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Research Article

Effect of stationary factors on the yield and chemical composition of essential oils of *Tetraclinis articulata* (Vahl) Masters from Sdama Chergui massif's (Tiaret, North-West Algeria)

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Abstract

This paper aims to study the influence of exposure, altitude, and soil pH on the yield and chemical composition of essential oils (EOs) from the leaves of *Tetraclinis articulata* (Vahl) Masters, developed in the Sdama Chergui massif in northwestern Algeria. These EOs were obtained by hydrodistillation, resulting in average yields of 0.33±0.13 %, 0.29±0.07 %, 0.24±0.07 %, and 0.2±0.03 %, recorded at the south, west, north, and east exposures, respectively. Upon analyzing the obtained EOs by gas chromatography coupled with mass spectrometry (GC/MS), a total of chemical molecules grouped into 22 families were identified. The predominant compounds were Bornyl acetate (9.14 %, 7.95 %, 11.20 %, and 9.78 %), Limonene (5.03 %, 7.18 %, 5.76 %, and 5.41 %), α-Pinene (4.97 %, 5.26 %, 5.09 %, and 4.79 %), Camphor (7.53 %, 3.69 %, 3.01 %, and 5.91 %), and Borneol (5.20 %, 6.26 %, 4.93 %, and 5.11 %). Additionally, the analysis of these molecules by Factorial Analysis of Mixed Data (PCAmix) revealed that exposure, altitude, and soil pH have little effect on the yield and chemical composition of the essential oils of *Tetraclinis articulata*. Finally, careful attention to stationary parameters is crucial for obtaining essential oils of high quality and optimal quantity.

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1. Introduction

Throughout history, diverse cultures worldwide have utilized medicinal plants to address various health issues and to maintain well-being (Salmerón-Manzano et al., 2020). These plants are valued for their healing and preventive properties, containing a wealth of bioactive substances used in traditional medicine for treating numerous diseases and infections (El Khasmi & Farh, 2022; Moussaoui et al., 2024). Medicinal plants contain secondary metabolites with complex chemical combinations that are considered as potential sources of valuable therapeutics compounds (Adeosun et al., 2024). It has been hightlighted that these plants have a positive effects againts atherosclerotic cardiovascular disease (Barkas et al., 2023), high cholesterol levels, the risk of breast cancer, and preventing neurodegenerative diseases (Labdelli et al., 2019). These same plants, in response to environmental factors, produce essential oils that contain concentrated aromatic and volatile compounds known for their fragrant, cosmetic and therapeutic properties (Aćimović, 2021; Ouarghidi & Abbad, 2019).

Algeria is recognized for its rich variety of plant species, thanks to its diverse landscapes and climate (Zemour et al., 2024). In fact, over 4000 different plant species have been mentionned in Algeria (Belhouala & Benarba, 2021), with more than 600 of them being medicinal and aromatic plants (Souilah et al., 2018). It contains 248 taxa, considered as endemic plant species (Meddour et al., 2023). Among them, *Tetraclinis articulata* (Vahl) Masters is an endemic species found in the Moroccan, Algerian, and Tunisian mediterranean basin (Walas & Taib, 2022). This tree, belonging to the Cupressaceae family, is acknowledged for its redwood with a strong odor. It is commonly used in artisanal marquetry, cabinetmaking, and as a source of fuel for heat production (Bourkhiss et al., 2016; Hadjadj & Letreuch, 2017). Also, *Tetraclinis articulata* is highly regarded for its medicinal properties, such as antibacterial (Bourkhiss et al., 2023; Zerkani et al., 2019), antioxidant (Rabib et al., 2020; Saber et al., 2021a), antifungal (Benjemaa et al., 2022; Hamdani et al., 2021), anti-inflammatory (El Jemli, 2017), insecticidal (Aboulfadl et al., 2023; Sadiki et al., 2022), antidiabetic (Khatib et al., 2024), and anti-cancer activities (Calderón-Montaño et al., 2021). Additionally, *Tetraclinis articulata* is often used to treat digestive disorders and is considered a good antiseptic (Senouci et al., 2023). Generally, and in order to use an essential oil effectively and safely, it is important to identify its volatile fraction that determines its properties and its toxicity (de Sousa et al., 2023).

Numerous studies have analyzed the biochemical composition of the essential oil of the different organs of *Tetraclinis articulata*. They found a remarkable diversity, mainly due to different factors: the species itself, the growth stage, the extraction methods, and the environmental conditions (Boussaid et al., 2022; El Hachlafi et al., 2024; Jlizi et al., 2021; Khatib et al., 2024; Saber et al., 2021b; Zerkani et al., 2019).

The objective of this study is to highlight the influence of geographical exposure, altitude and soil pH on the yield and chemical profile of the essential oils from the leaves of *Tetraclinis articulata*, harvested from the Sdama Chergui massif (Tiaret region, North-West Algeria).

2. Materials and methods

2.1 Study area and plant material

The leaf harvest occurred in the region of Sdama Chergui, a massif located in western Algeria within the Tiaret mountains (situated between 35 ° 5 ' and 35 ° 20 ' North latitude and between the 0 ° meridian 47 ' and 1 ° 30 ' East of the international meridian) (Figure 1). This area features a semi-arid climate and a diverse plant cover, primarily consisting of *Tetraclinis articulata*, *Quercus ilex*, and *Juniperus oxycedrus*. Additionally, there are other notable plant species such as *Rosmarinus officinalis*, *Ampelodesmos mauritanicus*, *Pistacia lentiscus, Phillyrea angustifolia,* and

Phillyrea latifolia. Thereafter, the leaves were dried in the open air at 20 °C, shielded from light, and then carefully stored to prevent exposure to humidity and contamination.

