

Research Article

Phytochemical Screening and Antioxidant Activity of *Artemisia Herba-Alba* and *Olea Europaea* L. Leaf Extracts Growing in the North-East of AlgeriaWIDAD FATMI ^{1*}, YUVA BELLIK ¹, NASREDDINE MEKHOUKH ¹, YASMINA SOUAGUI ¹, TAQIYEDDINE BENSOUILAH ², HASSINA GUERGOUR ¹¹ Laboratory of Health and Environment, Faculty of Nature and Life Sciences, University El-Bachir El-Ibrahimi, Bordj Bou Arreridj, 34000, Algeria.² Laboratory of Biology, Water and Environment, Faculty of Nature and Life Sciences, University May 8, 1945, Guelma, 24000, Algeria.**ARTICLE DETAILS***Article history:*

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ABSTRACT

As part of the valuation of local medicinal plants, we are interested in the study of the physicochemical and phytochemical composition, and the antioxidant properties of different extracts (acetonic, ethanolic and methanolic extracts) prepared from the leaves of *Artemisia herba-alba* and *Olea europaea* L. The physicochemical analysis of the plants powder revealed that *Artemisia herba-alba* contained approximately 7.86 % water, 5.99 % mineral salts, and 4.94 % fat, while *Olea europaea* L. contained 8.23% water, 3.51% mineral salts, and 2.81% fat. The results showed that the methanolic extract of *Artemisia herba-alba* exhibited the highest level of phytochemicals (24.8 %) whereas for *Olea europaea* L. it was the ethanolic extract which showed substantial level of bioactive compounds (37.65 %). The highest concentrations of polyphenol, flavonoid, tannin and chlorophyll pigments were obtained with acetonic extracts for both studied plants. The strongest anti-free radical activity with DPPH, reducing power, and total antioxidant capacity (TAC) were obtained with the acetonic extract for *Artemisia herba-alba* (IC₅₀ = 0.611 mg/mL, EC₅₀ = 5.03mg/mL, TAC = 97.91µg AAE/mg dw), and methanolic extract for *Olea europaea* L. (IC₅₀ = 0.56 mg/mL, EC₅₀ = 0.83 mg/mL, TAC = 150.49 µg AAE/mg dw).

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INTRODUCTION

The increasing interest in the beneficial effects of aromatic and medicinal plants, applied as herbal remedies to treat or prevent diseases or to promote health, has resulted in numerous research studies directed at determining their levels of phytochemicals and their specific health benefits [1]. Phytochemicals have been emerged as a potential source of natural antioxidants [2]. Most antioxidants isolated from higher plants are phenolic compounds. The goal of phytotherapy is to replace synthetic antioxidants in foods and pharmaceutical preparations. Furthermore, phenolic compounds are involved in the prevention of various diseases associated with oxidative stress, such as cancers and cardiovascular diseases [3].

The Mediterranean flora is known for its richness and abundance of therapeutic plants [4]. Among these medicinal plants, *Artemisia herba-alba* and *Olea europaea* L. which are widely used in traditional medicine thanks to their benefits as remedies for metabolic, respiratory, cardiovascular, spasmodic, inflammatory, eyesore and cancer diseases. A preliminary ethnobotanical survey conducted in the Bordj Bou Arreridj region revealed that these two plants are widely used by diabetics.

Artemisia herba-alba is an aromatic and medicinal herb of the Asteraceae family, from Middle East and North Africa. Previous phytochemical studies have revealed that the *Artemisia* genus is rich in polyphenols, flavonoids, tannins, and essential oils [3]. Particularly, *Artemisia herba-alba* has been shown to have a favourable antioxidant, antihepatotoxic, choleric, spasmolytic,

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anthelmintic, antiphlogistic, antibiotic or antimicrobial activities [5].

On another hand, the olive tree, *Olea europaea* L., belongs to the Oleaceae family [6]. The leaves contain high levels of bioactive compounds, particularly phenolic compounds, with diverse biological properties such as antioxidant, antimicrobial, anti-viral and anti-inflammatory effects [7].

The aim of this study is to determine the phytochemical compounds of the leaves extracts of *Artemisia herba-alba* and *Olea europaea* L. Moreover, the study aims at evaluating their *in vitro* antioxidant capacity through different tests including DPPH free radical scavenging activity, ferric reducing antioxidant power, and total antioxidant capacity.

