Trop J Nat Prod Res, March 2024; 8(3):6570-6581 **ISSN 2616-0684 (Print)**
ISSN 2616-0692 (Electronic)

Tropical Journal of Natural Product Research

Available online at https://www.tjnpr.org

Original Research Article

Comparative Chemical and Biological Analysis of Wood and Tar Essential Oils from *Cedrus atlantica* and *Juniperus oxycedrus* in Morocco

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ARTICLE INFO

Article history:
Received 04 January 2024
Revised 27 February 2024
Accepted 04 March 2024
Published online 01 April 2024

ABSTRACT

Wood tar essential oils have garnered attention for their potential therapeutic benefits and industrial applications. However, despite their historical significance, comprehensive analyses comparing artisanal and laboratory-produced variants are notably absent in current literature. Therefore, this research aims to bridge this gap by conducting chemical analysis to identify key constituents in artisanal and laboratory-produced essential oils of wood tar, as well as their antimicrobial and antioxidant activities. The DPPH and FRAP assays were employed to assess antioxidant activity to determine EC_{50} values. The results of the study revealed the following major phytoconstituents in the tar: β-himachalene (30.24%; 22.78%), α-himachalene (16.06%;14.23%), and γ-Gurjunene (9.69%;7.88%). Similarly, for AJTEO and LJTEO, the major constituents observed were α-Cedrene (32.85%; 27.89%), Terpinen-4-ol (7.06%;10.13%), and β-Cedrene (3.51%; 4.85%). In the antimicrobial assessment, laboratory wood tar EO shows stronger inhibition against tested organisms than artisanal wood tar EO. Juniper tar EO was more effective than cedar tar EO. LCTEO displayed potent antioxidant activity, with an EC_{50} value of 16.09 µg/mL, followed by ACTEO at 17.659 µg/mL. AJTEO and LJTEO exhibited robust antioxidant effects with EC_{50} values <1.25 µg/mL. In the FRAP assay, LCTEO demonstrated potent antioxidant activity with a value of 395.66 ± 0.02 µg/mL, while ACTEO showed an EC_{50} of 532.71±0.053 µg/mL. AJTEO displayed an EC_{50} value of 293.3±0.012 µg/mL, and LJTEO exhibited an EC_{50} of 238.12±0.014 µg/mL. This research compares artisanal and laboratory-produced wood tar essential oils and their potential for application in pharmacy and cosmetic industries as antimicrobial and antioxidant agents.

***Keywords*:** Essential Oils, Juniper tar, Cedar tar, Chemical analysis, antimicrobial & antioxidant activity.

Introduction

Wood tar, a viscous liquid derived from the pyrolysis of plant material, has found applications not only in protecting wood surfaces and marine ropes but also in pharmacy practice as stomachic and in treating skin conditions and cosmetics as soaps. However, variations in its physico-chemical properties and biological activity exist depending on plant species, geographical location, availability, and desired product characteristics.[1,2] *Juniperus oxycedrus*, commonly referred to as Cade,[3,4] holds prominence as the primary species used for tar production, followed by *Tetraclinis articulata*, *Cedrus atlantica*, and various Pinus species.[5–14] This study focuses on the essential oils extracted from *Juniperus oxycedrus* and *Cedrus atlantica*, both extensively utilized in Morocco for tar production.

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Citation: Ninich O, El Fahime E, Satrani B, Burri S, Ghanmi M, Aarabi S, Chauiyakh O, Ettahir A. Comparative chemical and Biological Analysis of Wood and Tar Essential Oils from *Cedrus atlantica* and *Juniperus oxycedrus* in Morocco. Trop J Nat Prod Res. 2024; 8(3):6570-6581. https://doi.org/10.26538/tjnpr/v8i3.15

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria

Juniperus oxycedrus, a member of the Cupressaceae family, is indigenous to southern Europe and the Mediterranean basin,[4,15–18] flourishing at altitudes between 1700 and 3000 meters in stony terrains.[3,19] Renowned for its ecological significance, *Juniperus oxycedrus* contributes to the regional economy through diverse applications, including firewood, natural ornamentation, and medicinal purposes. Its essential oil, esteemed for its multifaceted properties, such as antibacterial, antioxidant, and anti-inflammatory effects, has found utility in various domains[3,19], albeit recent concerns have arisen regarding its association with poisoning incidents.[20–25]

Cedrus atlantica, family Pinaceae, is an endemic Moroccan species prized for its essential oil possessing antimicrobial, antifungal, and antioxidant properties.[26,27,36–42,28–35] Widely utilized in construction due to its durability.[30] Cedarwood also holds significance in Morocco's traditional therapeutic practices,[43] where cedar wood tar forms an integral part of the Magico-medical system, [44] utilized for treating skin conditions and eliminating parasites in animals.[32] These traditional forest species, deeply rooted in folk medicine, intrigue researchers for their potential applications. [43,45]

This study presents a comparative analysis of the biological and antioxidant properties of wood tar essential oils extracted from *Juniperus oxycedrus* and *Cedrus atlantica*, using both lab-produced and artisanal samples. By exploring the composition and potential of these oils, the study aims to uncover similarities or distinctions between the two species, thereby contributing novel insights into their utility and applications in various fields.

Material and methods

Plant collection and identification

The artisanal wood tar samples were obtained from local producers located in Talgount (31°40'32.7"N 7°15'58.7"W) in March 2021 and Itzer (32°55'31.8"N 5°11'40.7"W) in June 2021. These producers employed traditional artisanal techniques.[2] On the other hand, the Laboratory wood tar was obtained by subjecting small wood pieces of *C. atlantica* and *J. oxycedrus* to heat within an iron container of "Pyrox" in the process of pyrolysis at 450 °C for 3 to 4 hours.

