

# Phenolic content, HPLC analysis and Antioxidant activity extract from *Tamarix Articulata*

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## ABSTRACT

Halophyte plants can easily adapt in saline habitats, salt marshes, mudflats, and inland deserts. This kind of plants can be utilized as new sources of natural antioxidants. The goal of the present investigation was to examine the antioxidant of methanolic extracts of the *Tamarix articulata* in southern Algeria (ElOued). Colorimetric methods were applied to assess the total phenolic and flavonoid contents, HPLC analysis was used to identify the individual polyphenols, and the amount was also determined, while scavenging assays of DPPH, FRAP, ABTS,  $\beta$ -carotène and total antioxidant capacity of methanol leaves extract from *Tamarix articulata* were used to measure antioxidant capacities. The resulted findings showed that total phenolic contents and flavonoid from the extract of this plant were found to be  $398.42 \pm 5.21$  mg GAE /g DW for phenolic content, and  $120.55 \pm 3.41$  mg CE/g DW. The HPLC analysis showed that there were at least 8 remarkable phenolic compounds of leaves extract, and the most important ones included vanillic acid, naringin, and caffeic acid. The seeds of this natural plant can be a promising source for new drugs because they contain antioxidant properties and phenolic compounds.

**Keywords:** *Tamarix articulata*, antioxidant activity, HPLC analysis.

## Introduction

Various saline habitats including coastal sand dunes, salt marshes and mudflats to inland deserts, salt flats and steppes can be suitable places for Halophytes to grow [1]. Because their salt tolerance limits and their original climate zone, these plants have been attributed a high physiological plasticity. Considering its various genera, based on the studies conducted by different researchers, *Tamarix* (Tamaricaceae) covers between 65 and 90 species which originally grow in Asia, Africa and Europe [2]. A lot of *Tamarix* species grow in salty areas such as deserts, sand dunes, salt marshes and ravines. Some species of *Tamarix* are grown as decorative plants in gardens or as wind breaks or shade trees [3-5]. *Tamarix articulata* belongs to this family

(Tamaricaceae) (Table 1), its generic name which is originated in Latin is believed to be *Tarfa green* in Algeria. *Tamarix River* in Spain is an annual plant, mostly seen in the Mediterranean region, Algeria, Tunisia, and Morocco. Its leaves have been used as traditional remedies for the treatment of various diseases such as antidiabetic agents [6], the stimuli of perspiration and diuretic [7], anthelmintic, antihemorrhoid hemostat and gingivitis. The plant has been used to cure dromedary galls and antimicrobial agents [8]. The galls of this plant absorb hydrolyzable tannins up to 48% of their total weight [9, 10]. In Algeria and surrounding regions, the plant has been medicinally applied to cure rheumatism, diarrhea, and other maladies [11]. There exist natural antioxidants in all parts of the plant, and phenolics, carotenoids and vitamins are the typical compounds representing antioxidant activities [12]. Considering all types of natural antioxidants, polyphenols can be regarded as the major potent compound, because of being abundantly applied in the food industry, cosmetic, pharmaceutical and medicinal materials [13]. Structurally, phenolics form an aromatic ring, including one or more hydroxyl substituent, and range from simple phenolic molecules to highly polymerized compounds [14]. The phytochemical screening showed a conspicuous absence of alkaloids in methanolic extract, then a remarkable presence

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of tannins in particular hydrolysable tannins. Other metabolites and bioactive compounds have been determined like flavonoids, cardiac glycosides, steroids and - terpenoids (Table 2) [15]. Considering all of these characteristics, the halophyte *Tamarix articulata* was selected to be examined in this study. In the current study, total phenolic content, flavonoid, antioxidant activities and HPLC profile of methanolic extract from *Tamarix articulata* which grows in southeast part of Algeria (ElOued) were investigated (Figure 01). Hence, the scavenging activities towards DPPH, ABTS, FRAP,  $\beta$ -carotène and total antioxidant activity were assessed for methanolic extract of *Tamarix articulata*.