MATERIALS AND METHODS

Plants Material and Extracts Preparation

During March 2021, fresh leaves of *Artemisia herba-alba* were collected from the region of Hamem Elbibane (ElM'hir), Bordj Bou Arreridj in Algeria while *Olea europaea* L. Leaves were collected from the region of Tefreg (Djaafra), Bordj Bou Arreridj in Algeria.

Leaves were cleaned, dried in shade, powdered, and then stored in an airtight container. About 10g of each plant powder was extracted with 150 mL of different solvents (70% acetone, 70% ethanol and 70% methanol) using a mechanical shaker for 48H in the dark at room temperature. The extracts were thereafter filtered. The filtrates were evaporated in a vacuum under 70°C on a rotary evaporator.

The yields of crude extract of *Artemisia herba-alba* were 24.8, 23.6 and 21% for methanolic, ethanolic and acetonic extract, respectively. In contrast, for *Olea europaea* L., the highest yield was detected in ethanolic extract (37.65%), followed by acetone extract (36.98%), then methanolic extract (35%).

Physicochemical Analysis of Plants Powder

Determination of Humidity

The moisture of the powder was determined by the oven drying method according to AOAC (2000) [8]. A quantity of 2g of leaf powder was placed in a previously tared porcelain crucible. The crucibles and their contents were then placed in an oven (Mettler type-ONE 7, Schutzart DIN EN 60529-IP 20. Germany), at

103±2°C for 24 hours. After cooling in a desiccator containing a desiccant (silica gel), the crucibles were weighed. The humidity percentage was calculated by the following formula:

$$H\% = (m - m' / m) \times 100.$$

m: mass of the sample before drying

m': mass of the sample after drying = mass (crucible + sample) - mass of the empty crucible

Determination of Ash Content

The ash content was determined according to AOAC (2000) [8]. After dry mineralization, in a porcelain crucible, previously tared, a quantity of 2g of each plant powder was introduced into a muffle furnace of the mettler type at a temperature of 800°C for 10 hours, until white ash was obtained (all the organic matter burns and only the inorganic part of the sample is recovered). It was left to cool in a desiccator, and then the weights of the crucibles were noted. The ash content was calculated according to the following formula:

$$AC (\%) = [(M - M') / E] \times 100\%.$$

AC: Ash Content.

M: final mass (crucible + total ash).

M': mass of the empty crucible.

E: test sample of the material.

Determination of lipid content

The fat content of the plants powder was determined by soxhlet extraction according to BIPEA (1976) [9]. A quantity of 10g of powder from each plant was placed in an extraction cartridge (Wattman cartridge), which was then inserted into the extraction bulb. A pre-weighed round-bottomed flask (W1) was filled with 2/3 petroleum ether. The sample was continuously extracted with boiling petroleum ether (P.E. 35°C) which gradually dissolved the fat. The solvent containing the fat returned to the flask by successive spills caused by a siphon effect in the lateral elbow. Since only the solvent can evaporate again, the fat accumulated in the flask until the extraction was complete. Once the extraction was complete, the ether was evaporated, on a rotary evaporator, and the flask containing the fat was weighed to obtain the weight (W2). The fat content was calculated according to the following formula:

$$FC (\%) = [(W2 - W1) \times 100] / SW.$$

Whereas,

FC: Fat Content.

W2: ball weight + mass of fat (g) dry.

W1: weight of empty balloon (g).

SW: sample weight (g).

Phytochemical Analysis

Determination of Total Phenolic Content

The Folin-Ciocalteu colorimetric method was used for the estimation of total polyphenol contents [10]. Aliquots (200 μ L) of appropriately diluted extracts were incubated for 2 min with Folin-Ciocalteu reagent, and then 75 mg/mL of sodium carbonate solution were added. After 2 h of incubation at 25°C, the absorbance at 765 nm was determined. The results were expressed as microgram of gallic acid equivalent (GAE) per milligram of dry weight (dw).

Determination of Total Flavonoid Content

Total flavonoid content was estimated according to the method described by Jain et al. (2011) [11]. The extract (1 mL) was incubated with 1 mL aluminium chloride. The absorbance at 430 nm was determined after 1 h of incubation at 25°C. The results were expressed as milligram of quercetin equivalent (QE) per gram of dry weight dw.