Essential oils

About 100 g of sawdust and 50 g of wood tar from each sample were subjected to hydrodistillation in a Clevenger-type apparatus. The round bottom flask holding the materials was half filled with distilled water and boiled for 8 hours.[43,46] The essential oil yields were calculated from the weight of the initial samples before distillation.[47]

GC-MS analysis

Chromatographic analyses were conducted using an electronic pressure-regulating gas chromatograph (HP 6890 series) with an HP-5 (5% phenyl-methyl-siloxane) capillary column (30 m×0.25 mm, film thickness: 0.25 µm). Detection utilized a flame ionization detector (FID) (250°C) supplied with a mixture of H_2/air gas and nitrogen served as the carrier gas at a flow rate of 1.7 ml/min. The device featured a split-splitless PVT injector, operating in split mode (split ratio: 1/50, flow rate: 66 mL/min), with 1 µL injected. Temperature programming ranged from 50 to 200°C over 5 minutes, with a gradient of 4°C/min. Control and monitoring were managed by a computer system type "HP ChemStation". Component identification relied on Kováts indices and gas chromatography-mass spectrometry (GC-MS), conducted on a Hewlett-Packard gas chromatograph (HP 6890 series) coupled with a mass spectrometer (HP 5 973 series).[48]

The components were characterized through a comparative analysis of their Kovats indices (KI) and mass spectra against established standards and literature data (Adams). Alkanes within the C9 to C18 range served as reference compounds for calculating KI values [28]. They were determined from the following equation:

$$KI = \left(\frac{TR_x - TR_n}{TR_{n+1} - TR_n} + n\right) \times 100$$

TRx represents the retention time of the compound, while TRn and TRn+1 indicate the retention times of linear alkanes with n and n+1 carbon atoms.

Antimicrobial activity

The antimicrobial activity was evaluated according to the technique of dispersing essential oils in 0.2% agar-agar as described by Mansouri *et al.* [49] and outlined in the literature. [28,37,47,50,51]

The essential oils are first diluted one-tenth in the agar-agar solution. Aliquots of this dilution were added to test tubes containing nutrient agar for bacteria and PDA for molds. They were then sterilized in the autoclave for 20 minutes at 121 °C, cooled to 45 °C, and poured into Petri dishes. The final concentrations of essential oils were 1/100, 1/250, 1/500, 1/1,000, 1/2,000, 1/3,000, and 1/5,000 (v/v). Controls, consisting of culture medium and 0.2% agar-agar solution alone, were also prepared. Inoculation is done by using a platinum loop to take the same volume of microbial culture to streak the Petri dishes. The plates were incubated at 37 °C for 24 hours for bacteria and 25 °C for seven days for fungi. Each assay was repeated three times to minimize experimental error.[49] The study analyzed four bacterial strains (*Bacillus subtilis*, *Escherichia coli*, *Staphylococcus aureus*, and *Micrococcus luteus*) and four wood-rotting fungal species (*Gloeophyllum trabeum*, *Coniophora puteana*, *Poria placenta*, and *Coriolus versicolor*). Minimum inhibitory concentrations (MIC) were determined using a standard method, defined as the concentration at which there is no visible growth on the Petri dishes.[37,47]

Antioxidant activity

DPPH free radical scavenging activity Assay

DPPH free radical scavenging activity of the EOs was evaluated according to the method described by Satrani *et al.*[52–55] The antioxidant capacity (expressed as EC_{50}, i.e., the effective concentration at which the antioxidant activity reaches 50% inhibition of free radicals) of the EOs was evaluated from the linear regression analysis of the percentage absorbance versus concentration of the EOs This parameter serves as a measure of the antioxidant potency of the tested natural extract. [7,15,52,53,55–59]

The Ferric Reducing Antioxidant Power (FRAP) assay

The Ferric Reducing Antioxidant Power (FRAP) assay was conducted according to the method described by Jaouadi *et al.*[48,60] The results were quantified as the effective concentration (EC_{50}) at which the absorbance reached 0.5, determined through linear regression analysis. EC_{50}, in this context, indicates the concentration of the antioxidant necessary to reduce ferric ions to the ferrous form by 50%, reflecting the antioxidant capacity of the sample.[48,58,61,62] Triplicate determination was carried out.

Results and Discussion

The determination of the proximate parameters of herbal products is important in establishing product standards and minimizing adulterations. In this study, the moisture content of the *C. atlantica* wood stumps was 2.40 ± 0.01%, while the EO yield was 4.03%. In contrast, *J. oxycedrus* exhibited a higher wood moisture content of 9.27 ± 0.01%, demonstrating significant moisture compared to *C. atlantica*. Also, the percentage EO yield of 0.26 was lower than that of *C. atlantica*. Indicating that *J. oxycedrus* wood contains lower essential oil content (Figure 1).

The extraction yield of 0.26% for *Juniperus oxycedrus* tar appears relatively low when compared to other studies. Higher yields have been reported by different authors, 1.703%, 1.20%, 11.0% and 2.0%, respectively.[64,65,66,67] However, Loizzo *et al.* reported 0.68% yield.[15] The yield of essential oil extraction from *Cedrus atlantica* wood was 4.03%, surpassing some previously reported results in the literature.[39,40] Chalchat et *al.* (1994) reported lower yields of 0.7% when using cedar wood sawdust.[68] Similarly, Satrani et *al.* (2006) obtained yields of 2.78% [47] and 0.05%.[69] Fidah *et al*. (2016) with 3.35%[70] and Al Kamaly *et al*. (2022) with 3.84%.[29]

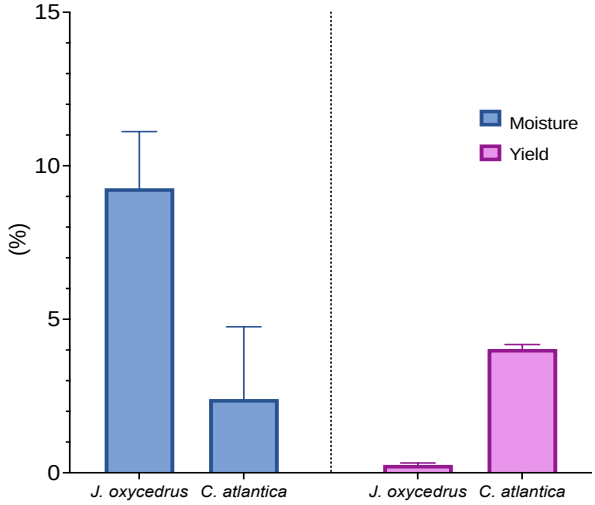

Figure 1: moisture and yield percentage (%)

Comparatively, there was a significant difference in both moisture content and EO yield of *J. oxycedrus* and *C. atlantica*. These results indicate that high moisture levels in the wood can significantly affect the volume of essential oil extracted, thereby influencing the extraction yield. This observation aligns with a study conducted by Derriche et *al.*, which highlights the role of plant nature and physiology in determining the quantity of extracted essential oils.[63] However, further studies are required to confirm this correlation.