**Table 1. Classification of *Tamarix articulata*.**

Division:	Magnoliophyta
Class:	Dicotyledons
Subphylum:	angiosperms
Branching:	spermaphytes
Genre:	Tamarix
Subclass:	Archichlamydes
Order:	Parlétales
Species:	Tamarix aphylla L. karst.

**Table 2: Phytochemical compounds identified of methanolic extract.**

Metabolite	Methanolic extract
Steroids	+
Terpenoids	+
Alkaloids	-
Flavonoids	+
Tannins	+
Cardiac glycosides	+
Cyanogens glycosides	-
Anthraquinones	-
Saponins	-
Reducing sugar	-

## Materials and Methods

### Chemical

Methanol (99%) and Folin Ciocalteu reagent were obtained from biochem chemopharma Co (Canada). 1,1-Diphenyl-2-picryl hydrazyl (DPPH) (99%), ascorbic acid (99.7%), gallic acid (99%), sodium carbonate (99%), AlCl<sub>3</sub> (99%), rutin (99%) were all bought from Merck Co, all other reagents used in the study were of analytical grade.

### Plant material

All the samples were collected from mature, field-grown plants in the region of ElOued, Algeria. Green leaves were obtained during the growing season (Sep 2016). For each species of the plant, the samples were collected, placed in plastic bags, taken to the laboratory, and left to air dry. The species of the plant was determined by Dr. Youcef HALIS, Associate Professor,

Scientific and Technical Research Centre for Arid Areas (CRSTRA), Biophysical Station, Touggourt, Algeria.

### Preparation of extract

Distilled water was used to wash the leaves of *T. articulata*, then, they were left at room temperature for 5 days in the dark, and dried in the oven for 1 h at 60 °C. The dried leaves of the plant were crushed into small parts, and then were finely powdered. The extract was obtained by magnetic stirring of 10 g dry powder in 100 ml pure methanol. Furthermore, the extract was maintained for 24 h at room temperature, then filtered through a Whatman n° 4 filter paper. Finally, the filtrate was concentrated under vacuum. Then, it was kept at 4 °C [16].

### Determination of total phenolic content

Singleton and Rossi (1965) used the Folin- Ciocalteu method with some modifications to determine the total phenolic content of the plant [17]. It was determined by the ultraviolet and visible spectrometer at 760 nm. For this study, different concentrations (0.3-0.03 mg/ ml) of samples were added to 0.5 mL Folin- Ciocalteu phenol reagent (diluted 10 times with distilled water). After 5 min, the mixture was mixed with 2 ml of saturated sodium carbonate solution (20%). The mixture was stirred after staying 30 min in the dark; the absorbance was read at 760 nm. A standard curve of Gallic acid was used. The analysis was conducted in triplicate, and the total phenolic content of the organs was represented as mg Gallic acid equivalents per gram of dry weight (mg GAE/g DW).

### Estimation of total flavonoid content

A colorimetric assay was used to identify the total flavonoids contents [18]. 1 ml of AlCl<sub>3</sub> (2 %) solution was added to 1 ml of methanolic extract and mixed well during 30 min to give pink color solution. At 430 nm, the absorbance of the mixture was identified. For calculating the standard curve, Rutin was utilized (0.1 and 0.02 g/L). The total flavonoid content was shown as mg of rutin equivalents (REs) per g of extract. The analysis of all the samples and the standards were done in triplicate.

### HPLC analysis

The high-performance liquid chromatography (HPLC) was used to determine polyphenols of the extract [19]. The Shimadzu (LC 20, Japan), the system comprised of an LC-10AD pump, a CTO-10A column oven, an SPD-10A UV-DAD detector, a CBM-10A interface and an LC-10 Workstation was applied. LC-18 column with properties of (250 mm x 4 mm i.d. x 5 mm) was used. 20  $\mu$ l of *Tamarix articulata* extract was added and the constituting elements of the 4 mixtures were divided by the gradient elution at 30 °C. The mobile phases were: A, 98:2 (v/v) acetic acid and B acetonitrile. Acetic acid and the elution gradient was: 0–5 min, 95% B; 10 min, 90% A; 11 min, 80% A; 30 min 60% A; 40 min 50% A; 50 min 20% A. The stream was 0.8 ml/min, and the absorbance of detection was 268 nm. Phenolic compound standards included: chlorogenic acid, rutin, gallic acid, caffeic acid, vanillic acid, vanillin, p-coumaric acid,

and naringin were dissolved in methanol used for the determination of polyphenols existing in the extract of *Tamarix articulata*. By comparing the retention time of the reference standards, the peak in HPLC was determined. Considering the peak area of the reference compounds, the concentration of each phenolic compounds in the extracts was identified and it was reported as mg/g of extract.