Determination of Total Pro-Anthocyanidins Content

Total pro-anthocyanidins content of each extract was determined according to a previously reported method with slight modifications [12]. A volume of 0.5 ml of each extract (1 mg/ml) was added to 3 ml vanillin solution (4%), previously prepared in methanol, and 1.5 mL of hydrochloric acid. The mixture was vortexed and incubated for 15 min at room temperature. Absorbance at 500 nm was recorded. The results were expressed as milligram of catechin equivalent (CE) per gram of dry weight (dw).

Determination of Carotenoids Content

Carotenoids were extracted according to Sass-Kiss et al. (2005) [13]. A quantity of 50 mg of the powder from each plant was added to 10 mL of n-hexane:acetone:ethanol (2:1:1). After stirring for 15 min, the mixture was centrifuged at 4500 rpm for 15 min. The upper layer was pipetted and the procedure was repeated. Combined extracts were diluted to 50 mL volume with hexane. The compounds were detected at 450nm. The results were expressed as microgram of β -carotene equivalent (β CE) per gram of dry weight (dw).

Determination of Chlorophyll Pigments Content

The determination of β -carotene, lycopene and chlorophyll contents of the various extracts was carried out by spectrophotometry according to the protocol described by Nagata and Yamashita, (1992) [14]. Briefly, 100 mg of each extract were added to 10 mL of an acetone-hexane mixture (4: 6 V/V). The whole mixture was vortexed and then filtered through Whatman No 4 paper. Optical density of the filtrates was recorded at 663 nm, 645 nm, 505 nm and 453 nm by the spectrophotometer at the same time.

The pigment content was calculated according to the equations indicated below and the results were expressed as milligram of chlorophyll, carotenoids or β -carotene per gram of dry weight.

- Chlorophyll a (mg/100 ml) = $0.999 \times A_{663} - 0.0989 \times A_{645}$
- Chlorophyll b (mg/100 ml) = $-0.328 \times A_{663} + 1.77 \times A_{645}$
- Lycopene (mg/100 ml) = $-0.0458 \times A_{663} + 0.204 \times A_{645} - 0.372 \times A_{505} + 0.0806 \times A_{453}$
- β -carotene (mg/100 ml) = $0.216 \times A_{663} - 1.22 \times A_{645} - 0.304 \times A_{505} + 0.452 \times A_{453}$

Antioxidant Activity

DPPH Radical Scavenging Activity

The DPPH test was carried out as described by Burits and Bucar, (2000) [15]. A volume of 50 μ L of various dilutions of the extracts was mixed with 5 mL of a 0.004% methanol solution of DPPH. After an incubation period of 30 min, the absorbance of the samples was read at 517 nm. Butylhydroxytoluene (BHT), quercetin and ascorbic acid were used as positive controls.

Ferric Reducing Antioxidant Power (FRAP)

FRAP analysis was evaluated according to the protocol of Oyaizu (1986) [16]. Extracts (200 μ L) were mixed with phosphate buffer (500 μ L, 2.0 M, pH 6.6) and potassium ferricyanide (2.5 mL, 1.0%), and the mixture was incubated at 50°C for 20min. A portion (2.5mL) of trichloroacetic acid (10%) was added to the mixture, which was then centrifuged at 650 x g for 10min. The upper layer of the solution (500 μ L) was mixed with distilled water (2.5mL) and ferric chloride (0.5 mL, 0.1%), and the absorbance was measured at 700 nm.

Increased absorbance of the reaction mixture indicated increased reducing power.

Total Antioxidant Capacity

The total antioxidant capacity of the extracts was evaluated by the phosphomolybdenum test according to the procedure described by Prieto et al. (1999) [17]. An aliquot of 0.1 mL of sample solution was combined with 1 mL of reagent solution (0.6 M sulfuric acid, 28 mM sodium phosphate and 4 mM ammonium). The mixtures were incubated for 90 min at 95°C. After cooling at room temperature, the absorbance was measured at 695 nm. The results were expressed as microgram of ascorbic acid equivalent (AAE) per milligram of dry weight (dw).

Statistical Analysis

All variables were verified for homoscedasticity using Levene's test and normality using a one-sample Kolmogorov-Smirnov test. To improve the normality of the data, non-normally distributed variables were \log_{10} transformed. The one-way ANOVA was used to evaluate the effect of extract type on the individual means of total phenolic, total flavonoid, and total proanthocyanidin contents. It was also used to compare the results of DPPH radical scavenging activity, ferric reducing antioxidant power, and total antioxidant capacity between the three types of extracts. Results are expressed as mean \pm SE. All tests are two tailed, and results were considered significant at $p < 0.05$. Statistical analyses were performed using SPSS Version 19.