Notably, some studies have reported much higher yields (9.19%) for *C. atlantica* from the Azrou region of Morocco.[37] In comparison, the result falls in the middle range of other studies, of 3.51% to 5.98% reported by Jaouadi *et al*. for sawdust of *C. atlantica.*[48] The variations in essential oil extraction yields can be attributed to different factors, including the extraction technique, wood origin, [46,71] wood quality, [71,72] the sanitary condition of the wood,[73] and the region's bioclimatic conditions.[71,72]
With respect to the wood tar EOs, *C. atlantica* gave an oil volume of 6.2 mL from its artisanal wood tar, while the laboratory wood tar was 12.5 mL. *J. oxycedrus* artisanal wood tar yielded 4 mL of essential oil, while the laboratory wood tar produced 6.6 mL (Figure 2).

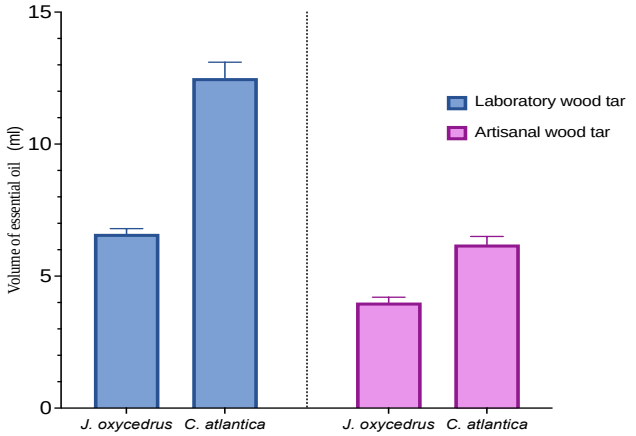

Figure 2: Volume of essential oil of artisanal and laboratory tar (ml)

The results indicate a significant difference in the volume of essential oil obtained between the laboratory tar and artisanal tar. For *C. atlantica*, the volume of essential oil extracted from the laboratory tar was almost double that obtained from the artisanal tar, measuring 12.5 mL compared to 6.2 mL. Similarly, for *J. oxycedrus*, the volume of essential oil obtained from the laboratory tar was also higher than that from the artisanal tar, with 6.6 mL compared to 4 mL. These findings highlight the influence of the wood tar extraction method and the specific species involved on the volume of essential oil obtained. Additionally, it's worth noting that the yields of essential oils from wood tar can be subject to variations based on factors such as the age of the species, the duration, and the method of extraction, as well as the plant part used in the process.[64] The following table presents the chemical composition of six essential oils analyzed (Figure 3 and Table 1).
The essential oils are Artisanal Cedar tar (ACTEO), Laboratory cedar tar (LCTEO), Cedar (CEO), Artisanal juniper tar (AJTEO), Laboratory Juniper tar (LJTEO), and Juniper (JEO). The analysis identified 57 phytochemicals for ACTEO, 55 for LCTEO, 66 for CEO, 76 for AJTEO, 35 for LJT EO, and 30 for JEO. All cedar oil samples exhibited the presence of 28 common compounds, which collectively accounted for 74.76%, 72.59%, and 67.06% in ACTEO, LCTEO, and CEO, respectively. These shared compounds encompass a variety of terpenes, including sabinene, α-phellandrene, 9-epi-(E)-caryophyllene, and β-cedene. Monoterpenols such as cis-carveol and α-terpinene-7-al were also identified, alongside sesquiterpenes like (α-; δ-; γ-) cadinene, γ-gurjunene , and allo-aromadendrene. Esters such as cedryl acetate and trans-carvyl acetate were also present. The cedar oil samples additionally contained various other compounds. The cedar essential oil samples exhibited distinct molecules that stood out in percentage composition. Notably, three molecules demonstrated prominence: α-Himachalene, γ-Gurjunene, and β-himachalene. α-himachalene displayed varying percentages of 16.06%, 14.23%, and 11.36%. Similarly, γ-Gurjunene was present at percentages of 9.69%, 7.88%, and 8.88%. Moreover, β-Himachalene exhibited rates of 30.24%, 22.78%, and 26.74% across ACTEO, LCTEO, and CEO, respectively. Some molecules were detected as traces, while others were exclusive to ACTEO and LCTEO. For instance, the ACTEO revealed 0.33% terpinolene, while the LCTEO contained 0.83% Cedrane, 0.74% ρ-Cymen-9-ol, and 2.63% Octanol acetate.
common molecules. These molecules collectively constituted 49.63%, 59.81%, and 40.34% of AJTEO, LJTEO, and JEO, respectively. The most significant molecules in AJTEO were α-Cedrene (32.85%), followed by neoiso-3-Thujanol acetate (9.75%) and Terpinen-4-ol (7.06%). Similarly, LJTEO prominently featured α-Cedrene (27.89%), Terpinen-4-ol (10.13%), and cis-Thujopsadiene (9.55%). In JEO, key molecules were Cedrol (19.54%), cis-Thujopsadiene (15.99%), and 5-Cedranone (15.29%). Additionally, laboratory juniper tar oil contained 0.92% neo-3-Thujanol and 1.21% Cedril methyl ketone. Notable molecules in JEO encompassed Germaerene D (0.9%), β-Atlantane (0.62%), 8-Cedren-13-ol (0.75%), (2Z,6Z)-farnesol (1%), and Cedryl acetate (1.1%). AJTEO exclusively contained Fenchone (0.51%), Vestitenone (0.35%), and α-Muurolene (2.76%), while others were present at lower percentages. Interestingly, a single molecule, δ-cadinene, was detected across all six essential oil samples, with varying percentages: 1.7% for ACTEO, 2.99% for LCTEO, 2.22% for CEO, 0.42% for AJTEO, 0.76% for LJTEO, and 1.01% for JEO. Analyzing *Juniperus oxycedrus* essential oil samples unveiled eleven Numerous studies have delved into the composition of cedar essential oil, revealing a distinct array of molecules. These include (α, β, γ) himachalane, [29,32,37,39,47,71,73] cedrol, [39] isocedranol, [39,47] himachalol, [47,69] I-epicubenol, [47] α-pinene, [39,69] Zα-Atlantone, [47,70] E-γ-Atlantone, [70] E-α-Atlantone, [32,37,47,70,71,73] 5-isocedrano, [70] 9-iso-thujopsanone, [70] cedranone, [70] 8-Cedren-13-ol, [37] cedroxide, [37] deodarone, [37,47,74] α-cedrene, [75] β-cedrene, [75] isovanillincamphor, [75] β-caryophyllene , [69,75] caryophyllene oxide, [69,75] p-cresol, [75] α-humulene, [69] and guaiacol. [75]