## Antioxidant assays

### Evaluation of total antioxidant capacity

The phosphomolybdenum method was based on the reduction of Mo (VI), to Mo (V) by the extract plant that produces the formation of a green phosphate Mo (V) complex at acid Ph<sup>201</sup>. 1 ml of reagent solution (0.6 M sulfuric acid, 0.28 M sodium phosphate and 0.04 mM ammonium molybdate) was added to 0.1 ml of methanolic extract.

The samples were put in a thermal block at 95 °C for 90 min. After that, the absorbance of each solution was assessed at 695 nm after the cooling of the mixture at room temperature. The analysis of all the samples were done in triplicate. The antioxidant capacity was represented as mg Gallic acid equivalent per gram dry weight (mg GAE/g DW).

### DPPH assay

DPPH quenching ability of methanolic extract was calculated in order to scavenge the 2, 2-diphenyl-1-picrylhydrazil free radical<sup>21</sup>. 1ml of the extract at known concentrations was added to 0.25 ml of methanolic solution DPPH. The mixture was placed at room temperature for 30 min in the dark, the ability of the extracts to decrease the stable radical DPPH to the yellow-colored was assessed before evaluating the absorbance at 517 nm, and the ability of extracts to decrease the stable radical DPPH to the yellow-colored DPPH was also measured. The ability to scavenge the DPPH radical was calculated using the following equation:

$$\text{DPPH\_scavenging effect \%} = [(A_0 - A_1) / A_0] \times 100$$

Where:

$A_0$  is the absorbance of the control at 30 min.

$A_1$  is the absorbance of the sample at 30 min.

The antioxidant capacity of the extract was represented as IC50. The IC50 was measured from the antioxidant activities (%) of the pure compound used at various concentrations. The analysis of all the samples were done in triplicate. A lower IC50 value showed the higher antioxidant activity of the plant extract.

### FRAP assay

The ability of the extracts to reduce Fe<sup>3+</sup> was determined by the method described by Benzie and Strain with some modifications<sup>22</sup>. FRAP reagent contained: 2.5 ml of TPTZ (Tripyridyltriazine, 10 mM) mixed with 10 ml of HCl (40 mM) added to 2.5 ml of 20 mM FeCl<sub>3</sub>. After that, 25 ml of

0.3M acetate buffer (PH 3.6) was mixed with the above reaction. 1.8 ml of the freshly prepared was added to the volume of 0.2 ml of methanol extract (concentration of extract 0.9 mg/ml) or standardized (Ascorbic acid) was added to 1.8 ml of freshly prepared FRAP reagent. The absorbance of each sample solution was subsequently measured at 595 nm using a spectrophotometer (Shimadzu UV-1800, Japan). The results were represented as mg AAE/g dry weight, using the equation obtained from the calibration curve:

$$Y = 1.9249 X + 0.0039.$$

All the determinations were performed in triplicate.

### ABTS assay

The antioxidant activity of methanolic extract of *Tamarix articulata* was evaluated by ABTS scavenging assay radical<sup>23</sup>. ABTS reacted with potassium per sulfate (K<sub>2</sub>S<sub>2</sub>O<sub>8</sub>) to form the cationic radical ABTS<sup>+</sup> · blue-green color. The antioxidant activity was determined by the discoloration of the solution, and was expressed by the percent inhibition (PI) of the absorbance at 734 nm and calculated using the following equation:

$$\text{ABTS radical scavenging activity} = [(\text{Abs}_{\text{control}} - \text{Abs}_{\text{sample}}) / \text{Abs}_{\text{control}}] \times 100$$

Where:

Abs control: Is the absorbance of ABTS radical + ethanol.