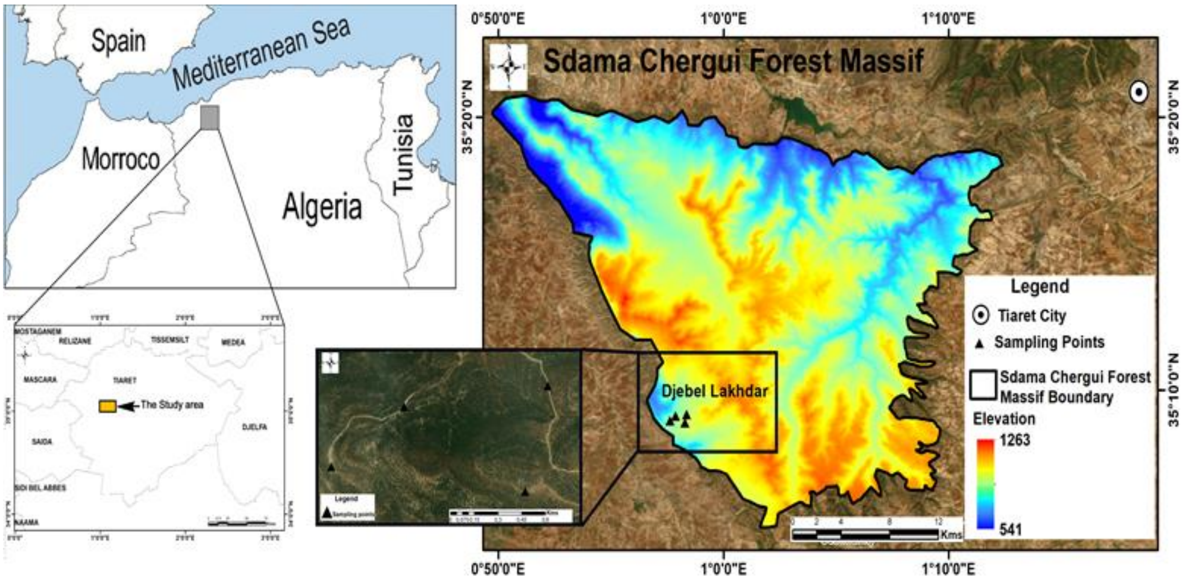


Figure 1. Map of the study area

2.2 Extraction of essential oil

The essential oils of *Tetraclinis articulata* were extracted by hydrodistillation using a Clevenger-type apparatus, using 100 g of dry matter immersed in 750 mL of distilled water, for 3 hours (Chefnaj et al., 2023). The essential oil is recovered after decantation, placed in an airtight and opaque tube and finally stored at 4 °C until its use. The dry extract yield is calculated according to the following formula:

$$\boldsymbol{Yield\ EOs\ \%} = \frac{\textbf{Weight of EOs (g)}}{\textbf{Weight dry of plant matter (g)}}\ \mathbf{x\ 100}$$

2.3. Chemical composition analysis

The qualitative analysis of essential oils was carried out using a Bruker Scion SQ Gas Chromatograph coupled to a quadruple Mass Spectrometer (GC/MS). The ion source maintains a constant temperature of 28 °C; fragmentation is carried out by electronic impact under an electric field of 70 eV, with mass exploration in a range of 35 to 600 Da.

The device was equipped with a DB-5 type capillary column (length: 25 m; diameter: 0.220 mm; film thickness: 0.25 μm). The temperature program was set from 50 °C for 10 minutes to 250 °C, and held for 15 minutes, with an increase of 2 °C per minute. For injection, 0.2 μL of the sample was injected in Split mode, with a ratio of 1/100. Helium was used as a gas carrier, with a constant flow rate of 1 mL/min throughout the operation.

The components were determined by comparing their retention times (RT) with those of reference samples, calculated using a homologous series of n-C8 to n-C28 alkanes. Subsequently, the detected mass spectra were compared with those listed in the commercial MS NIST (Mass Spectrometry National Institute of Standars and Technology) 2020 and Wiley 07 libraries, but also with the results obtained from previous studies.

3. Results and discussion

3.1 Essential oil yield

The obtained essential oils from leaves of *Tetraclinis articulata* were light yellow, slightly liquid at room temperature, with pronounced balsamic odour giving yields of 0.33 ± 0.13 %, 0.29 ± 0.07 %, 0.24 ± 0.07 % and 0.2 ± 0.03 % for the South, West, North and East respectively (Figure 2). The analysis of variance revealed that the effect of geographical exposures (North, South, East, and West) is not significant on this yield ($p < 0.05$).

In their study, Boufares et al. (2019), Hamdani et al. (2021) as well as Boussaid et al. (2022) noted that there was variability in the percentage yield of essential oils of this species harvested in three different sites in Algeria; Tiaret, Chlef and Tlemcen with values of 0.6 %, 1.1 % and 0.66 %, respectively. Similarly, different yields were recorded elsewhere: 1.71 % in Egypt (Ibrahim et al., 2017), 0.5 % in Morocco (Rabib et al., 2020), and 0.4 % in Tunisia (Benjemaa et al., 2022).