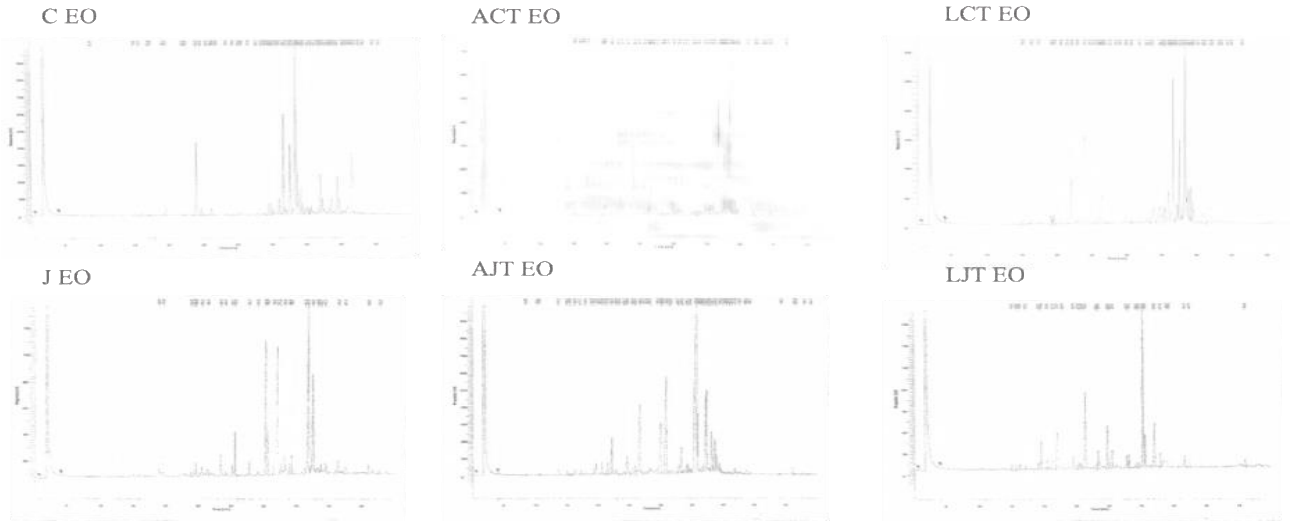

Figure 3: Chromatogram of gas chromatography analysis of essential oils of *Cedrus atlantica* and *Juniperus oxycedrus*

Table 1: Chemical composition of essential oils of *Cedrus atlantica* and *Juniperus oxycedrus*

IK	Compound Name	ACT EO	LCT EO	C EO	AJT EO	LJT EO	J EO
815	(2E)-Octene	-	-	-	0.04	-	-
824	Cyclohexane	-	-	0.09	-	-	-
836	Isopropyl butanoate	-	-	-	0.17	-	-
844	(3E)-Hexenol	-	-	-	0.06	-	-
871	Methyl 2-methylpentanoate	-	-	-	0.25	-	-
908	Isobutyl isobutyrate	-	-	-	0.35	-	-
921	tricycle	0.28	0.31	0.24	0.05	0.93	-
924	α-thujene	-	-	-	0.08	-	-
932	α-pinene	0.09	-	0.08	0.25	0.43	-
946	camphene	0.07	0.33	-	-	0.58	-
953	thuja-2,4(10)-diene	-	-	-	0.04	-	-
959	n-heptanol	-	-	-	0.25	-	-
961	verbenene	0.05	-	0.05	-	-	-
969	sabinene	0.15	0.19	0.12	-	0.47	-
974	β-pinene	-	-	-	0.29	-	-
995	isobutyl-(2E)-butenoate	-	-	-	0.09	-	-
1001	δ-2-carene	-	-	-	0.41	-	0.52
1002	α-phellandrene	0.44	0.84	0.16	0.17	-	-
1008	δ-3-carene	0.17	0.36	0.11	0.81	0.84	-
1014	α-terpinene	1.24	0.95	0.62	-	0.77	-
1020	ρ-cymene	-	-	-	0.29	4.19	1.68
1032	(Z)-β-ocimene	0.4	0.26	-	0.89	1.01	-
1044	(E)-β-ocimene	-	-	-	0.12	-	-
1054	γ-terpinene	0.51	0.39	-	0.89	1.33	-
1065	cis-sabinene hydrates	2.36	3.87	0.08	-	-	-
1071	ρ-Cresol	-	-	-	2.79	4.9	-
1078	Camphenilone	-	-	0.06	-	-	-
1083	Fenchone	-	-	-	0.51	-	-
1086	Terpinolene	0.33	-	-	-	-	-
1095	6-Camphenone	-	0.24	-	0.06	0.78	-
1104	α-fenchocomphorone	-	0.28	-	-	-	-
1111	6-camphenol	9.32	-	7.78	-	-	-
1112	trans-thujone	-	-	-	0.22	-	0.33
1119	trans-pinene hydrate	-	-	-	0.06	-	0.65
1128	allo-ocimene	-	-	-	1.55	2.95	1.4
1130	1-terpineol	1.97	-	0.92	-	-	-
1143	trans-dihydro-α-terpineol	0.18	7.86	-	-	-	-
1147	neoiso-3-thujanol	-	-	-	0.83	0.84	1.27
1149	neo-3-thujanol	-	-	-	-	0.92	-
1156	cis-dihydro-β-terpineol	-	-	0.12	-	-	-
1158	m-cresol acetate	-	-	-	0.81	0.92	-
1159	trans-β-Terpineol	0.27	-	-	-	-	-
1162	δ-Terpineol	1.41	-	0.53	-	-	-
1174	Terpinen-4-ol	3.51	-	0.12	7.06	10.13	0.47
1179	ρ-Cymen-8-ol	0.36	1.35	-	0.31	-	-