RESULTS

Physicochemical Analysis

The results of the physicochemical analysis of the studied plants leaves powder are shown in Table 1. The moisture, ash, and fat levels obtained for *Artemisia herba-alba* leaf powder are 7.86%, 5.99%, 4.94%, respectively. Whereas, leaf powder of *Olea Europaea* L. showed 8.23% moisture, 3.5% ash content and 2.81% lipid content.

Table 1: physicochemical parameters of plants powder

	Humidity	Ash Content	Lipid Content
<i>Artemisia Herba-alba</i>	7.86%	5.99%	4.94%
<i>Olea Europaea</i> L.	8.23%	3.5 %	2.81%

Phytochemical Analysis

Total Phenolic Content

Acetonic extract of *Artemisia herba-alba* showed the highest concentration of polyphenols (178.14 ± 0.92 $\mu\text{gGAE/mg dw}$), followed by methanolic extract (96.77 ± 0.81 $\mu\text{g GAE/mg dw}$) and ethanolic extract (96.75 ± 0.19 $\mu\text{g GAE/mg dw}$). In the case of *Olea europaea* L., the highest level of polyphenols was detected in methanolic extract (150.20 ± 0.72 $\mu\text{g GAE/mg dw}$) followed by acetonic extract (139.36 ± 0.75 $\mu\text{g GAE/mg dw}$) and ethanolic extract (129.09 ± 1.75 $\mu\text{gGAE/mg dw}$) (Fig.1).

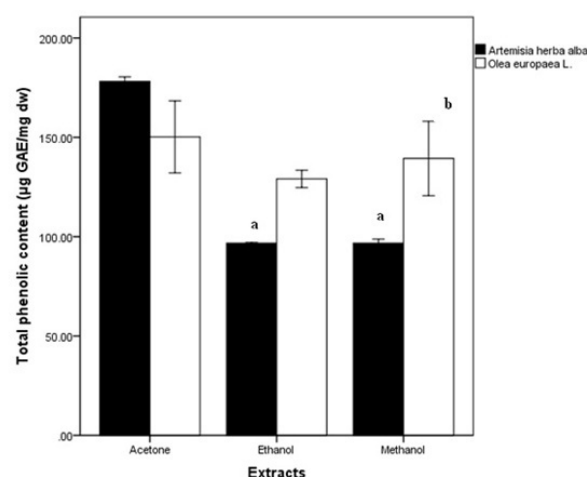


Figure 1: Total phenolic content of plants.

^a $p < 0.001$ versus acetonic extract of *Artemisia herba-alba*.

^b $p < 0.01$ versus ethanolic extract of *Olea europaea* L.

Total flavonoid

According to the data in the Fig. 2, the total flavonoid content varies depending on the solvent utilized. Acetonic extract of *Artemisia herba-alba* presented the highest concentration of flavonoid with an average value of 30.97 ± 0.14 mg QE/g dw, followed by methanolic extract with an average concentration of 17.83 ± 0.11 mg QE/g dw, then ethanolic extract with an average rate of 17.75 ± 0.13 mg QE/g dw. Methanolic extract of *Olea europaea* L. had the highest concentration of flavonoid with an average value of 16.74 ± 0.64 mg QE/g dw, followed by acetonic extract (12.36 ± 0.39 mg QE/g dw), and ethanolic extract (10.21 ± 0.80 mg QE/g dw).

Total Pro-Anthocyanidins

The analysis of condensed tannins showed that the high concentration for *Artemisia herba-alba* was obtained with acetonic extract (37.21 ± 1.83 mg CE/g dw), followed by ethanolic extract (22.77 ± 0.78 mg CE/g dw), and methanolic

extract (19.07 ± 0.78 mg CE/g dw). As for *Olea europaea* L., the highest concentration was recorded with the acetonic extract (27.03 ± 0.33 mg CE/g dw), followed by methanolic extract (6.94 ± 0.22 mg CE/g dw), and ethanolic extract (6.34 ± 0.69 mg CE/g dw) (Fig.3).

Carotenoids

Artemisia herba-alba presented the highest carotenoid concentration (34.2 ± 0.125 μ g/g dw)

in comparison with *Olea europaea* L. (11.12 ± 0.17 μ g/g dw).