Our study highlighted that *Tetraclinis articulata* shows significant variation in essential oil yield depending on the region of origin (Boussaid et al., 2022). Also, comparing our results to those obtained by Fadel et al. (2019) on the volatile fraction of other species belonging to the Cupressaceae family (*Juniperus oxycedrus* with a rate of 0.02 % and *Cupressus sempervirens* with 0.01 %), *Tetraclinis articulata* tends to have a higher essential oil yield. This variation offers a diversity of options for different applications in aromatherapy, perfumery, and traditional medicine (Daud et al., 2022).

Several factors contribute to the variation in these yields, including factors specific to the plant such as age, vegetative stage and genetic variability (Boufares et al., 2019; Bourkhiss et al., 2011). Also, harvest time, methods and duration of drying, and extraction techniques, play an important role (Bourkhiss et al., 2009; Herzi et al., 2013; Zerkani et al., 2019). According to Hamdani et al. (2021), Boussaid et al. (2022) and Laftouhi et al. (2023), climatic conditions can also contribute to this variation.

Previous studies have found that the more sunlight the plant receives, the higher its yield. Thus, the modulation of light, in terms of intensity and spectrum, can optimize the production of bioactive compounds and essential oils of medicinal plants, improving their therapeutic properties (Zhang et al., 2021). However, Milenkovic et al. (2021) have reported that EOs of some species such us Thym, Marjoram and Origano synthetized more EOs under shading conditions.

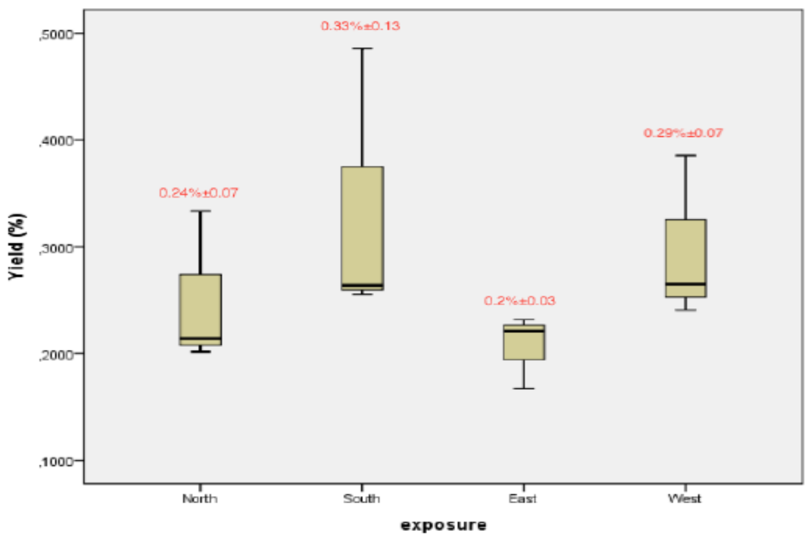


Figure 2. Average essential oil yield (%) of Tetraclinis articulata

3.2 Chemical composition of the essential oil

The GC/MS analysis allows to highlight the presence of approximately 89, 93, 89 and 85 chemical molecules for the North, South, East and West exposures representing 94.45 %, 92.73 %, 98.77 % and 90.84 % of the total composition of this oil, respectively. The results of the chemical fraction of the essential oil from the four regions (North, South, East and West) are presented in Table 1 and Figure 3.

Table 1. Chemical composition (%) of the essential oil of leaves of Tetraclinis articulata from different exposures in semi-arid conditions

RT	Compounds	Exposure East (%)	Exposure West(%)	Exposure North(%)	Exposure South(%)
5.8	Santolina triene	0.04	0.08	0.05	0.06
6.75	Tricyclene	1.80	2.13	1.74	1.67
7.12	α-Thujene	0.08	0.15	0.16	0.27
7.5	**α-Pinene**	**5.09**	**4.79**	**4.97**	**5.26**
8.28	Camphene	1.08	1.64	1.95	1.53
8.6	Thuja-2,4(10)-diene	0.07	0.26	0.36	0.21
10.13	Sabinene	1.80	1.75	2.18	2.20
11.87	β-Pinene	4.36	3.13	3.05	3.08
12.72	α-Phellandrene	0.08	0.24	0.19	0.27
14.02	α-Terpinene	0.24	0.38	0.18	0.50
15.03	para- cymene	0.28	1.14	1.04	0.54
15.42	**D-Limonene**	**5.76**	**5.41**	**5.03**	**7.18**
18.13	cis-Sabinene hydrate	0.08	0.05	0.04	0.33
18.7	*γ-Terpinene*	0.50	0.65	0.32	0.66
20.16	p-Mentha-3,8-diene	-	-	0.08	0.07
20.42	trans-Linalool oxide (furanoïd)	-	-	-	0.17
21.7	α-Terpinolene	0.32	0.94	0.63	1.20
21.8	p-Cymenene	-	-	0.63	-
22.04	Camphen-6-one	-	0.00	0.25	0.11
23.41	Linalool	0.86	1.16	0.63	1.37
23.87	1,3,5-p-Menthatriene	0.55	0.57	1.00	0.25
24.06	Endo-Fenchol	-	-	-	0.40
24.89	trans-p-Mentha-2,8-dien-1-ol	0.30	0.23	0.05	0.40
25.13	α-Campholenal	0.20	1.15	1.80	0.27
26.19	Trans-Pinocarveol	0.15	-	-	-
26.5	**Camphor**	**3.01**	**5.91**	**7.53**	**3.69**
26.81	camphene hydrate	1.31	2.09	2.98	1.11
27.75	Isoborneol	0.62	-	0.41	0.22
27.95	trans-Pinocamphone	-	-	0.15	0.19
28.12	Pinocarvone	-	-	0.59	0.29
28.72	**Borneol**	**4.93**	**5.11**	**5.20**	**6.26**
29.03	*p-Mentha-1,5-dien-1-ol*	0.25	1.32	0.59	0.28
29.66	Terpin-4-ol	2.07	2.51	3.03	2.02
30.26	trans-1(7),8-p-Menthadien-2-ol	0.24	0.12	0.19	0.27
30.72	p-Cymen-8-ol	0.09	-	1.46	0.50
30.89	α –Terpineol	2.53	-	3.07	2.84
31.32	Myrtenol	0.10	0.76	0.97	0.65
32.07	Verbenone	0.07	0.66	1.55	0.35