1186	α-Terpineol	-	0.28	0.08	0.07	-	-
1197	Verbanol	-	-	-	0.15	-	-
1199	γ-Terpineol	0.1	0.19	-	-	-	-
1204	ρ-Cymen-9-ol	-	0.74	-	0.2	-	-
1211	Octanol acetate	-	2.63	-	-	-	-
1215	transCarveol	0.14	0.58	-	0.38	0.72	-
1219	cis-Sabinene Hydrateacetate	-	-	-	0.19	-	-
1226	cis-Carveol	0.41	0.26	0.33	-	3.33	3.45
1239	Carvone	0.15	0.3	0.04	-	-	-
1244	Car-3-in-2-one	-	-	-	1.1	-	0.33
1255	Carvenone	1.48	0.14	-	-	-	-
1259	cis-Carvoneoxide	-	-	-	4.41	6.01	-
1267	iso-3-Thujanolacetate	-	-	-	0.2	0.72	1.29
1273	trans-Carvone oxide	-	1	0.2	-	-	-
1273	neo-3-Thujanol acetate	-	-	-	9.75	2.44	4.14
1283	α-Terpinen-7-al	0.64	0.23	0.16	-	-	-
1295	3-Thujanol acetate	-	-	-	0.26	-	-
1299	Terpinen-4-ol acetate	0.14	-	-	0.21	-	-
1305	ρ-cresyl isobutyrate	-	-	-	0.15	-	-
1306	Dihydro-carveol acetate	-	-	0.07	-	-	-
1308	iso-Verbanol acetate	-	-	-	0.25	-	-
1311	cis-Pinocarvyl acetate	0.19	0.47	-	-	-	-
1328	neo-iso-Verbenol acetate	-	-	-	0.22	-	-
1335	δ-Elemene	-	-	-	0.97	2.16	1.26
1339	Trans-Carvyl acetate	0.11	0.35	0.32	-	-	-
1345	α-Cubebene	0.31	0.2	-	1.94	1.83	-
1361	(Z)-β-Damoscenone	0.72	1.33	0.14	0.1	-	-
1365	cis-Carvylacetate	0.17	2.36	-	-	-	-
1371	Longicyclene	-	-	0.2	0.62	-	-
1374	α-Copaene	-	-	-	0.69	0.38	0.25
1387	β-Cubebene	0.57	-	1.12	0.24	0.56	-
1389	iso-Longifolene	1.6	2.36	1.2	0.2	-	-
1400	β-Longipinene	0.93	1.37	0.5	-	-	-
1407	Longifolene	0.69	1.48	-	-	-	-
1410	α-Cedrene	-	1.07	-	32.85	27.89	13.85
1419	β-Cedrene	1.89	1.28	0.28	3.51	4.85	4.5
1429	cis-Thujopsene	-	-	-	0.14	-	-
1431	β-Gurjunene	1.44	2.93	-	-	-	-
1434	γ-Elemene	-	-	1.68	0.15	-	-
1441	Cedrane	-	0.83	-	-	-	-
1444	Vestitenone	-	-	-	0.35	-	-
1447	epi-Cedrane	-	-	-	0.34	-	0.75
1449	α-Himachalene	16.06	14.23	11.36	-	-	-
1454	(E)-β-Farnesene	-	-	0.72	-	-	-
1458	Allo-Aromadendrene	0.32	0.25	0.72	5.99	-	-
1464	9-epi-(E)-Caryophyllene	0.58	1.71	0.56	-	-	-
1465	cis-Thujopsadiene	-	-	-	4.74	9.55	15.99

1475	γ-Gurjunene	9.69	7.88	8.88	0.07	-	0.82
1478	γ-Muurolene	-	-	-	0.12	-	-
1481	γ-Himachalene	0.09	0.35	0.47	-	-	-
1484	Germaerene D	-	-	-	-	-	0.9
1489	β-selinene	-	-	-	2.7	1.81	-
1495	cis-Cadina-1,4-diene	-	-	-	0.29	0.22	1.28
1500	β-himachalene	30.24	22.78	26.74	-	-	-
1500	α-Muurolene	-	-	-	2.76	-	-
1508	Germacrene A	0.64	-	2.84	1.27	-	-
1513	γ-Cadinene	1.93	2.63	2.38	-	-	-
1522	δ-Cadinene	1.7	2.99	2.22	0.42	0.76	1.01
1530	γ-dihydro-ar-Himacholene	0.67	1.42	0.89	-	-	-
1533	trans-Cadina1.4diene	-	-	-	0.16	0.78	1.6
1537	α-Cadinene	0.36	0.97	0.47	-	-	-
1541	8.14-Cedranoxide	-	-	0.36	-	-	-
1544	α-Colacorene	-	-	-	0.03	-	0.53
1559	Germacrene B	-	0.26	1.32	0.1	-	-
1574	α-Cedrene epoxy	-	-	0.39	-	-	-
1574	Germacrene D-4-ol	-	-	-	0.11	-	-
1577	trans-Sesquisabinene hydrat	-	-	-	0.08	-	-
1578	Epoxy-Himachalene	0.41	0.89	0.46	-	-	-
1587	Davanone	-	-	-	0.03	-	-
1589	Allo-cedrol	-	0.19	0.1	-	-	-
1600	Cedrol	-	-	0.42	0.42	1.81	19.54
1612	cis-Isolongifolamone	0.51	0.9	3.57	-	-	-
1615	β-Himachalene oxide	0.18	0.73	-	-	-	-
1618	epi-Cedrol	-	-	-	0.18	-	-
1626	2-epi-α-Cedrene-3-one	-	-	1.31	-	-	-
1628	5-Cedranon	-	-	-	-	0.95	15.29
1639	1.7-di-epi-α-cedrenal	-	-	0.11	-	-	-
1649	β-Eudesmol	0.19	0.19	-	-	-	-
1652	Himachalol	-	-	1.12	-	-	-
1652	α-Cadinol	-	-	-	0.07	-	1.02
1661	Allohimachalol	0.09	0.27	-	-	-	-
1661	Dihydroeudesmol	-	-	1.61	-	-	-
1666	14-hydroxy-(Z)-Caryophyllene	-	-	-	0.25	-	-
1668	β-atlantone	-	-	-	-	-	0.62
1675	Cadalene	0.27	-	0.22	0.05	-	-
1688	Cedr-8-en-13-ol	-	-	-	-	-	0.75
1694	(Z)-γ-atlantone	0.59	1.23	3.97	-	-	-
1698	Deodarone	0.15	-	2.2	-	-	-
1698	(2Z,6Z)-Farnesol	-	-	-	-	-	1
1706	(E)-γ-atlantone	-	0.16	-	-	-	-
1713	Cedroxide	-	-	-	-	-	-
1717	(Z)-α- atlantone	-	-	0.32	-	-	-
1728	Longifolol	-	-	0.18	-	-	-
1741	Cedr-8(15)-en-9α-ol	-	-	0.34	-	-	-