Abs sample: Is the absorbance of ABTS radical + ethanol extract or standard.

### β-Carotene linoleic acid bleaching assay

The free radicals' scavenging is calculated by measuring the inhibition of the production of volatile organic compounds and the formation of conjugated diene hydroperoxides arising from linoleic acid oxidation<sup>24</sup>. β-Carotene was prepared; 10 ml chloroform was added to 2 mg of this reagent, after that, 2 ml of β-Carotene solution was added. For removing the chloroform, the rotator evaporator was used at 40 °C.

The resulted reaction was added to 100 ml of aerated distilled water under vigorous shaking in the dark. 0.2 ml of the extract was added to 4.8 ml of the above mixture. A control sample was prepared using 0.2 ml methanol and 4.8 ml of β-carotene reaction. The mixture was incubated at 50 °C for 2 h. The capacity of extract to inhibit β-Carotene radical was measured at 470 nm. The antioxidant capacity was estimated by (IC<sub>50</sub> μg/ml) using the following equation:

$$\% \text{ antioxidant activity} = ((A_0 - A_t) / (A_0^\circ - A_t^\circ)) \times 100$$

Where:

$A_0$  and  $A_t$  are respectively the absorbance calculated at the beginning of the incubation for simple extracts and control.

$A_0^\circ$  and  $A_t^\circ$  are the observances unregistered at 2 h,

respectively for simple extract and control. All measurements were prepared in triplicate.

## Statistical Analysis

Experimental values are given as means  $\pm$  standard deviation (SD) of the three replicates for antioxidant activity. One way analysis of variance (ANOVA), correspondence analysis and determination of the Pearson correlation coefficient (q) were used during this work to evaluate and correlate results between them. The statistical calculations were carried out by Origin Pro Version 9.1 software (Origin Lab Corporation). The significance level was set at  $p < 0.05$ .

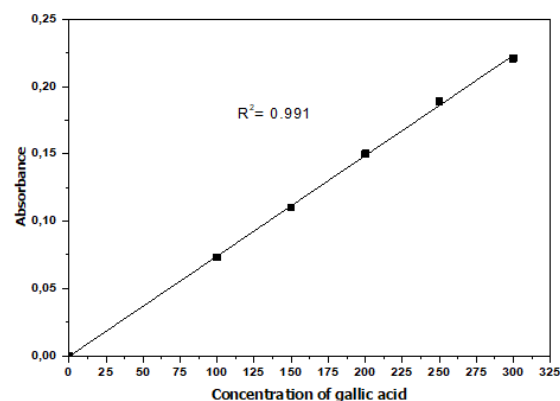
## Results and Discussion

### Total phenolic and flavonoids contents

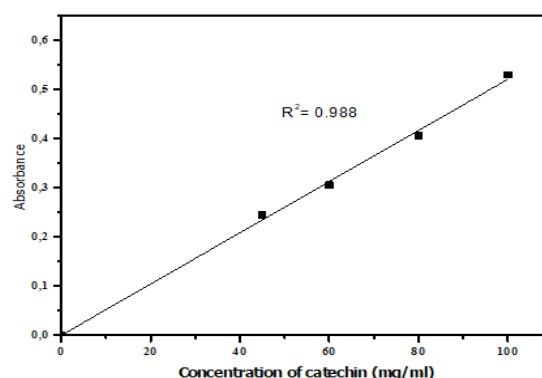
Preliminary evaluation of the phytochemical composition of methanolic extract of *Tamarix articulata* gave results represented in the Table 2. From this table, the methanolic extract of the plant studied were compared with the methanolic extract of *Tamarix articulata* growth in Adrar<sup>[15]</sup> containing polyphenols and flavonoids. The total polyphenol content of the methanolic leaves extract of *Tamarix articulata* obtained by maceration method was expressed as Gallic acid equivalent per gram of dry weight (Figure 1) using UV-Visible (Shimadzu UV-1800, Japan) and the equation of calibration curve:  $Y = 0.00778 x$ ,  $R^2 = 0.991$ ,  $x$  was the absorbance and  $Y$  was the gallic acid equivalent. All the findings were shown as means ( $\pm$ SEM), and the analysis of them were done in three replications. The edible leaves of the plant and *Tamarix articulata* growth in Adrar were found to be rich in polyphenols, a high concentration of phenolic content was found in *Tamarix articulata*  $398.42 \pm 5.21$  mg of GAE/g of dry weight and was found to be  $262.26 \pm 1.96$  mg of GAE/g of dry weight in *Tamarix articulata* extract growth in Adrar. The difference existing in the results indicated that the quality, nature of the soil and the region were responsible for the obtained results.

