by Adams and other [7], in his study of France *J. oxycedrus* when the fresh leaves were treated by steam distillation and found that the largest group of constituents in the essential oil was monoterpenes (75.5%), the major components were α-pinene (53.2%), δ-3-carene (5.1%) and limonene (3.5%) in monoterpene fraction. Also, in Moroccan leaf essential oil of *J. oxycedrus* ssp. *oxycedrus*, α-pinene (45.3%) and δ-3-carene (13.9%) were the major constituents, followed by C_{10}-dienol acetate (5.8%) [7].

3.2. Analysis of enantiomeric distribution of monoterpenes oil

We report, for the first time, the studies chiral essential oil compounds exhibit a discrete enantiomeric composition which.

For the sake of comparison, we have reported in Figure 2 the chromatographic profile of the standard mixture used for the calibration of enantiomeric compounds; Figure 2(a) presented hydrocarbons monoterpene chiral and Figure 2(b) presented oxygenated monoterpenes chiral.

The percent composition of compounds enatiomeric obtained with capillary column internally coated with 20% *β*-cyclodextrin in 35% phenyl methyl polysiloxane are reported in Table 2, and Figure 3 reported the chromatographic profile of essential oil of *J. oxycedrus* extracted by steam distillation (SD), the enantiomeric distribution of α-pinene, sabinene, camphene, δ-3-carene, β-pinene, limonene, linalool, terpinen-4-ol, bornyl acetate, camphor, citronellol and borneol present in essential oil of *J. oxycedrus.*

Only one study of the enantiomeric distributions of monoterpenes fraction in *J. oxycedrus* needles and berries obtained by SPME, Foudil-Cherif and Yassaa determined three enantiomeric pairs α-pinene, camphene and β-pinene, and three absolute enantiomers (+)-sabinene, (+)-limonene and (+)-β-phellandrene (100%) of needles oil of *J. oxycedrus* [27], three compounds (α-pinene, camphene and β-pinene enantiomers) of them also separated in the present study (Table 2). α-pinene (88.6%), camphene (79.07%), δ-3-carene (74.29%), β-pinene (56.41%), limonene (95.12%), bornyl acetate (94.47%) and borneol (68.93%) showed a dominant (+)-absolute configuration, while sabinene (50.94%), linalool (63.41%) and terpinene-4-ol (77.46%), were present in (-)-absolute configuration. However, the enantiomeric pair (±)-β-citronellol and (±)-camphor no separated in this column HP chiral-20B (Figure 2(b)). In order to confirm the identity of enantiomers and eliminate possible interferences, chiral analysis was also carried out, using an MS detector.

Table1: Relative content of essential oil of *J. oxycedrus* extracted by steam distillation (SD) calculated from peak areas.