1767	Cedryl acetate	0.52	0.36	0.07	-	-	1.1
1775	Cedril methyl ketone	-	-	-	-	1.21	-
1777	(E)-α atlantone	-	-	6.02	-	-	-
-	NI	-	-	0.11	-	-	-
-	NI	-	-	0.16	-	-	-
-	NI	-	-	0.05	-	-	-
-	NI	-	-	0.05	-	-	-
-	NI	-	-	0.06	-	-	-
-	NI	-	-	0.05	-	-	-

* KI: Kováts indice ; **NI: Not identified

In a Slovakian study, the primary constituents in commercially available cedar essential oil were identified as δ-cadinene, (Z)-β-farnesene, β-himachalene, viridi-florol, and himachala-2,4-diene.[76] Many authors have also identified (α, β) himachalenes, methyl-1,4-cyclohexadiene, [12,43] α-cedrene, [77] trans-cadina-1(6),4-diene, [12] 6-camphenol,[12] and sabinene hydrate [12] as the significant constituents in Atlas cedar tar essential oil.

Several studies exploring *Juniperus oxycedrus* essential oil have unveiled its intricate chemical composition. This complexity is underscored by identifying diverse compounds, including isovanillin, (-)-a-cedrene, camphor, b-caryophyllene, caryophyllene oxide, p-cresol, and guaiacol.[75] Furthermore, the distillation of oil from *J. oxycedrus* wood reveals a range of components such as cubenene, α-copaene, β-caryophyllene, thujopensene, α-humulene, γ-cadinene, calamenene, calacorene, 1,6-dimethyl naphthalene, 2,3-dimethyl-5-methoxy phenol, elemol, epicubenol, and cubenol.[78] Alcohols, including cedrol, T-muurolol, α-muurolol, α-cadinol, and the intermediate compound 1,6-dimethyl-4-ispopropyl naphthalene intermediol, alongside eudesmol, cis-caryophyllene epoxide, and calamenenol, are also present.[78] Juniper tar is characterized by β-cedrene and cis-thujopsene.[43] The dominant constituents include *δ*-cadinene, γ2-cadinene, calamenene, and α -humulene, and caryophyllene oxide.[67] *J. oxycedrus* EO derived from aerial parts contains compounds such as α-pinene and β-cedrene and phenols like guaiacol and cresol.[15,79] The tar of *J. oxycedrus* contains α-pinene, γ-cadinene, and germacrene D.[65] Notably, the EO displays a higher concentration of alcohols compared to the oil distilled from empyreumatic (tar) oil.[78] Furthermore, the chemical compositions of *J. oxycedrus* essential oils vary across different regions, resulting in distinct majority compounds.[52,80]

The study aligns with the findings documented in existing literature. Previous research has identified key components within *C. atlantica* essential oils, including himachalenes, atlantones, cedrene, α-pinene, β-caryophyllene, α-humulene, caryophyllene oxide, himachalol, and various other compounds. However, the distribution of these major constituents varies across different samples. This variance in chemical composition and constituent percentages can be attributed to several factors, such as the specific plant part subjected to extraction, geographical location, harvesting timeframe, plant age, and extraction methods.[16,28,29,35,40,41,47,78,81] Wood tars' chemical composition is influenced by many factors, encompassing the type of plant tissue involved. Notably, components like lignin, cellulose, and hemicellulose play a significant role in shaping the chemical composition of tars. [82–84] Additionally, distinctions arise from different tree sections and extraction processes, [45] as well as the botanical origin of the plant itself. [77]

The results of the antimicrobial activities of the EOs are shown in Table 2. The results revealed that at 1/100 v/v, all samples inhibited bacteria and fungi. ACTEO inhibited all bacteria and *C. versicolor* and *P. placenta* at 1/500 v/v, and *G. trabeum* and *C. puteana* at 1/1000 v/v. LCTEO inhibited *B. subtilis* and *P. placenta* at 1/500 v/v, others at 1/1000 v/v. CEO had weaker inhibition, requiring 1/500 v/v for four bacteria and *C. puteana* and 1/250 v/v for *G. trabeum*, *C. versicolor*, and *P. placenta*. The samples exhibited significant antimicrobial activity against various tested microorganisms, although resistance levels varied. CEO, particularly effective against *G. trabeum*, had a lower minimum inhibitory concentration (MIC) of 1/250 v/v in the current study, compared to 1/1000 v/v in a prior investigation. In this study, *C. puteana* was inhibited at 1/500 v/v, while the earlier study reported inhibition at 1/400 v/v.[70] Saab *et al*. (2018) reported robust antimicrobial activity of cedar essential oil against *E. coli, S. aureus, M. luteus*, and *B. subtilis.* Notably, *S. aureus* exhibited greater resistance to inhibition compared to *B. subtilis* and *E. coli.*[35] The study observed similar inhibition across all strains, with a MIC of 1/500 v/v. Another study demonstrated that *C. atlantica* essential oil effectively inhibited *M. luteus* and *Penicillium spp.*[76] Cedarwood essential oil's major components are primarily responsible for its antimicrobial activity,[35,43,77,84] especially against Gram-positive bacteria. Monoterpenes, hydrocarbons, and terpinenes have exhibited moderate to strong antimicrobial properties. [35] In a study conducted in the Middle Atlas region, *C. atlantica* essential oil displayed antimicrobial effects against *E. coli*, *B. subtilis*, and *B. cereus*, with MIC values ranging from 0.2 to 0.4 µL/mL. [37] This antibacterial effect is primarily attributed to terpenes with aromatic rings and phenolic hydroxyl groups, which can form hydrogen bonds with target enzymes. Other terpenes, alcohols, aldehydes, and esters may also contribute to its antimicrobial properties.[37,47,85,86] Terpenes and terpenoids are pharmacologically important molecules with hydrophobic properties that can disrupt the membranes of human-pathogenic fungi, leading to cell death. [47,86,87] The lipophilic hydrocarbon skeleton and hydrophilic functional groups play a crucial role in their antimicrobial action.[85] Himachalene and atlantone have been reported to possess therapeutic activities in cancer, microbial infections, and neuroprotection. [88] Cedarwood essential oils exhibit antimicrobial and antifungal activity due to their diverse chemical composition and synergistic interactions.[47,86] The effectiveness varies with different bacterial strains and oil compositions. The health of cedar trees affects oil quality.[73] However, recent research in 2021 conducted by Ez-Zriouli *et al*. demonstrated significant antimicrobial activity of *C. atlantica* essential oil, surpassing some synthetic antibiotics against various bacterial and yeast strains.[86]