The presented results were in agreement with those published earlier indicating *Tamarix articulata* was an accessible source of natural phenolic compounds<sup>[25, 26]</sup>. Previous studies have reported the correlation between phenolic content with antioxidant activities, and some of the most important bioactive of phytochemical constituents such as flavonoids and tannins possess antioxidant<sup>[27]</sup>. The results showed that extracts from *Tamarix articulata* contained  $120.55 \pm 3.41$  mg of catechin/g of dry weight (Figure 2) using the equation obtained from the calibration curve:

$Y = 0.5617x$ ,  $R^2 = 0.985$  where  $x$  is the absorbance and  $Y$  is the catechin equivalent.



**Figure 1:** Standard curve for determination of phenolic content expressed in mg GAE/g of dry weight



**Figure 2:** Standard curve for determination of flavonoid content expressed in mg CE/g of dry weight.

### HPLC analysis:

The chromatographic profile of methanolic extract of *Tamarix articulata* has been represented in Figure 3. The results of the quantitative analysis of phenolic compounds showed that in methanolic extracts, there were the standard compounds of gallic acid, chlorogenic acid, vanillic acid, caffeic acid, vanillin, p-Coumaric acid, rutin, and naringin. Figure 4 showed the chromatograms of the standard markers' mixture. Peaks 1, 2, 3, 4, 5 and 6 were ascorbic acid, Gallic acid, chlorogenic acid, caffeic acid, vanillin and rutin; respectively.

The contents of these components in different extracts were determined according to the calibration curves of ascorbic acid  $y = 2260.81x$  ( $R^2 = 0.998$ ), Gallic acid ( $R^2 = 0.991$ ), chlorogenic acid  $y = 37492.06x$  ( $R^2 = 0.999$ ), caffeic acid  $y = 70429.77x$  ( $R^2 = 0.999$ ), vanillin  $y = 80555.42x$  ( $R^2 = 0.989$ ) and rutin  $y = 3118.94x$  ( $R^2 = 0.988$ ), where  $y$  was the peak area and  $x$  was the concentration of analytes (0–80  $\mu$ g/ml). The contents of these components in different extracts were determined according to the calibration curves<sup>[28]</sup>.

The quantitative results have been specified in Table 3. For *Tamarix articulata*, four major peaks were observed and eluted at 13.39, 16.27, 23.81 and 45.05 corresponding chlorogenic acids, caffeic acid, p-coumaric acid, and quercetin consequently. One compound with retention times of 28.37 was not identified. It was the first time to investigate the HPLC

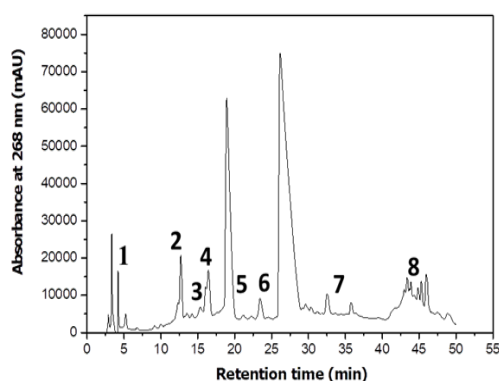
profile of *Tamarix articulata* leaves extract. However, because of its strong polarity, some compounds were not concentrated by methanolic extract [29]. *Tamarix articulata* was identified for the first time in this study. For a reliable characterization, the experimental data were compared with the literature [4, 30] and with reference standards commercially available. Most of the polyphenols identified in this study were determined as

compounds previously recognized in other species belonging to the genus *Tamarix*, which included gallic acid, ellagic acid, flavones and flavonols [31]. The data provided above demonstrated that the HPLC method developed in this study represented a reliable tool for the analysis of the major phenolic compounds in *T. articulata*.