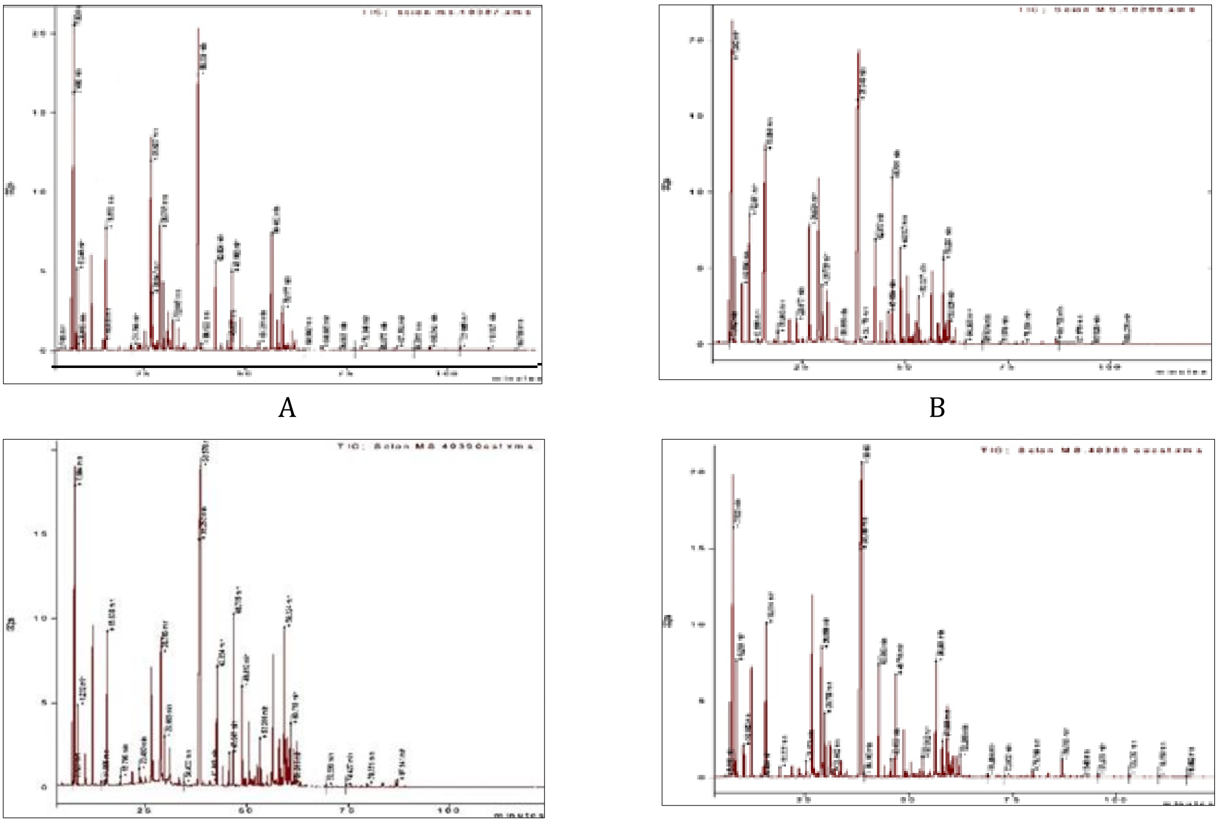
Continued Table 1

RT	Compounds	Exposure East (%)	Exposure West(%)	Exposure North(%)	Exposure South(%)
32.51	Cymen-9-ol	-	-	0.23	-
32.86	Iso-dihydrocarveol	0.11	0.15	0.15	0.48
33.16	endo-Fenchyl acetate	0.35	0.37	0.17	0.93
33.45	*trans*-Carveol	0.31	1.14	1.52	0.48
33.73	trans-p-Mentha-1(7),8-dien-2-o1	0.01	0.13	0.16	0.30
34.4	Cis-Carveol	0.16	0.28	0.41	0.27
34.84	Carvone	0.16	0.56	0.63	0.25
35.17	carvacrol methyl ether	0.03	0.06	0.08	0.13
35.65	Carvotanacetone	-	0.00	0.17	0.23
36.52	Iso-3-thujanol acetate	0.10	0.12	0.09	0.17
36.97	perilla aldehyde	0.15	0.18	0.06	0.10
37.72	cis-Verbenyl acetate	0.34	0.18	0.42	0.21
38.08	**Bornyl acetate**	**11.20**	**9.78**	**9.14**	**7.95**
39.15	Undecan-2-one	0.26	0.43	0.68	0.42
39.66	cis-Pinocarvyl acetate	0.06	0.11	0.13	0.10
40.86	Myrtenyl acetate	**0.55**	0.10	0.13	0.08
41.18	linalool propanoate	-	0.08	0.12	0.17
41.7	δ-Elemene	0.03	0.05	0.04	0.09
42.37	Silphinene	0.04	0.03	0.12	0.00
42.61	α-Terpinyl acetate	2.97	2.62	2.60	2.49
43.24	Neoiso dihydro carveol acetate	-	-	-	0.17
43.7	α-Ylangene	0.04	0.07	-	0.13
4399	α-Copaene	0.88	0.57	0.48	0.89
44.46	β-Bourbonene	0.05	0.12	-	0.12
45.63	β-Cubebene	1.52	0.73	0.50	1.12
45.7	β-Elemene	0.20	0.23	-	0.87
45.89	Tetradecane	0.04	0.09	012	0.08
46.02	N-Methyl methyl anthranilate	-	-	0.14	-
46.45	Methyl eugenol	0.06	0.17	0.14	0.21
46.72	(Z)-Caryophyllene	4.03	2.67	2.51	3.23
47.31	α-Gurjunene	0.14	0.08	0.07	0.50
47.75	β-Copaene	0.02	0.09	-	-
48.79	α-Humulene	2.89	1.80	1.49	2.29
49.39	geranyl acetone	0.34	0.32	0.10	0.37
49.68	E-β-Farnesene	0.29	0.17	0.11	0.42
50.21	γ-Gurjunene	0.14	0.08	0.00	0.15
50.53	Germacrene D	2.71	1.05	0.45	2.34
50.78	cis-β-guaiene	0.13	0.10	-	-
51.11	δ-selinene	0.29	0.37	0.21	0.40
51.48	γ-amorphene	0.29	0.16	0.07	0.49
51.91	α-Muurolene	0.43	0.26	0.16	0.56
52.62	γ-Cadinene	0.86	0.46	0.24	1.04
53.32	δ-Cadinene	2.38	1.02	0.63	1.79
53.7	trans-cadina-1,4-diene	0.15	0.15	-	0.13
53.98	10-Epi-Cubebol	0.21	0.13	-	0.18
54.22	α-Calacorene	0.07	0.09	0.05	0.06
54.88	Elemol	0.04	0.17	0.07	-

Continued Table 1

RT	Compounds	Exposure East (%)	Exposure West(%)	Exposure North(%)	Exposure South(%)
55.05	GermacreneB	-	-	0.24	0.37
56.06	(E)-Nerolidol	0.52	0.32	0.27	0.31
57.04	Salvial-4(14)-ene-1-one	0.18	0.45	0.15	0.53
57.97	Humulene epoxide II	3.30	2.31	2.07	1.31
58.31	β-Oplopenone	1.10	0.85	0.64	0.57