Peak number	RI[a]	Compound[b]	%[c]	Methods of identification
1.	924	Tricyclene	1.0	RI, MS
2.	938	**α-Pinene**	**36.7**	RI, MS, Co-GC [e]
3.	947	α-Fenchene	0.2	RI, MS
4.	949	Camphene	0.7	RI, MS, Co-GC
5.	954	Thuja-2,4(10)-diene	tr [d]	RI, MS
6.	970	Verbenene	0.1	RI, MS
7.	973	Sabinene	0.9	RI, MS
8.	977	**β-Pinene**	**1.7**	RI, MS, Co-GC
9.	990	**Myrcene**	**4.9**	RI, MS
10.	1011	**δ-3-carene**	**10.6**	RI, MS, Co-GC
11.	1024	*p*-Cymene	0.6	RI, MS
12.	1031	**Limonene**	**5.8**	RI, MS, Co-GC
13.	1051	(E)-β-Ocimene	tr	RI, MS
14.	1066	γ-Terpinene	tr	RI, MS
15.	1082	Fenchone	tr	RI, MS, Co-GC
16.	1087	Terpinolene	1.2	RI, MS
17.	1099	Linalool	0.7	RI, MS, Co-GC
18.	1106	*cis*-Rose oxide	tr	RI, MS
19.	1120	*exo*-Fenchol	0.4	RI, MS
20.	1124	α-Campholenal	0.3	RI, MS
21.	1144	**Camphor**	**4.1**	RI, MS, Co-GC
22.	1147	Camphene hydrate	0.2	RI, MS
23.	1155	Isoborneol	tr	RI, MS
24.	1158	*trans*-Pinocamphone	tr	RI, MS
25.	1160	Pinocarvone	tr	RI, MS
26.	1165	Borneol	0.6	RI, MS, Co-GC
27.	1172	cis-Pinocamphone	tr	RI, MS
28.	1175	Terpinen-4-ol	0.3	RI, MS, Co-GC
29.	1183	*p*-Cymen-8-ol	0.1	RI, MS
30.	1188	α-Terpineol	0.1	RI, MS
31.	1193	Myrtenal	tr	RI, MS
32.	1207	Verbenone	0.1	RI, MS, Co-GC
33.	1216	*endo*-Fenchyl acetate	0.3	RI, MS
34.	1225	Citronellol	0.1	RI, MS, Co-GC
35.	1235	*trans*-Chrysanthenyl acetate	0.1	RI, MS
36.	1239	Carvone	0.1	RI, MS, Co-GC

37.	1251	Linalool acetate	1.1	RI, MS
38.	1270	*neo*-Isopulegyl acetate	0.3	RI, MS
39.	1282	**Bornyl acetate**	**6.0**	RI, MS, Co-GC
40.	1315	(2E, 4E) Decadienol	0.1	RI, MS
41.	1331	δ-Elemene	0.2	RI, MS
42.	1344	α-Terpenyl acetate	1.6	RI, MS
43.	1370	α-Copaene	0.2	RI, MS
44.	1373	Geranyl acetate	0.2	RI, MS
45.	1378	β-Bourbonene	0.1	RI, MS
46.	1386	β-Elemene	0.4	RI, MS
47.	1406	α-Gurjunene	1.8	RI, MS
48.	1413	β-Caryophyllene	0.5	RI, MS, Co-GC
49.	1427	γ-Elemene	0.1	RI, MS
50.	1447	α-Humulene	0.7	RI, MS
51.	1457	*cis*-Cadina-1(6),4-diene	tr	RI, MS
52.	1462	*cis*-Muurola-4(14),5-diene	tr	RI, MS
53.	1470	*trans*-Cadina-1(6),4-diene	0.2	RI, MS
54.	1474	γ -Muurolene	0.1	RI, MS
55.	1480	Germacrene D	1.2	RI, MS
56.	1489	β-Selinene	0.2	RI, MS
57.	1494	α-Muurolene	0.3	RI, MS
58.	1508	Germacrene A	0.2	RI, MS
59.	1510	γ-Cadinene	tr	RI, MS
60.	1517	δ-Cadinene	1.9	RI, MS
61.	1528	*trans*-Cadina-1,4-diene	tr	RI, MS
62.	1532	(Z)-Nerolidol	2.3	RI, MS
63.	1535	α-Calacorene	0.1	RI, MS
64.	1543	Elemol	0.5	RI, MS
65.	1550	Germacrene B	1.9	RI, MS
66.	1556	(E)-Nerolidol	0.1	RI, MS
67.	1568	Germacrene D-4-ol	0.2	RI, MS
68.	1576	Caryophyllene oxide	0.3	RI, MS, Co-GC
69.	1592	Cedrol	0.1	RI, MS, Co-GC
70.	1601	Humulene epoxide II	1.4	RI, MS
71.	1620	1epi-Cubenol	0.5	RI, MS
72.	1628	γ-Eudesmol	tr	RI, MS
73.	1633	epi-α-Muurolol	tr	RI, MS
74.	1637	α-Muurolol	tr	RI, MS
75.	1641	β-Eudesmol	0.8	RI, MS
76.	1649	α-Cadinol	tr	RI, MS
77.	1662	Eudesma-4(15),7-dien-1β-ol	0.1	RI, MS
78.	1671	Shyobunol	0.2	RI, MS
79.	1706	(2E ,6Z)-Farnesol	0.2	RI, MS
80.	1981	Manool oxide	0.5	RI, MS
81.	2067	Abietadiene	0.1	RI, MS
		Total	**98.5%**	
		Component Group		
		Monoterpene hydrocarbons	**64.6%**	
		Oxygenated monoterpenes	**17.1%**	