**Table 3: Quantification of identified individual phenolic compounds of *Tamarix articulata* extract at 268 nm using HPLC system.**

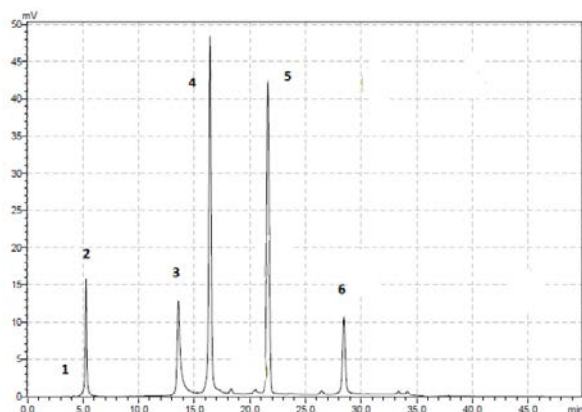
Identified compound	Retention Time (min)	Equation curve	Area	Area (%) in fraction	Height (uV)	Quantity (mg/g DW)
Gallic acid	5.29	y=54681x	136852	0.820	5449	2.574±0.01
Chlorogenic Acid	13.39	y=21665x	123706	0.741	3883	5.791±0.02
Vanillic Acid	15.53	y=65077x	266213	1.595	5439	4.090±0.02
Caffeic Acid	16.27	y=84066x	461760	2.767	15150	5.492±0.01
Vanillin	21.46	y=58930x	134417	0.805	2599	2.280±0.01
p-Coumaric Acid	23.81	y=49495x	282983	1.696	6903	5.717±0.02
Rutin	28.37	y=28144x	-	-	-	ND
Naringin	34.78	y=19377x	33723	0.202	1364	1.740±0.01
Quercetin	45.05	y=55378x	254411	1.525	10985	5.606±0.02

ND: Not detected



**Figure 3:** The HPLC chromatogram of leaves extract from *Tamarix articulata* at 268 nm.

- 1: Gallic acid; 2: Chlorogenic Acid; 3: Vanillic Acid; 4: Caffeic Acid; 5: Vanillin; 6: p-Coumaric Acid; 7: Naringin; 8: Quercetin.



**Figure 4.** Chromatogramme Profil of mélange étalons

- 1: ascorbic Acid; 2: Gallic acid; 3: Chlorogenic Acid; 4: Caffeic Acid; 5: vanilline; 6: Rutin

## Total antioxidant activity

Several tests and methods have been developed to evaluate the antioxidant capacity of plant extracts. Each assay represented the antioxidant activity of the test material from a different point of view. Applying a combination of two or more complementary test systems has been the most appropriate approach to identify the antioxidant activity of a plant extract [32]. The phosphomolybdenum method has been based on the decrease of Mo (VI) to Mo (V) by the antioxidant compounds, and the formation of green Mo (V) complexes with a maximal absorption at 695 nm. The total antioxidant activity of *Tamarix articulata* was shown to be  $345.92 \pm 8.45$  mg AGE/ g DW (Table 3). In comparison with the Tunisian *T. articulata*, the leaves' extract had interesting antioxidant capacity with the shown value of  $262.26 \pm 1.96$  mg AGE/ g DW. This strong result confirmed high antioxidant activity of the eaves' extract of the plant. This antiradical activity could be due to the phenolic compounds. In fact, it has been found that antioxidant molecules such as polyphenols, and flavonoids reduced the ability of free radicals [33]. The antiradical properties of polyphenols were mostly due to their redox capacity, which let them act as reducing agents, hydrogen donors, and singlet oxygen quenchers. Many synthetic drugs preserved against oxidative damage but they had negative side effects. Using natural antioxidants from food supplements like traditional medicines, can be considered as a possible solution to the problem. Phenolic compounds have been communicated to have very biological properties, such as antioxidant and anti-inflammatory. Recent evidence has suggested that diets rich in polyphenols play a leading role in oxidative stress-related disorders in the view of the fact of their antiradical capacity.