Chlorophyll Pigments

As shown in Table 2, *Artemisia herba-alba* extracts displayed high concentrations of chlorophyll a and chlorophyll b, and only trace amounts of other pigments. Whereas, *Olea europaea* L., extracts exhibited a low quantity of fat-soluble pigments.

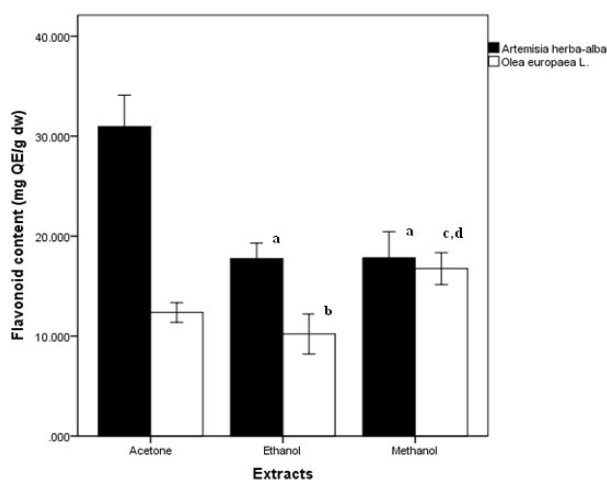


Figure 2: Total flavonoid content of plants

^ap < 0.001 versus acetone extract of *Artemisia herba-alba*.

^bp < 0.05 versus acetonic extract of *Olea europaea* L.

^cp < 0.001 versus acetonic extract of *Olea europaea* L.

^dp < 0.001 versus ethanolic extract of *Olea europaea* L.

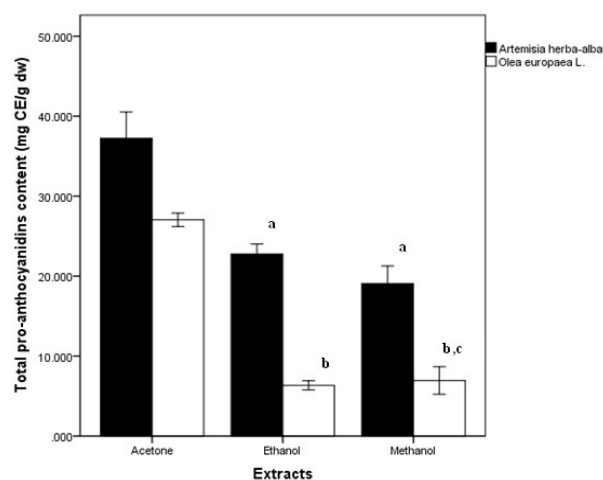


Figure 3: Total pro-anthocyanidin content of plants

^ap < 0.01 versus acetonic extract of *Artemisia herba-alba*.

^bp < 0.001 versus acetonic extract of *Olea europaea* L.

^cp < 0.05 versus ethanolic extract of *Olea europaea* L.

Table 2: Liposoluble pigment content (mg/g dw) of the various extracts of *Artemisia herba-alba* and *Olea europaea* L.

	<i>Artemisia herba-alba</i>			<i>Olea europaea</i> L.		
Solvent	acetone	ethanol	methanol	acetone	ethanol	methanol
chlorophyll a (mg/g dw)	134	113 ^{a,b}	84.53 ^a	49.32	7.55 ^{c,d}	18.7 ^c
chlorophyll b (mg/g dw)	224	47.66 ^{a,b}	97.13 ^a	14.82	8.74 ^{c,d}	7.55 ^c
Lycopene(mg/g dw)	23.5	13.7 ^{a,b}	01.06 ^a	/	/	/
β- carotene (mg/g dw)	26.23	16.06 ^{a,b}	06.53 ^a	13.45	/	/

^ap<0.001 versus Acetonic extract of *Artemisia herba alba*. ^bp<0.001 versus methanolic extract of *Artemisia herba alba*.

^cp<0.001 versus Acetonic extract of *Olea europaea*L. ^dp<0.001 versus methanolic extract of *Olea europaea*L.

Antioxidant Activity

DPPH Radical Scavenging Activity

Due to its solubility and efficiency, DPPH radical test is one of the most extensively utilized techniques for rapid and direct assessment of antioxidant activity [18].

The results revealed that acetonic extract of *Artemisia herba-alba* exhibited the greatest antiradical activity with an IC₅₀ of 0.611 mg/mL, followed by methanolic extract and ethanolic extract with an IC₅₀ of 0.912 mg/mL and 0.956

mg/mL, respectively. For *Olea europaea* L. methanolic extract was the most active, with an IC₅₀ of 0.56 mg/mL followed by ethanolic extract with a value of 0.60 mg/mL, then acetonic extract with a value of 0.61 mg/mL (Fig.4).