		Sesquiterpene hydrocarbons	**9.27%**	
		Oxygenated sesquiterpenes	**6.9%**	
		Diterpenes	**0.6%**	

[a] Retention indices as determined on a DB-5 column using the homologous series of *n*- alkanes.
[b] Compounds listed in order of elution from a DB-5 column.,[c] Relative area was given according to FID area percentage data, [d]Trace (<0.1%), [e] Co GC = identification was based on retention times of authentic compounds on DB-5 capillary column.

Table 2: enantiomeric distribution of chiral components of essential oil of *J. oxycedrus* from Algeria.

Peak number	**Time**	**Enantiomer compound[a]**	**% [b]**	**% [c]**	**ee (%) [d]**	**Identification methods [e]**
1.	38.079	1S, 5S-(-)-α-Pinene	2.15	11.40		1, 2
2.	38.93	1R, 5R-(+)-α-Pinene	16.7	88.60	77.2	1, 2
3.	41.094	1S, 5S-(-)-Sabinene	0.27	50.94	10.88	2
4.	41.312	1R, 5R-(+)-Sabinene	0.26	40.06		2
5.	42.737	1S, 4R-(-)-Camphene	0.09	20.93		1, 2
6.	43.647	1R, 4S-(+)-Camphene	0.34	79.07	58.14	1, 2
7.	44.229	1S-(-)-δ-3-carene	0.09	25.71		2
8.	44.591	1R-(+)-δ-3-carene	0.26	74.29	48.58	1, 2
9.	45.611	1R, 5R-(+)-β-Pinene	0.22	56.41	12.82	1, 2
10.	46.151	1S, 5S-(-)-β-Pinene	0.17	43.59		1, 2
11.	47.497	4S-(-)-Limonene	0.08	4.88		1, 2
12.	48.111	4R-(+)-Limonene	1.56	95.12	90.24	1, 2
13.	68.883	R-(-)-Linalool	0.26	63.41	26.82	1, 2
14.	69.063	S-(+)-Linalool	0.15	36.59		1, 2
15.	78.111	(±)-Camphor*	6.2	100	100	1, 2
16.	80.944	4R-(-)-Terpinen-4-ol	1.1	77.46	54.92	1, 2
17.	81.164	4S-(+)-Terpinen-4-ol	0.32	22.54		2
18.	84.234	1R-(+)-Bornyl acetate	8.03	94.47	88.94	1, 2
19.	84.541	1S-(-)-Bornyl acetate	0.47	5.53		2
20.	87.018	(±)-β-Citronellol*	0.29	100	100	1, 2
21.	90.234	1S, 4S-(-)-Borneol	1.6	31.07		1, 2
22.	90.833	1R, 4R-(+)-Borneol	3.55	68.93	37.86	1, 2

[a] The order of elution of the different compounds and their enantiomers from the chiral column was as indicated in the table.
[b] Relative content of individual enantiomers in the oil
[c] relative content of enantiomeric pairs.
[d] enantiomeric excess
[e] Identification methods: 1; CoGC and 2; GC/MS-Chiral
* enantiomeric pairs no separated in column HP-Chiral 20B