58.69	(2, 7Z) Bisaboladien-4-ol	0.32	0.38	0.17	0.27
59.22	1-epi-cubenol	5.05	3.17	2.97	2.74
59.61	Caryophylla-4(12),8(13)-dien5α-ol	1.56	1.11	1.30	0.77
59.97	α-muurolol	3.04	1.25	0.55	1.21
60.27	Z-14-hydroxy caryophyllene	0.61	0.58	0.30	0.37
60.7	*α-Cadinol*	1.57	2.01	1.16	1.45
60.83	14-hydroxy- 9-e pi-(E)- caryophyllene	0.27	-	-	-
61.1	Z-α-Santalol	0.18	-	0.12	0.11
61.6	Eudesma-4(15),7-dien-1β-ol	1.77	1.01	1.01	0.42
61.81	Germacra-4(15),5,10(14)-trien-1-α-ol	2.12	1.83	1.17	0.89
Total		**98.77**	**90.84**	**94.45**	**92.73**

The Chromatogram of *Tetraclinis articulata* essential oils obtained by GC/MS from different exposures is shown in the Figure 3.



A B

C D

Figure 3. Chromatogram of Tetraclinis articulata essential oils obtained by GC/MS from different exposures (A: Chromatogram North exposure, B: Chromatogram South exposure, C: Chromatogram East exposure, D: Chromatogram West exposure)

It is important to note that the volatile composition of this oil was qualitatively alike but showed quantitative variations. The predominant organic fraction of essential oils extracted from the leaves of *Tetraclinis acrticulata* represented by Bornyl acetate (9.14 %, 7.95 %, 11.20 % and 9.78 %), Limonene (5.03 %, 7.18 %, 5.76 % and 5.41 %), α-Pinene (4.97 %, 5.26 %, 5.09 % and 4.79 %), Camphor (7.53 %, 3.69 %, 3.01 % and 5.91 %) and finally Borneol (5.20 %, 6.26 %, 4.93 % and 5.11 %) respectively for the North, South, East and the West. Also, it has been noted that the presence of β-Pinene (3.05 %, 3.08 %, 4.36 % and 3.13 %), Z-Caryophyllene (2.51 %, 3.23 %, 4.03 % and 2.67 %) and 1-epi-Cubenol (2.97 %, 2.74 %, 5.05 % and 3.17 %) for the same exposures, was remarkable.

Our results can be compared to those obtained by the chromatographic analysis of two essential oils from different regions of Algeria. Thus, in Tiaret, Boufares et al. (2019) revealed that Bornyl acetate (28.42 %), camphor (18.6 %), α-pinene (16.45 %), Limonene (12.75 %) and borneol (8.41 %) were the main constituents of this oil, while the presence of the same constituents was remarkable but with different rates in the essential oils of the leaves collected in Chlef, where Hamdani et al. (2021) found a majority composition of Camphor (28.03 %), Bornyl Acetate (20.5 %), α-Pinene (11.34 %), Limonene (11.09 %) and Borneol (8.01 %).