Ferric Reducing Antioxidant Power (FRAP)

The ability of a substance to transfer an electron or liberate a hydrogen atom is known as reducing power [19]. The findings from this study showed that the investigated extracts have demonstrated proven reducing power. The

reducing power of *Artemisia herba-alba* extracts is categorized as follows: acetonetic extract ($EC_{50} = 5.03$ mg/mL) > methanolic extract ($EC_{50} = 5.20$ mg/mL) > ethanolic extract ($EC_{50} = 5.35$ mg/mL). The reducing power of *Olea europaea* L. is classified as follows: methanolic extract ($EC_{50} = 0.83$ mg/mL) > acetonetic extract ($EC_{50} = 1.47$ mg/mL) > ethanolic extract ($EC_{50} = 2.16$ mg/mL) (Fig.5).

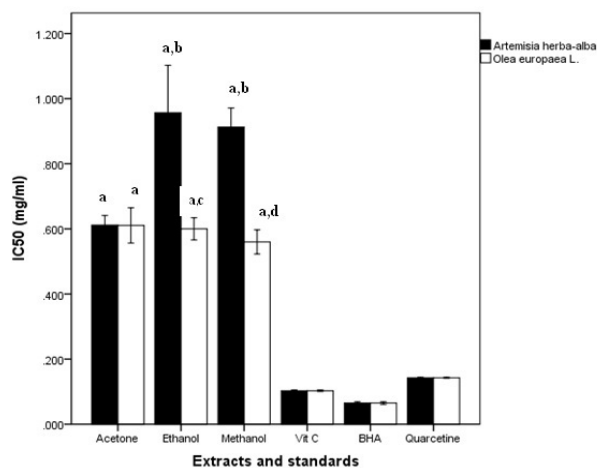


Figure 4: DPPH scavenging activity of plants and standards

^a $p < 0.001$ versus vit C, BHA, Quercetin.

^b $p < 0.001$ versus acetonetic extract of *Artemisia herba-alba*.

^c $p < 0.05$ versus acetonetic extract of *Olea europaea* L.

^d $p < 0.01$ versus acetonetic extract of *Olea europaea* L.

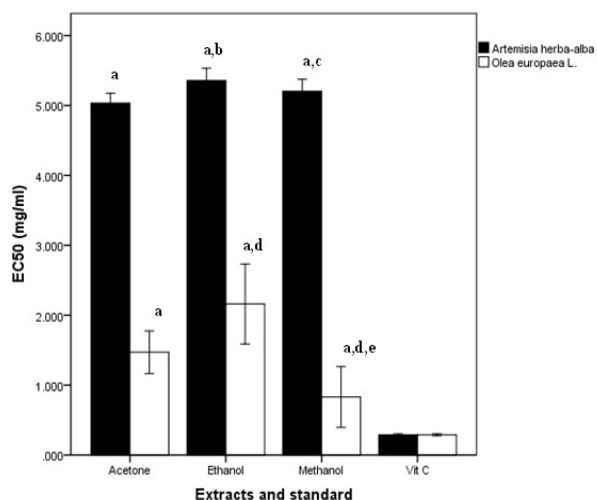


Figure 5: Ferric reducing antioxidant power of plants and standard

^a $p < 0.001$ versus Vit C.

^b $p < 0.01$ versus acetonetic extract of *Artemisia herba-alba*.

^c $p < 0.05$ versus ethanolic extract of *Artemisia herba-alba*.

^d $p < 0.001$ versus acetonetic extract of *Olea europaea* L.

^e $p < 0.001$ versus ethanolic extract of *Olea europaea* L.

Total Antioxidant Capacity

The TAC of *Olea europaea* L. extracts is classified in the following order: ethanolic extract (131.03 μ g AAE/mgdw), acetonetic extract (141.03 μ g AAE/mg dw) and methnolic extract (150.49 μ g AAE/mg dw). While for *Artemisia herba-alba* extracts, the TAC is classified as follows: methanolic extract (76.05 μ g AAE/mg dw), ethanolic extract (84.18 μ g AAE/mg dw) then acetonetic extract (97.91 μ g AAE/mg dw) (Fig.6).