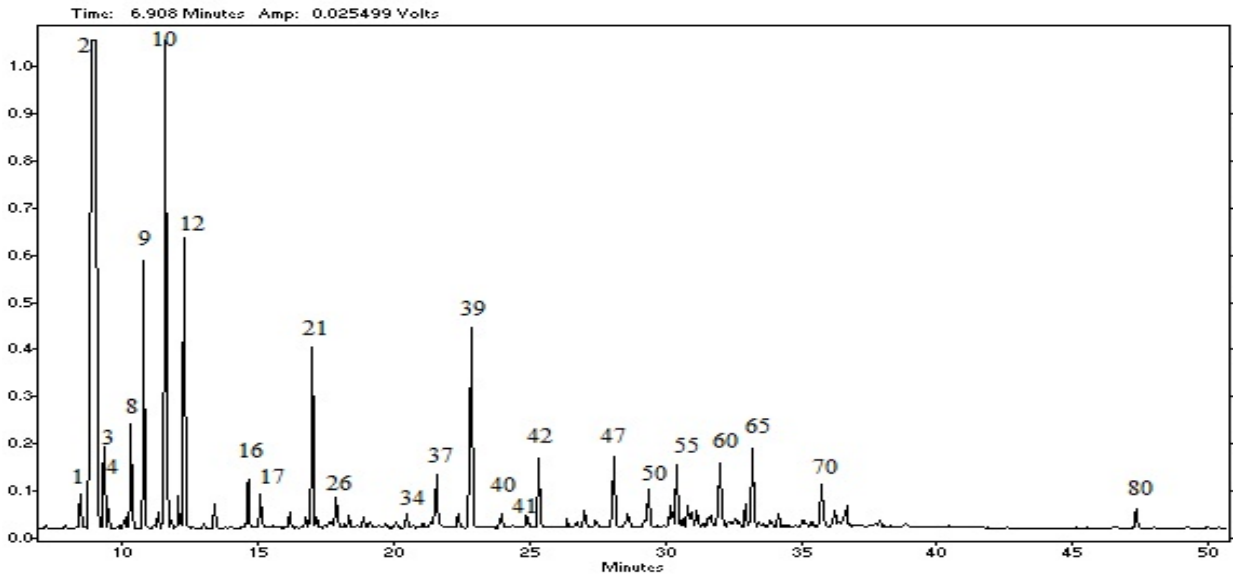


Figure1: GC-FID chromatographic profiles in apolar column DB-5 of essential oil of *J. oxycedrus*. Numbers refer to compounds identified in Table 1.