In Morocco, studies conducted by Bourkhiss et al. (2023) and El Hachlafi et al. (2024) on the volatile fraction of *Tetraclinis articulata* leaves are qualitatively similar to results of the present study. The difference was in the rate of each element. On the other hand, as they analyzed the same organ from the Aguelmous region (Khénifra Province), Eddabbeh et al. (2021) indicated the predominance of α-Pinene (29.5 %), Cedrol (19.22 %), β-Phellandrene (7.67 %), Sabinene (5.3 %) and Bornyl Acetate (1.52 %). Ibrahim et al. (2017) reported that the composition of essential oil recovered from the aerial part from Giza in Egypt was characterized by a completely different fraction. Rich in Camphor (21.23 %) and Bornyl Acetate (15.03 %) but poor in α-Pinene (5.92 %) and the presence of Fenchol, Fenchone and Isobornyl Acetate with a rate of 13.85 %, 9.48 % and 8.39 %, respectively.

In Tunisia, studies carried out by Benjemaa et al. (2022) indicated that *Tetraclinis articulata* contained Camphor with 22.4 % (*Tetraclinis articulata* ct Camphor). However, Ben Ghnaya et al. (2016) demonstrated that α-pinene (56.21 %), isobornyl acetate (7.46 %), and β-mycrene (3.08 %) were the dominant constituents of this oil. On the other hand, the results obtained by Herzi et al. (2013) revealed that the chemical composition of *Tetraclinis articulata* essential oil mainly consisted of α-Pinene (24.9 %), Linanol acetate (21.44 %), and Caryophyllene oxide (4.24 %).

Previous studies report that the diversity in the chemical composition of essential oils results from different factors, including harvest time, extraction mode, genotype and life cycle (Boussaid et al., 2022), drying time (El Hachlafi et al., 2024; Zatout et al., 2022) and the geographical origin of the plant (Khatib et al., 2024; Rabib et al., 2019). Also, the adaptation of *Tetraclinis articulata* to different conditions such as altitude, climate and soil can contribute to the biosynthesis of certain chemical molecules (Ben Ghnaya et al., 2016; Khalil et al., 2020; Zerkani et al., 2019).

This research demonstrates that the variability in concentration of volatile compounds in essential oils could be attributed to the amount of light received from the four geographic exposures. Indeed, recent studies indicate that light and duration of sunshine (the length of the day) contribute significantly to the biosynthesis of terpenes (Boukhatem et al., 2010; Wei et al., 2023).

Table 2 demonstrates the chemical compounds of the same family (defining 22 families) (https://www.nist.gov; https://pubchem.ncbi.nlm.nih.gov), where the percentage, for North, South, East and West exposures were calculated in relation to all of the compounds.

Table 2. Rate of chemical compounds of differents families in essential oils of Tetraclinis articulata as affected by the exposures

Familly	Chemicals Compounds	Exposures (%)			
		North	South	East	West
Thuyene	α-thuyene ; sabinene ; cis-sabinene hydrate ; thuya-2,4(10) - diene, Iso-3-thujanol acetate	2.99	3.42	2.15	2.56
Pinene	α-pinene ; β-pinene, verbenone	10.13	9.37	9.63	9.44
Camphene	Camphene ; camphen-6-on ; α-campholenal ; camphor ; camphene hydrate ; trans-pinocamphone	15.52	7.44	5.66	11.87
Terpinene	α- terpinene ; γ- terpinene ; α- terpinolene ; terpin-4-ol ; α-terpinéol ; α-terpinyl-acétate	10.4	10.47	8.73	7.81
Menthol	trans-p-mentha-2,8-dien-1-ol ; p-mentha-1,5-dien-1-ol ; trans-1(7), 8-p-menthadien-2-ol ; trans-p-mentha-1(7),8-dien-2-ol	1.04	1.34	0.8	1.98
Phellandrene	α-phellandrene	0.2	0.29	0.08	0.26
Menthene	p-mentha-3,8-diene ; 1,3,5-p-menthatriene	1.14	0.34	0.55	0.62
Myrtenol	myrtenol ; myrtenyl-acetate	1.16	0.78	0.55	0.94
Cymene	Para-cymene ; P-cymenene ; P-cymen-8-ol ; cymen-9-ol	3.55	1.12	0.37	1.25
Carvone	Trans-pinocarveol ; pinocarvone ; iso-dihydrocarveol ; trans-carveol ; cis-carveol ; carvone ; carvotanacetone	3.67	2.15	0.9	2.34
Linalool	Trans-linalool oxyde ; linalool ; linalool propanoate	0.79	1.84	0.87	1.36
Fenchol	Endo-fenchol ; endo-fenchyl acétate	0.17	1.43	0.35	0.4
Borneol	Iso-borneol ; bornyl acetate ; borneol	15.61	15.56	1.,95	16.39
Limonene	D- Limonene	5.32	7.74	5.83	5.95
Copaene	α-copaene ; β-copaene	0.5	0.95	0.91	0.72
Elemene	δ-elemene ; β-elemene ; elemol	0.11	1.03	0.27	0.49
Caryophyllene	(Z) caryophyllene ; Caryophyllene oxide ; caryophilla-4(12), 8(13)-dien 5α-ol ; Z-14-hydroxy caryophyllene ; 14-hydroxy-9-epi-(E)-caryophyllene ; α- humulene ; humulene epoxyde II	12.05	11.18	16.18	13.43
Gurjunene	α- Gurjunene ; γ- Gurjunene	0.07	0.7	0.28	0.17
Germacrene	Germacrene D ; Germacrene B ; Germacra-4(15),5,10(14)-trien-1-α-ol	1.96	3.88	4.89	3.17
Muurolene	α- Muurolene ; α- Muurolol	0.75	1.9	3.51	1.66
Cadinene	γ-cadinene ; Δ-cadinene ; trans-cadina-1-4-diene ; α-cadinol	2.14	4.75	5.02	4.00
Cubenol	10-epi-cubenol ; 1-epi-cubeno	3.14	3.14	5.32	3.63