In Table 3, the results of antimicrobial tests on juniper essential oils samples are presented. At a 1/100 v/v concentration, all microorganisms were inhibited. However, AJTEO inhibited some microorganisms at 1/1000 v/v and others at 1/2000 v/v. LJTEO showed similar results. JEO inhibited some microorganisms at 1/500 v/v and others at 1/1000 v/v. Evidently, certain microorganisms exhibit higher susceptibility compared to others across the various samples tested. In a broader assessment, juniper wood tar essential oils samples showed better antimicrobial activity compared to wood essential oils in terms of their efficacy in inhibiting bacterial growth. This study showed that *J. oxycedrus* samples have antimicrobial properties against various microorganisms. Active extracts inhibited the growth of several microbes at different concentrations. Notably, the essential oil of *J. oxycedrus* showed antimicrobial activity against *S. aureus* at varying minimum inhibitory concentrations, with AJT, AJTEO, and JEO at an MIC of 1/2000 v/v and LJTEO at an MIC of 1/3000 v/v. Marongiu *et al*. (2003) found that supercritical carbon dioxide-extracted *J. oxycedrus* wood oil had a different chemical composition with a reduced level of sesquiterpene hydrocarbons and an increased amount of oxygenated sesquiterpenes properties that showed no antibacterial activity.[66]

Table 2: Results of the antimicrobial activity of *C. atlantica* esseantial oils (EO) samples Dilution (v/v)

	1/100 (v/v)			1/250 (v/v)			1/500 (v/v)			1/1000 (v/v)			1/2000 (v/v)			1/3000 (v/v)			1/5000 (v/v)		
	ACT EO	LCT EO	C EO	ACT EO	LCT EO	C EO	ACT EO	LCT EO	C EO	ACT EO	LCT EO	C EO	ACT EO	LCT EO	C EO	ACT EO	LCT EO	C EO	ACT EO	LCT EO	C EO
E.coli	-	-	-	-	-	-	M	-	M	+	M	+	+	+	+	+	+	+	+	+	+
B. subtilis	-	-	-	-	-	-	M	M	M	+	+	+	+	+	+	+	+	+	+	+	+
S. aureus	-	-	-	-	-	-	M	-	M	+	M	+	+	+	+	+	+	+	+	+	+
M. luteus	-	-	-	-	-	-	M	-	M	+	M	+	+	+	+	+	+	+	+	+	+
G.trabeum	-	-	-	-	-	M	-	-	+	M	M	+	+	+	+	+	+	+	+	+	+
C.versicolor	-	-	-	-	-	M	M	-	+	+	M	+	+	+	+	+	+	+	+	+	+
C.puteana	-	-	-	-	-	-	-	-	M	M	M	+	+	+	+	+	+	+	+	+	+
P. placenta	-	-	-	-	-	M	M	M	+	+	+	+	+	+	+	+	+	+	+	+	+

*(-): Inhibition; (+): Growth; M: Minimum Inhibitory Concentration. ; ACT EO : Artisanal *Cedar tar* Essential oil ; LCT EO : Laboratory *Cedar tar* Essential oil ; C EO : *C. atlantica* Essential oil

Table 3: Results of antimicrobial activity of *J. oxycedrus* samples Dilution (v/v)

	1/100 (v/v)			1/250 (v/v)			1/500 (v/v)			1/1000 (v/v)			1/2000 (v/v)			1/3000 (v/v)			1/5000 (v/v)		
	AJT EO	LJT EO	J EO	AJT EO	LJT EO	J EO	AJT EO	LJT EO	J EO	AJT EO	LJT EO	J EO	AJT EO	LJT EO	J EO	AJT EO	LJT EO	J EO	AJT EO	LJT EO	J EO
E.coli	-	-	-	-	-	-	-	-	-	-	-	-	M	-	M	+	M	+	+	+	+
B. subtilis	-	-	-	-	-	-	-	-	-	-	-	-	M	-	M	+	M	+	+	+	+
S. aureus	-	-	-	-	-	-	-	-	-	-	-	-	M	-	M	+	M	+	+	+	+
M. luteus	-	-	-	-	-	-	-	-	M	M	-	+	+	-	+	+	M	+	+	+	+
G.trabeum	-	-	-	-	-	-	-	-	-	M	-	M	+	M	+	+	+	+	+	+	+
C.versicolor	-	-	-	-	-	-	-	-	M	M	-	+	+	M	+	+	+	+	+	+	+
C.puteana	-	-	-	-	-	-	-	-	M	M	-	+	+	-	+	+	M	+	+	+	+
P. placenta	-	-	-	-	-	-	-	-	M	-	-	+	M	M	+	+	+	+	+	+	+

*(-): Inhibition; (+): Growth; M: Minimum Inhibitory Concentration.; *AJT* EO: Artisanal *Juniper tar* Essential oil; LJT EO : Laboratory *Juniper tar* Essential oil ; J EO : *J. oxycedrus* Essential oil

The oil extracted from the *Juniperus oxycedrus* in the Ait Bouguemez region is rich in monoterpenes and possesses significant antibacterial properties.[80] The complexity of *J. oxycedrus* oil attributes its activity to a combination of phytochemicals.[89] Okut *et al*. (2018) noted antibacterial activity in *J. oxycedrus* leaf oil.[90] Johnson (2001) observed antibacterial activity in *J. oxycedrus* tar but not against *Salmonella typhosa* and *Proteus morgani*.[5] It could be opined that the complexity of essential oils' chemical composition of these tars contributes to their activity.[43] The results of the antioxidant activity evaluation for the samples are presented in Figure 4. The DPPH assay of cedar essential oils demonstrates varying levels of antioxidant activity, ranging from 15% to 50%. CEO exhibits the lowest activity (> 20 µg/mL), ACTEO shows a moderate level (17.659 µg/mL), and LCTEO is the most active (16.09 µg/mL). On the other hand, juniper essential oils display a wider range of activity, spanning from 25% to 70%. JEO exhibits the lowest activity (19.253 µg/mL), AJTEO and LJTEO demonstrate a strong antioxidant effect (< 1.25 µg/ml). Notably, LJCEO exhibits the most impressive overall antioxidant capacity. Shifting to the FRAP activity curves measured at 700 nm, they reveal optical density (OD) values ranging from 0.1 to 0.9 for both species. Artisanal and laboratory tar essential oils exhibit higher OD values.