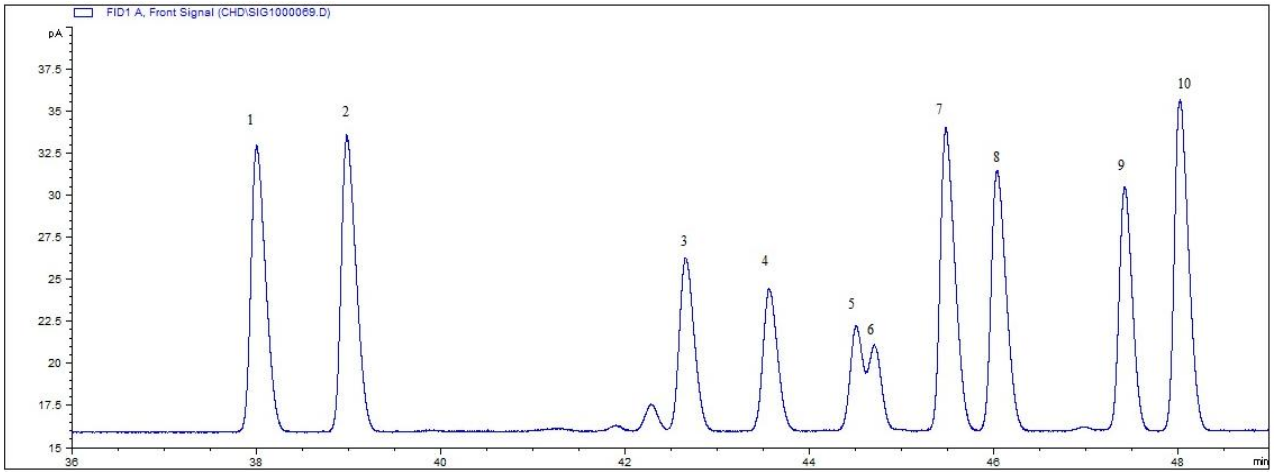
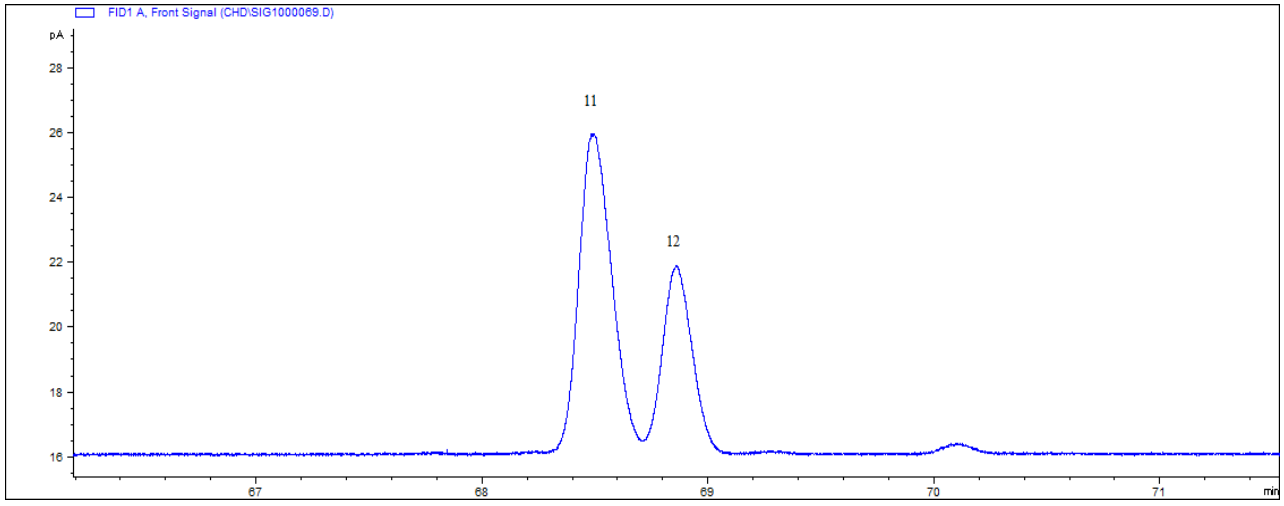


Figure 2(a) : enantio-GC- FID analysis of the hydrocarbons monoterpenes chiral; (1): (-)-α-pinene, (2): (+)-α-pinene, (3): (-)-camphene, (4): (+)-camphene, (5): (+)-δ-3-carene, (6): (-)-α-phellandrene, (7): (+)-β-pinene, (8) :(-)- β-pinene, (9) : (-)-limonene, (10): (+)-limonene.