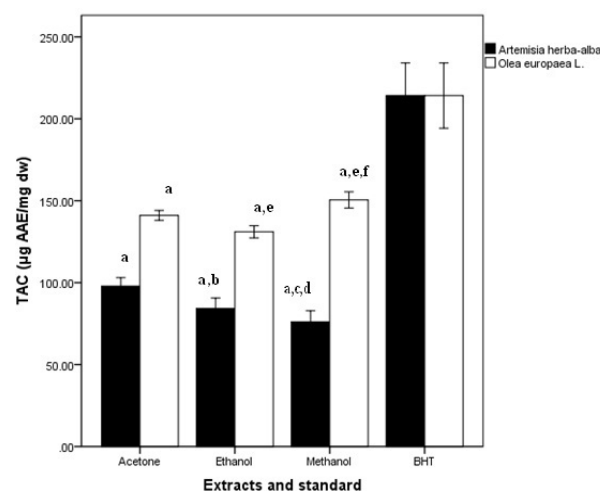


Figure 6: Total antioxidant capacity of plants and standard

^a $p < 0.001$ versus BHT.

^b $p < 0.01$ versus acetonetic extract of *Artemisia herba-alba*.

^c $p < 0.001$ versus acetonetic extract of *Artemisia herba-alba*.

^d $p < 0.05$ versus ethanolic extract of *Artemisia herba-alba*.

^e $p < 0.01$ versus acetonetic extract of *Olea europaea* L.

^f $p < 0.001$ versus ethanolic extract of *Olea europaea* L.

DISCUSSION

In this study, three solvents were used for the extraction of bioactive compounds from the leaves of *Artemisia herba-alba* and *Olea europaea* L., namely 70% acetone, 70% ethanol and 70% methanol. The results showed that the three solvents used gave better extraction yields. We used organic solvents at 70% to create a moderately polar medium that allows the best extraction of bioactive compounds [20]. Furthermore, the type and polarity of the solvent can affect the transfer of the hydrogen atom [21]. The extraction was performed at room temperature and with stirring to allow the obtaining of a maximum of bioactive compounds and prevent their degradation [22].

In the present study, the results showed that the methanolic extract of *Artemisia herba-alba* exhibits the highest yield. This is in agreement with the findings of De Abreu and Mazzafera (2005) and Falleh et al. (2008) [23, 24], who recommended methanol for the extraction of phenolics. While for *Olea europaea* L., ethanol was found to extract the highest level of bioactive compounds. Accordingly, Blasi et al. (2016) reported ethanol was the optimum extraction solvent of phenolics from *Olea europaea* leaves [25].

Artemisia herba-alba and *Olea europaea* L. were characterized in terms of total phenolics, flavonoids, condensed tannins and chlorophyll pigments. The total phenolic, flavonoid and condensed tannin contents of *Artemisia herba-alba* varied between 96-178 µg GAE/mg dw, 17-30 mg QE/g dw, and 19-37 mg CE/g dw, respectively. Our results are in quite agreement to that reported by Bourgou et al. (2016), who worked on the *Artemisia herba-alba* aerial parts using methanol as extraction solvent, and who recovered a total flavonoid amount of 16-43 mg CE/g dw [3]. However, our results are significantly higher in terms of total phenol content (8-17 mg GAE/g dw) and condensed tannin content (0.18-0.87 µg/g dw).

Total phenolic and flavonoid levels in *A. herba-alba* extract were comparable to those reported elsewhere [26], and higher than those identified by Djeridane et al. (2006) and Al Mustafa and Al Thunibat (2008) [27, 28].

Furthermore, Akroutet al. (2011) indicated that Asteraceae family, especially the *Artemisia* genus, is a valuable source of phenolic compounds [29]. This abundance could be linked to the hard climate conditions found in the Asteraceae's natural habitat (hot temperatures, intense sun exposure, dryness and salt), which stimulate the biosynthesis of secondary metabolites like polyphenols. Indeed, plant phenolic content is influenced by both intrinsic (genetic) and extrinsic (environmental, handling and storage) factors [30].

Total phenolic, flavonoid, and condensed tannin concentrations in *Olea europaea* L. ranged from 129 to 150 µg GAE/mg dw, 10 to 16 mg QE/g dw, and 6 to 27 mg CE/g dw, respectively. The total phenolic content was higher than that reported by Goldschmidt Linset al. (2018) (131 mg GAE/g dw) and Fernández-Poyatoset al. (2019) (31.52

mg GAE/g dw) [31, 32]. Flavonoid content was slightly lower than that found by Goldschmidt Linset al. (2018) (19.4 mg QE/g dw) [31].