Also, in the FRAB assay, *Cedrus atlantica* essential oils, show EC_{50} values as follows: ACTEO (532.71 ± 0.053 µg/mL), while LCTEO exhibits a lower EC_{50} of 395.66 ± 0.02 µg/mL. Conversely, CEO demonstrates the highest EC_{50}, with a value of 1027.7 ± 0.037 µg/mL, indicating the lowest antioxidant activity among the three samples. These results indicate that LCTEO possesses the most potent antioxidant activity, followed by ACTEO, while CEO exhibits the weakest antioxidant potential. The essential oils extracted from *Juniperus oxycedrus* exhibited the following EC_{50} values: AJTEO (293.3 ± 0.012 µg/mL), LJTEO (238.12 ± 0.014 µg/mL), and JEO (541.19 ± 0.061 µg/mL). The results showed that LJTEO demonstrated potent antioxidant activity.

Comparisons were made between cedar essential oil samples and previous literature findings on antioxidant activity. The results align with other studies demonstrating significant antioxidant activity.[76] For instance, Jaouadi *et al*. showed a radical scavenging activity of cedar essential oils, with EC_{50} values of 15.559 ± 0.715 mg/mL and 16.264 ± 0.285 mg/mL for cedarwood essential oils.[48] These findings suggest potential industrial applications for cedar essential oil due to its strong antioxidant properties.[52,59] The antioxidant activity of *J. oxycedrus* was also compared with existing literature, revealing significant variability. In Morocco, Satrani *et al.* reported a EC_{50} of 4.90 µg/ mL for the essential oil of *J. oxycedrus* twigs, while in Lebanon, they found an EC_{50} of 7.42 µg/ mL for the essential oil of *J. oxycedrus* fruits. [52] Findings align with El Jemli's results, showing an essential oil's antioxidant capacity with an EC50 of 26.91×10^3 ± 0.13 µg/ mL.[7] In Lebanon, Loizzo *et al.* reported an EC_{50} of 1.45±0.05 µl/ mL for oil from juniper wood.[15] The study revealed a higher FRAP antioxidant potential in juniper essential oils when compared to the values reported in other studies.[7,15,48,52] The essential oil samples demonstrate promising antioxidant properties, likely attributed to their chemical composition.[17,43,52,91] However, given the intricate composition of essential oils, elucidating the precise active compounds presents a challenge, primarily due to potential synergistic interactions among these compounds. For instance, while *J. oxycedrus* essential oil samples exhibit potential as natural antioxidants, it is imperative to identify the specific compounds responsible for this activity. Future research

avenues could involve investigating the individual contributions of various compounds, exploring potential synergistic effects, and assessing how different extraction methods may influence the bioactivity of these oils.

Conclusion

The studies conducted on essential oils derived from the wood and wood tar of *Cedrus atlantica* and *Juniperus oxycedrus* have revealed the complexity of their chemical composition. This complexity varies based on several factors, including extraction techniques and the species used. The experimental findings demonstrate that, overall, the laboratory-derived wood tar essential oils exhibited greater efficacy for both species compared to both artisanal wood tar and wood essential oils. Specifically, *Juniperus oxycedrus* displayed significantly stronger bioactivities than *Cedrus atlantica*. These results suggest a noteworthy difference in the effectiveness of essential oils derived from different sources, highlighting the potential importance of considering the origin and production methods when evaluating their biological activities. Future studies will focus on evaluating the effects of time and environmental stressors on the chemical composition and biological functionality of these species.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

Acknowledgments

The author sincerely thanks the research team of the National Agency for Water and Forests - Rabat and the National Center for Scientific and Technical Research (CNRST) of Morocco for granting access to the technical facilities of the UATS Division. Special thanks are also due to Mr. Said, the Responsible of the Talgount cooperative, and Mr. Moulay Ahmed, the Responsible of the Itzer cooperative.

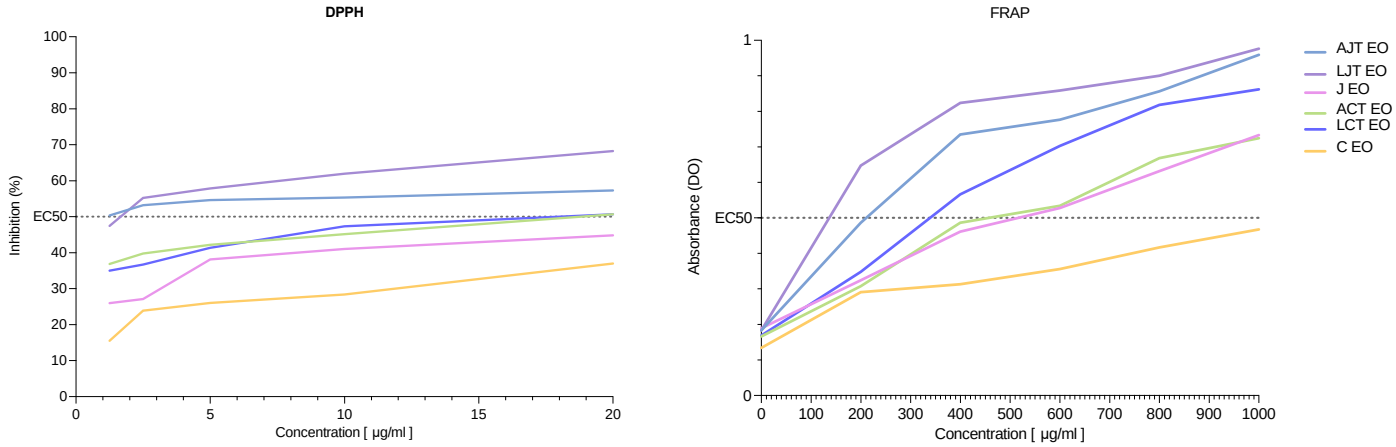

Figure 4: Representative curves of antioxidant activity.

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