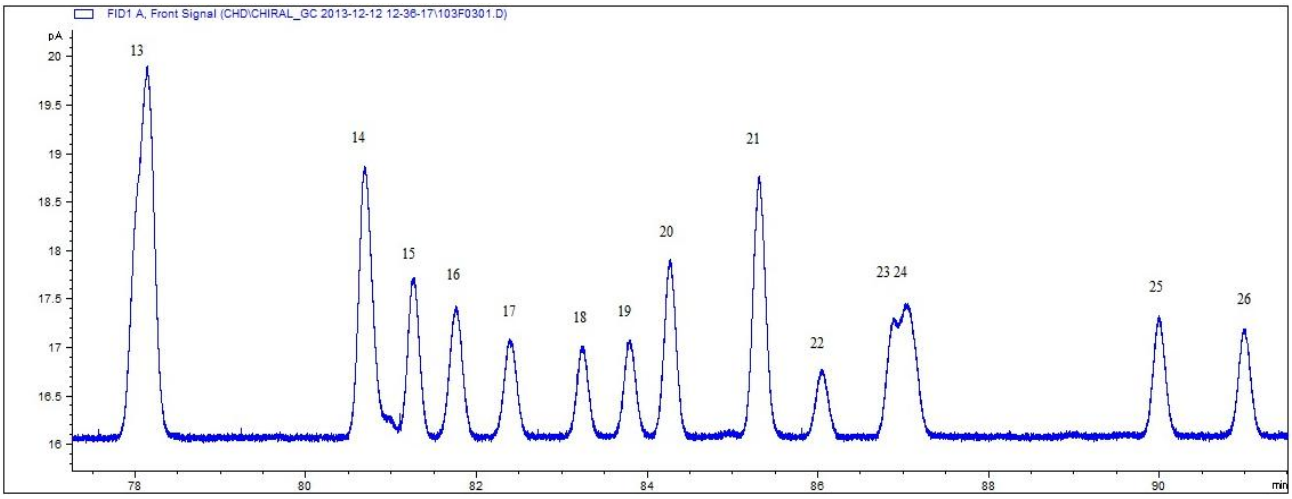


Figure 2 (b): enantio-GC- FID analysis of the oxygenated monoterpenes chiral; (11): (-)-linalool, (12): (+)-linalool, (13): (±)-camphor, (14):(+)-α-fenchol, (15): (-)-terpinene-4-ol, (16): (+)-pulegone, (17): (-)-pulegone, (18): (+)-menthol, (19): (-)-menthol, (20): (+)-bornyl acetate, (21): (-)-verbenone, (22): (+)-carvone, (23) and (24): (±)- citronellol, (25): (-)-borneol, (26): (+)-borneol.