The amount of condensed tannins obtained in this study is significantly higher when compared to that obtained by Ben Mansour-Gueddeset al. (2020) who used an aqueous extract of *Olea europaea* L. (0.24- 0.84 mg CE/g dw) [33]. The level of proanthocyanidins found in a plant depends on two main factors: the stage of vegetative development and the environmental conditions. Their concentration varies greatly between plant species and even within the same species, depending on maturity, age of the leaves, age of the flowers and season [34]. To protect themselves against environmental stresses like high temperatures and UV radiation, olive trees develop a variety of secondary metabolites [35]. Interestingly, Vita et al. (2018) also pointed out that the qualitative and quantitative biocompound profiles of olive trees differ based on cultivar, phenological stage, leaf maturation degree, phytosanitary state, climate, and cultivation area [36].

The amount of chlorophyll pigment in the leaves of the plants analyzed exhibited considerable variability. Therefore, climate circumstances influenced chlorophyll a, chlorophyll b, Lycopene, and β-carotene concentrations. The pigment levels found in olive leaves in this study were much lower than those found by Ben Mansour-Gueddes et al. (2020), who proved that the amount of chlorophyll a and b in olive cultivars fell dramatically under drought stress [33]. Drought is, in fact, one of the elements that influence photosynthesis and chlorophyll content.

The antioxidant activity of various extracts was measured using three different procedures: DPPH radical scavenging activity, ferric reducing antioxidant power and total antioxidant capacity. The results of the antiradical activity of *A. herba-alba* showed that the acetone extract is the most active (IC₅₀ = 0.611 mg/mL) compared to the ethanol and methanol extracts. This activity is close to that of the standards including BHA, vitamin C and quercetin, which demonstrate IC₅₀ values of 0.065, 0.103 and 0.142 mg/mL, respectively. Indeed, the studied extract showed very effective antioxidant activity when compared to that of Tunisian *A. herba-alba* methanol/water extract which showed an IC₅₀ of 20.64 mg/L [26], or to that reported by

Bourgouet *al.* (2016)(3 mg/L) [3]. Furthermore, acetic extract exhibited high reducing power with an EC₅₀ of 5.03 mg/mL. This result is significantly higher to that recorded by Bourgouet *al.* (2016) (EC₅₀ varying from 100 to 360 µg/mL) [3]. The reducing power of an active compound can serve as a significant indicator of its potent antioxidant activity [37,38].

Methanolic extract of *Olea europaea* L. showed greatest antiradical activity with an IC₅₀ of 0.56 mg/ml. This result is lower than that reported by Goldschmidt Linset *al.* (2018) (IC₅₀ = 0.13 mg/mL) [31]. As for the reducing power, methanolic extract was also the most active with an EC₅₀ of 0.83 mg/mL which is lower than that of Lahmadiet *al.* (2019) (EC₅₀ = 0.68 mg/L) [39]. In comparison to the aqueous extract published by Rouibah et *al.* (2019) (173 µg AAE/mg dw), the three extracts prepared from the leaves of *Olea europaea* L. (acetone, ethanol and methanol) exhibited a strong antioxidant ability (141.03, 131.03, and 150.49 µg AAE/mg dw, respectively) [7].

The antioxidant activity of the extracts can be attributed to the presence of a large proportion of phenolic compounds. Bentahar et *al.* (2016) indicated that polyphenolic compounds improve the status of different oxidative stress biomarkers [37]. Their antioxidant powers have been connected to their ability to scavenge free radicals, break radical chain reactions, directly reduce peroxides, and improve antioxidative defense enzyme activities. Their structure, which includes numerous hydroxyl substituents, including a 3-OH group and a conjugated double bond system, is responsible for these actions. The stability of the aroxyl radical produced when the flavonoid donates a hydrogen atom to the radical is related to these characteristics [40,41].

CONCLUSION

This research work has revealed that Algerian *Artemisia herba-alba* and *Olea europaea* L. leaves extracts are rich in polyphenols, flavonoid, carotenoid, condensed tannin and chlorophyll pigments. In addition, a study of the antioxidant potential of the extracts using various complementary methods (scavenging of the free radical DPPH, the reducing power, and the total antioxidant capacity) revealed that the extracts exhibited substantial antioxidant activity.

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