The main identified families are clearly mentioned in Table 2. These families show qualitative and quantitative variation according to different exposures. Thus:

- The North exposure includes the Bornel (15.61 %), Camphene (15.52 %) and Caryophyllene (12.05 %) families.
- The southern exposure is dominated by the Borneol family (15.56 %), Caryophyllene (11.18 %) and Terpinene (10.47 %)
- The eastern exposure is dominated by the Bornel (16.95 %), Caryophyllene (16.18 %) and Pinene (9.63 %) families
- The Western exhibition is dominated by the Borneol (16.39 %), Caryophyllene (13.43 %) and Camphene (11.87 %) families.

3.3 Relation: Environmental parameters - yield - chemical composition of Tetraclinis articulata essential oils obtained by Factor Analysis of Mixed Data (PCAmix)

To study the effect of environmental parameters (altitude, soil pH and exposure) on the yield, nature and distribution of chemical compounds of *Tetraclinis articulata* essential oils, a Factorial Analysis of Mixed Data (PCAmix) was carried out. It consists of simultaneously processing mixed data (quantitative and qualitative), using the Xlsat 2019 software.

PCAmix is a method similar to Principal Component Analysis of a set of observations, which combines qualitative and quantitative variables. It includes ordinary Principal Component Analysis (PCA), and Multiple Correspondence Analysis (MCA), as cases particular (Chavent et al., 2014; Escofier, 1979; Hill & Smith, 1976; Pagès, 2004).

The result of PCAmix is shown in four (4) graphs (Figure 4):

1. Correlation circle (Graph A): Correlations between the components and the initial variables.
2. Map of modalities (Graph B): Main coordinates of pH, yield and exposure modalities.
3. Mixed graph (Graph C): Representing the Squared Loadings of the quantitative and qualitative variables.
4. Altitude modalities graph (Grap D)

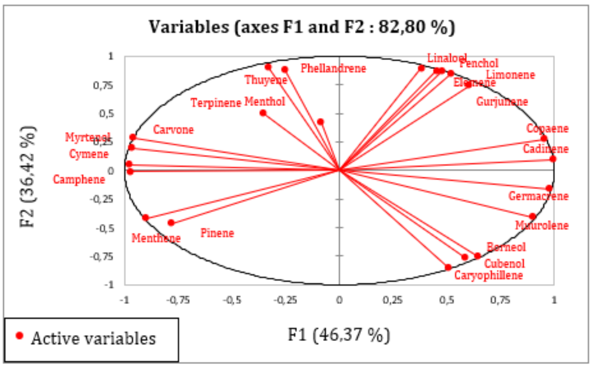
Two axes are interpreted:

- F1: represents 46.37 % of point cloud (information can be explained in this dimension).
- F2: represents 36.42 % of point cloud can be explained in this axis.

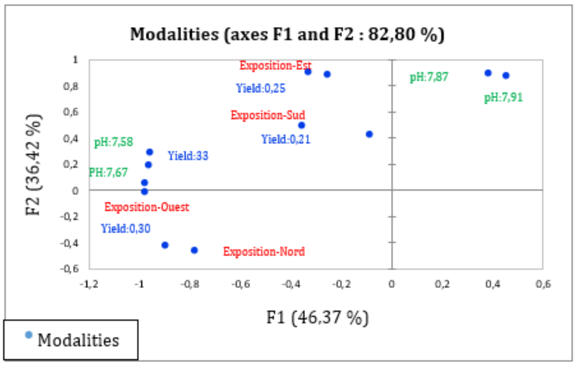
The graphs in Figure 4 indicate the quantitative variables, the qualitative variables and their links with the factor axes F1 and F2.

- Axis 1 was strongly negatively correlated with the following variables: Thuyene, Pinene, Camphene, Terpinene, Menthol, Phellandrene, Menthene, Myrtenol, Cymene and Carvone. The highest yield values were found in the northern and western exposures, with pH values of 7.58 and 7.67 and an altitude of 896 m.

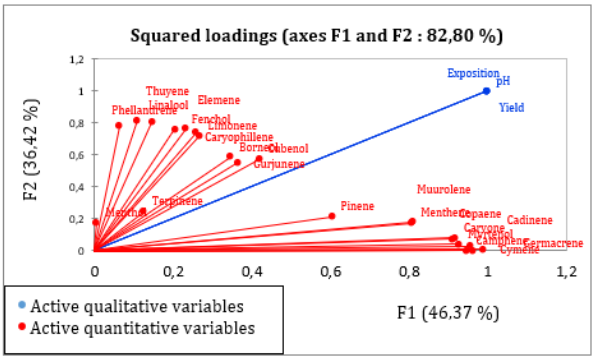
  On the positive side, the following species: Linalool, Fenchol, Borneol, Limonene, Copaene, Elemene, Caryophyllene; Gurjunene, Germacrene, Muurolene, Cadinene and Cubenol, corresponded to pH values 7.87 and 7.91 and altitude of 931 and 959 m.
- On axis 2, the variables Phellandrene, Linalool, Fenchol, Limonene, Elemene and Gurjunene were positively correlated with this axis. This suggests that these essential oil compositions had significant values in the Eastern and Southern exposures.