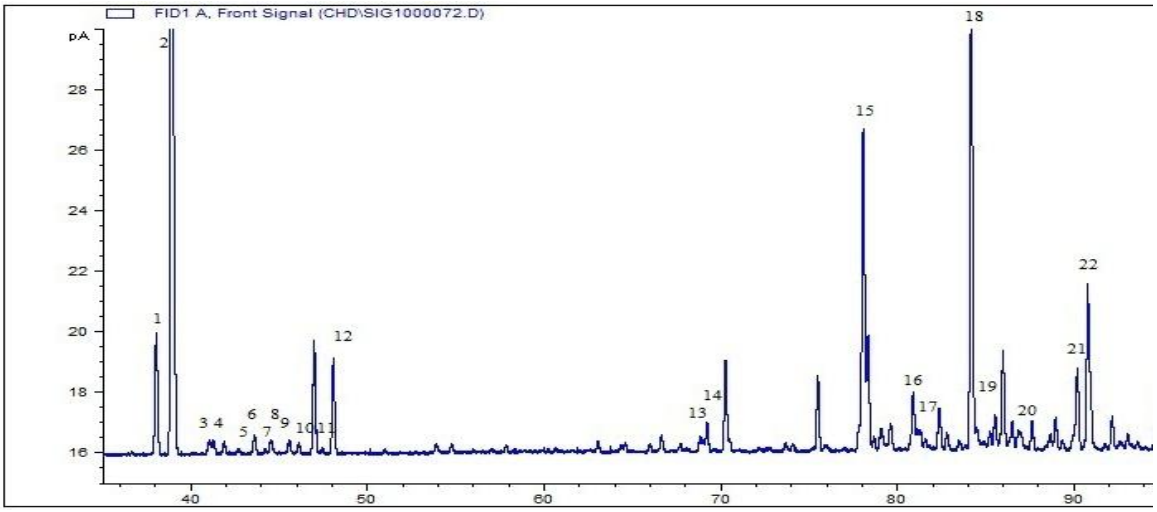


Figure 3: Zoomed GC- FID chiral chromatographic profiles of essential oil of *J. oxycedrus*. (1): (-)-α-pinene, (2): (+)-α-pinene, (3): (-)-sabinene (4): (+)-sabinene, (5): (-)-camphene, (6): (+)-camphene, (7): (-)-δ-3-carene, (8): (+)-δ-3-carene, (9): (+)-β-pinene, (10) :(-)- β-pinene, (11) : (-)-limonene, (12): (+)-limonene, (13): (-)-linalool, (14): (+)-linalool, (15): (±)-camphor, (16):(-)-terpinene-4-ol, (17): (+)-terpinene-4-ol, (18): (+)-bornyl acetate, (19): (-)-bornyl acetate, (20): (±)-citronellol, (21): (-)-bornrol, (22): (+)-borneol.

Conclusion

In this first report on the essential oil of *J. oxycedrus* obtained by steam distillation (SD) from Algeria, we present the chemical composition and their chiral compounds. The essential oil of aerial parts of *J. oxycedrus* growing in Algeria was characterized by high content of α-pinene, δ-3-carene and limonene. The chiral analysis of terpenes present in the oil steam distilled of *J. oxycedrus* was performed using enantio-GC-FID and chiral GC-MS. Large variability in the enantiomers distributions of terpenes was observed. While α-pinene, limonene and bornyl acetate showed a strong preference to (+)-enatiomer (>80%), terpinen-4-ol showed a strong preference to (-)-enatiomer. Sabinene, camphene, δ-3-carene, β-pinene, limonene, linalool and borneol showed rather a racemate.