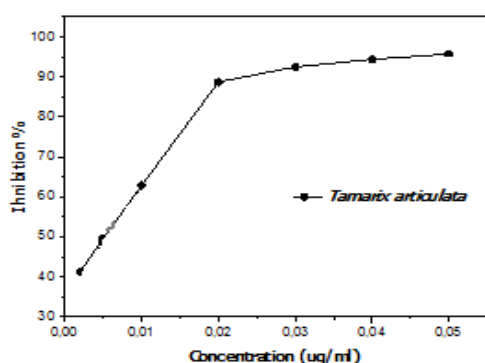
**Table 4: Phenolic content, flavonoids content, Scavenging activity of DPPH and AAT of Tamarix articulata leaves extracts. Antioxidant activity of DPPH expressed as % inhibition IC50 values (mg/ml).**

	<i>Tamarix articulata</i>
Phenolic content (mg GAE /g DW)	398.42 ± 5.21
Total flavonoids (mg CE/g DW)	120.55 ± 3.41
ATT (mg GAE/g DW)	345.92 ± 8.45
DPPH (IC50= µg/ml)	5.71± 0.22

### DPPH scavenging radical activity

The DPPH free radical can be considered a stable organic free radical, and has been generally accepted as a method for the evaluation of free radical scavenging potential of antioxidants [34] which has been a very simple and useful assay to detect the presence of antioxidant potential in the extracts [35].

The inhibitory concentrations of leaves' extract required 50% scavenging of the DPPH radical. The IC50 values of the antioxidant activity of the leaves' extract has been presented in Table 3 (Figure 5). It shows that methanolic leaves extract of *Tamarix articulata* exhibited the highest DPPH radicals with IC50= 5.71 ± 0.22 µg/ml. In comparison with the Algerian *T. articulata*, the leaves' extracts had interesting antioxidant activities against DPPH radicals shown by IC50 values of 9.1 µg/ml [36]. The results showed that high polyphenol contents in methanolic extract had the most potent radical scavenging capacity. Moreover, the concentrations of phenolic compounds were higher in the extract which could suggest that the high antioxidant capacity observed might be attributed mainly to the polyphenols. In a previous study, the high DPPH radical scavenging activity has been documented that could be due to the high content of phenolic compounds [37].



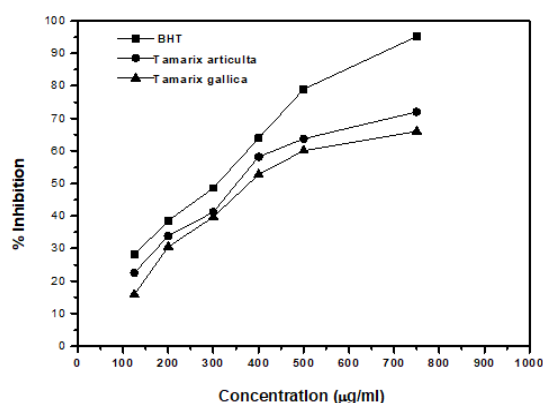
**Figure 5: DPPH scavenging activity of methanolic leaves' extract from *Tamarix articulata*.**

### ABTS assay

Figure 6 showed the dose-response curves of ABTS scavenging activities of methanolic extracts from *Tamarix articulata* and *Tamarix gallica* and reference antioxidant (BHT) on ABTS radicals. The ABTS radical scavenging activity of all extracts started with low values of 24.51 ± 0.66 % ; and 17.95 ± 0.52 % at a concentration of 125 µg/ml, respectively. After a rapid

growth, reached to stabilization of 75.81 ± 3.4 %, and 67.64 ± 3.55 at a concentration of 750 µg/mL, respectively. The abilities of scavenging ABTS radicals were in descending order: BHT > *Tamarix articulata* > *Tamarix gallica*.