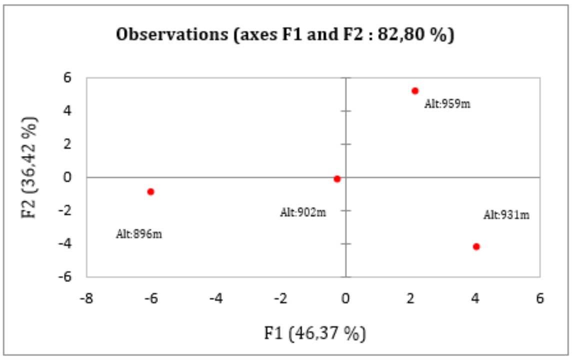
Graph A



Graph B



Graph C



Graph D

Figure 4. Relation: Environmental parameters - Yield - Chemical composition of Tetraclinis articulata essential oils obtained by Factor Analysis of Mixed Data (PCAmix): (Graph A: Circle of chemical species, Graph B: Map (pH, yield and exposures), Graph C: Mixed graph (Chemical species, yield, pH and exposures), Graph D: Altitude modalities graph)

Factor analysis of mixed data (PCAmix) revealed that stationary factors (exposure, altitude and pH) have little effect on the yield and volatile composition of essential oils in the said study. According to Fernández-Sestelo & Carrillo (2020) and Neffar & Benabdrrahmane (2013), plant populations from different exposures can present a completely diverse majority composition. Thus, the work carried out by Guerroudj et al. (2021) on essential oils from *Juniperus oxycedrus* berries shows that altitude and geographical exposure have a significant effect on its quantity and quality.

The altitude variations in the harvesting area can impact the production of active compounds (Dizajeyekan et al., 2016; Ghavam et al., 2021; Loya et al., 2023). The results of Boussaid et al. (2015) revealed that the yield of essential oils from the leaves and flowers of *Tetraclinis articulata* is not influenced by variation in altitude

Tetraclinis articulata generally thrives in slightly basic conditions, with a pH between 7.5 to 8.9 (Azzaoui et al., 2017), so pH is another factor influencing the quality and quantity of essential oils. A study conducted by Diomonde et al. (2014) on the relationship between soil and the chemical composition of *Lippia multiflora* indicates that there is little significant link between pH and the constituents of its essential oil.

Bornyl acetate, the main component of the essential oil from *Tetraclinis articulata*, holds significant value in the pharmaceutical field for its anti-inflammatory and neuroprotective properties (Lee et al., 2023; Yang et al., 2014). Due to its high concentration in the eastern exposure of the plant, focusing on the parts facing the eastern side is essential for efficient extraction of the essential oil from this species.

4. Conclusion

The evaluation of the impact of stationary parameters on the rate and chemical composition of the essential oil of *Tetraclinis articulata* leaves harvested in the Tiaret region in Algeria was the objective of this study. The results obtained reveal a qualitative and quantitative disparity. Thus, the yields of essential oils from the different exposures are 0.24±0.07 %, 0.33±0.13 %, 0.2±0.03 % and 0.29±0.07 % for the North, South, East and West respectively. The chromatographic analysis of this oil consists mainly of Bornyl acetate (9.14 %, 7.95 %, 11.20 % and 9.78 %), Limonene (5.03 %, 7.18 %, 5.76 % and 5.41 %), α-Pinene (4.97 %, 5.26 %, 5.09 % and 4.79 %), Camphor (7.53 %, 3.69 %, 3.01 % and 5.91 %) and Borneol (5.20 %, 6.26 %, 4.93 % and 5.11 %) for the North, South, East and West respectively. Furthermore, the different molecules identified were grouped into 22 chemical families. The study of stationary factors revealed that exposure, altitude and pH had a lesser effect on the qualitative and quantitative aspects of the essential oils of *Tetraclinis articulata*. Deepening this work would make it possible to improve the yield and chemical composition of essential oils produced through appropriate silvicultural practices, while taking into account environmental factors, cultivation methods and processing techniques. The importance of essential oil characterization lies in the precise identification of chemical molecules, this guarantees quality, purity and safety of use in different sectors. Additionally, it facilitates the understanding and prediction of their pharmacological and medical properties. Thus, given the high level of active biomolecules in the essential oil of *Tetraclinis articulata*, in particular Bornyl acetate, the incorporation and use of this oil in the pharmaceutical and cosmetic fields could greatly contribute to the preservation of the human health. *Tetraclinis articulata*, endemic to North Africa, represents a renewable and valuable species. However, certain factors contribute to the regression of forests, endangering their biodiversity and the ecological aspects they offer. Its preservation is therefore essential, by adopting a comprehensive approach combining various conservation strategies, as well as by raising awareness and educating local populations and the general public on the importance of this species.

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Author's declaration and contribution

The authors have no competing interests to declare. BN provided the GC/MSanalysis and the wrinting of this acticle. MM (Professor) provides proofreading and correction of the manuscript before submission. MZZ (Master student in Biodiversity and Plant Ecology) carried out statistical analysis of the data. ZK contributes to the correction and linguistic revision.
All authors have read and approved the final version of the manuscript.

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