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Reference

1. Adams R. P., Junipers of the world: The genus Juniperus. 4th ed., Trafford Publ., Bloomington, IN (2014).
2. Farjon A., A Handbook of the World's Conifers (Vol. 2). Koninklijke Brill NV, Leiden, Boston. The Netherlands (2010).
3. Masango P., *J. Clean. Prod.* 13(2005) 833.
4. Cassel E., Vargas R. M. F, *J. Mex. Chem. Soc*. 20 (2006) 126.
5. Adams R. P., *Biochem. Syst. Ecol.* 26 (1998) 637.
6. Cavaleiro C., Salgueiro L. R., Da Cunha A. P., Figueiredo A. C., Barroso J. G., Bighelli A., Casanova *J., Biochem. Syst. Ecol*. 31(2003) 193.
7. Adams R. P., Morris J. A., Pandey R. N., Schwarzbach A. E., *Biochem. Syst. Ecol.* 33 (2005) 771.
8. Cavaleiro C., Pinto E., Gonçalves M. J., Salgueiro L., *J. App. Microbiol*. 100 (2006) 1333.
9. Machado M., Santoro G., Sousa M. C., Salgueiro L., Cavaleiro C., *Flavour. Frag. J*. 25 (2010) 156.
10. Adams R. P., Altarejos J., Fernandez C., Camacho A., *J. Essent. Oil Res.* 11(1999) 167.
11. Adams R. P., *Biochem. Syst. Ecol.* 28 (2000) 515.
12. Salido S., Altarejos J., Nogueras M., Sanchez A., Pannecouque C., Witvrouw M., Clercq E. D., *J. Ethnopharmacol*. 81(2002) 129.
13. Boti J. B., Bighelli A., Cavaleiro C., Salgueiro L., Casanova J., *Flavour Frag. J*. 21(2006) 268.
14. Adams R. P., Terzioglu S., Mataraci T., *Phytologia* 92 (2010) 156.
15. Adams R. P., Mataraci T., *Phytologia* 93 (2011) 293.
16. Angioni A., Barra A., Russo M. T., Coroneo V., Dessi S., Cabras P., *J. Agric. Food Chem.* 51 (2003) 3073.
17. Valentini G., Bellomaria B., Maggi F., Manzi A., *J. Essent. Oil Res.* 15 (2003) 418.
18. Milos M., Radonic A., *Food Chem.* 68 (2000) 333.
19. Stassi V., Verykokidou E., loukis A., Harvala A., philianos S., *J. Essent. Oil Res.* 7 (1995) 675.
20. Sezik E., Kocakulak E., Baser K. H. C, Ozek T., *Chem. Nat. Compd.* 41 (2005) 352.
21. Medini H., Marzouki H., Chemli R., Khouja M. L., Marongiu B., Piras A., Porcedda S., Tuveri E., *Chem. Nat. Compd.* 45 (2009) 739.
22. Medini H., Elaissi A., Khouja M. L., Chraief I., Farhat F., Hammami M., Chemli R., Harzallah-Skhiri F., *Chem. Biodivers* 7 (2010) 1254.
23. Medini H., Marongiu B., Aicha N., Chekir-Ghedira L., Harzallah-Skhiri F., Khouja M. L., *J. Chem.* (2013) http://dx.doi.org/10.1155/2013/389252.
24. Ismail A., Lamia, H., Mohsen H., Bassem J., *Asian J. Appl. Sci*. 4 (2011) 771.
25. Riahia L., Chograni H., Ziadi S., Zaouali Y., Zoghlami N, Mliki A., *J. Essent. Oil Res.* 25 (2013) 324.
26. Dob T., Dahmane D., Chelghoum C., *Pharm. Biol*. 44 (2006) 1.
27. Foudil-Cherif Y., Yassaa N., *Food Chem.* 135 (2012) 1796.
28. Achak N., Romane A., Alifriqui M., Adams R. P., *J. Essent. Oil Res.* 21(2009) 337.
29. Mansouri N., Satrani B., Ghanmi M., El Ghadraoui L., Aafi A., Farah A., *Phytotherapie* 8 (2010) 166.
30. Derwich E., Chabir R., *Asian J. Pharmac. Cilinical Res.* 4 (2011) 50.
31. Vourlioti-Arapi F., Michaelakis A., Evergetis E., Koliopoulos G., Haroutounian S. A., *Parasitol Res*., 110 (2012) 1829.
32. Marongiu B., Porcedda S., Caredda A., De Gioannis B., Vargiu L., La Colla P., *Flavour Frag. J.* 18 (2003) 390
33. Dahmane D., Dob T., Chelghoum C., *J. Mater. Environ. Sci*., 6 (2015) In Press.
34. Krimat S., Dob T., Toumi M., Kesouri A., Noasri A., *J. Mater. Environ. Sci*., 6 (2015) 70.
35. Adams R. P., Cedar wood oil - Analysis and properties. pp. 159-173. in: Modern Methods of Plant Analysis, New Series: Oil and Waxes. H.-F. Linskens and J. F. Jackson, eds. Springler- Verlag, Berlin (1991).
36. Van den Dool H., Kratz P. D., *J. Chromatogr*. 11 (1963) 463.
37. Adams R. P., Identification of essential oil components by gas chromatography/mass spectroscopy. 4th edn, Allured Publishing Corporation, Carol Stream (2007).
38. Konig W. A., Hochmuth D. H., Joulain D., Terpenoids and related constituents of Essential Oils. Library of Mass Finder 2.1, Institute of Organic Chemistry, Hamburg (2001).
39. Cherchar H., Berrehal D., Khalfallah A., Kabouche A., Kabouche Z., *Mor. J. Chem.* 2 (2014) 85.
40. Bouratou A., Ferhat M., Kabouche A., Laggoune S., Touzani R., Kabouche Z., *J. Mater. Environ. Sci*., 5 (2014) 1214.