Leaves' extract of *T. Gallica* showed significant antioxidant activities against ABTS+ cation radicals illustrated by IC50 values of 316.7 µg/ml [38]. The compounds with antioxidant activity were mainly phenolic acids, flavonoids and polyphenols [39]. Phytochemical studies have shown that *Tamarix articulata* extracts contain phenolic antioxidant compounds, such as flavonoids (luteolin, quercetin, apigenin, kaempferol, isorhamnetin, tamarixetin, quercetin dimethyl-ether, kaempferide and kaempferol-7,4'-dimethyl-ether) and phenolic acids (gallic acid, caffeic acid, p-coumaric acid and ellagic acid) [40].



**Figure 6: ABTS radical scavenging activity of *Tamarix articulata***

### FRAP assay

This technique uses the direct reduction of hexacyanoferrate(III) anion [Fe(CN)6] 3- into hexacyanoferrate(II) anion [Fe(CN)6] 4-. On a structural point of view, this observation has been in agreement with the lipophilic properties of phenols with a high number of hydrogen and carbon atoms [22]. In this work, FRAP antioxidant activity of methanolic extract from *Tamarix articulata* showed a wide range of the activity observed for *Tamarix articulata* (74.44 ± 1.05 mg AAE/g). The antioxidant scavenging activities of the extract for FRAP assay were shown in Table 5. It has been the first time to investigate the FRAP antioxidant activity of methanolic extract from *Tamarix articulata*.

**Table 5: The antioxidant scavenging activities of the two extracts for FRAP assay and ABTS assay**

	Antioxidant activity	
	ABTS 50% (µg/ml)	FRAP (mg/g)
<i>Tamarix articulata</i>	385.76 ± 2.64	479.23 ± 6.74

The values have been expressed as means ± SD of the triplicate measurements.

### β-Carotene

In β-Carotene model free radical scavenging, the oxidation of linoleic acid areas were attacked by the strongly unsaturated β-

Carotene radical, resulting in a diminution of absorbance at 470 nm. The existence of antioxidants agent could retard the extent of  $\beta$ -Carotene blanching by the neutralization of the linoleate free radicals formed in the system. The results of inhibition activity (50 %) of the extract has been shown in Table 6, the value obtained in *Tamarix articulata* was ( $IC_{50} = 43.52 \pm 2.43 \mu\text{g/ml}$ ). It has been the first time to investigate the  $\beta$ -Carotene model free radical scavenging of methanolic extract from *Tamarix articulata*. The interaction of a potent antioxidant with  $\beta$ -Carotene depended on the concentrations extracts. The results indicated and supported that the polyphenols can moderately prevent the degradation of  $\beta$ -Carotene caused by the radical reactions.

**Table 6:  $\beta$ -Carotene bleaching activities of *Tamarix articulata* leave extracts. Antioxidant activity was expressed as % inhibition  $IC_{50}$  values ( $\mu\text{g/ml}$ )**

Samples concentrations ( $\mu\text{g/ml}$ )	Reaction time (min)		Antioxidant activity $IC_{50} = (\mu\text{g/ml})$
	30	120	
<i>Tamarix articulata</i> Extract	20	35.37 $\pm$ 0.32	19.47 $\pm$ 0.59
	40	52.44 $\pm$ 1.31	41.08 $\pm$ 1.02
	60	83.42 $\pm$ 2.02	68.54 $\pm$ 1.85

Data are expressed as means  $\pm$  standard deviation of triplicate samples.

## Conclusion

In this study, phytochemical investigation, HPLC analysis, in vitro antioxidant activities of methanolic leaves' extract from *Tamarix articulata* acquired by the maceration technique have been examined. The richness of the extracts with the phenolic and flavonoid content was remarkable, and might be considered as a source of these compounds. Furthermore, in vitro assays showed high antioxidant activity, sufficient ability to inhibit the DPPH radical and phosphomolybdate. Furthermore, with a powerful analytical HPLC technique, the identification and quantification of 8 phenolic compounds were achieved for *Tamarix articulata* which can be used as antiradical drugs. In this context, the data presented in this work suggested that *Tamarix articulata* could be a new source of polyphenols, antioxidant activity, which can be a dare for new medicament.

## Conflicts of Interest

The authors declared no conflicts of interest.

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