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Antioxidant and Antibacterial Activities Valorisation of Two Extracts of *Pistacia atlantica* Desf

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Abstract **Original Research Article**

Medicinal plants are considered the essential raw material source for the discovery of new molecules necessary for the development of future drugs. The antioxidant activity of extracts of *Pistacia atlantica* Desf. The extracts were evaluated using several methods; the antioxidant activity was evaluated by DPPH and the reducing power essay. Moreover, the antimicrobial evaluation by Staphylococcus aureus ATCC25923 and Bacillus subtilis ATCC6633. This plant contains higher levels of total phenolic and a very important antioxidant activity.
Keywords: *Pistacia atlantica* Desf, Antioxidant, antibacterial.

INTRODUCTION

Pistacia atlantica Desf. Subsp *atlantica* is a tree from Ancardiaceae [1] .The genus Pistacia (family of Anacardiaceae) includes over 600 species. *P. vera, P. atlantica, P. terebinthus, P. khinjuk,* and *P. lentiscus* are the most famous species of Pistacia that are widely distributed in the Mediterranean and Middle Eastern area [2,3]. *Pistacia atlantica* Desf. is a deciduous tree which can reach up to 18 m in height. The big one's subjects can easily reach 1,000 years. The hard place produces resin putty that can be distilled but exudes naturally in hot weather. It's a resin putty, somehow a medieval ancestor of chewing gum, which local people were formerly some use and whose pharmacy was long served for the manufacture of ointments [4, 5, 6, 7].

The aerial parts and/or resin of plant has been also used in traditional medicine for the treatment of eczema, paralysis, diarrhea, throat infections, renal stones, jaundice, asthma, stomach-ache, and also as an astringent and a pectoral stimulant[8].

Secondary metabolites are the subject of much research, they have multiple interests, and they are put to use both in the food industry, cosmetics that pharmaceutical. They are widely used in therapy as vascular-protective, anti-inflammatory, enzyme inhibitors, antioxidants and, anti-free radicals [9].

Due to the economic importance, we study the antioxidant and antibacterial activities for use in the Algerian pharmacopoeia.

MATERIALS AND METHODS

Plant material

Pistacia atlantica. Desf Batt was harvested from the massif of Boutaleb (X1 E: 5° 30' 2.46" Y1 N: 35° 44' 41.74"). In May 2017.

Preparation of methanol extracts

The leaf of *Pistacia atlantica.* Desf. Was powdered and macerated in 80 % methanol for 24, 48 and 72 h, at the laboratory temperature (1:10 w/v, 10 g of dried herb). After maceration, the extracts were collected, filtered and evaporated to dryness under vacuum [10]. The dry extracts were stored at a temperature of -18 °C for later use.

Preparation of aqueous extracts

The method for preparing aqueous extracts from leaf of *Pistacia atlantica.* Desf thas been already described by Predrag et *al.* [11]. Briefly, dried plant material (10 g) was stirred in 100 ml of distilled water for 15 min at 90 ◦C followed by rapid filtration through four layers of gauze and then by a more delicate filtration through Whatman filter paper #1. The resulting filtrate evaporated to dryness under vacuum. The powder was stored at −10 ◦C until required

Determination of Total Phenolic Content

For total polyphenol determination, the Foline Ciocalteu method was used [12]. The samples (0.2 mL) were mixed with 1 mL of the Folin-Ciocalteu reagent

previously diluted with 10 mL of deionized water. The solutions were allowed to stand for 4 min at 25 °C before 0.2 mL of a saturated sodium carbonate solution (75 mg/mL) was added. The mixed solutions were allowed to stand for another 120 min before the absorbance were measured at 765 nm. Gallic acid was used as a standard for the calibration curve. The total phenolic compounds content was expressed as mg equivalent of Gallic acid per gram of extract (mg EAG/GE).

Determination of total flavonoids contents

The flavonoids content in our extracts were estimated by the Aluminium chloride solution according to the method described by Bahorun *et al*. [13]. Briefly, 1 mL of the methanol solution of the extracts was added to 1 mL of 2 % $AlCl_3$ in methanol. After 10 min, the absorbance was determined at 430 nm. Quercetin was used as a standard. Results were expressed as mg equivalent Quercetin per gram of extract (mg EQ/GE).

DPPH Assay

The donation capacity of extract was measured by bleaching of the purple-coloured solution of 1, 1-diphenyl-2-picrylhydrazyl radical (DPPH) according to the method of Hanato *et al*. [14]. One millilitre of the extracts at different concentrations was added to 0.5 mL of DPPH-methanol solution. The mixtures were shaken vigorously and left standing at the laboratory temperature for 30 min in the dark. The absorbance of the resulting solutions was measured at 517 nm. The antiradical activity was expressed as IC_{50} (micrograms per millilitre). The ability to scavenge the DPPH radical was calculated using the following equation:

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

Where:
A_0: the absorbance of the control at 30 min
A_1: is the absorbance of the sample at 30 min. Butylated hydroxytoluene (BHT) was used as standard.

Antimicrobial activity

Bacteria Strains were obtained from the American Type Culture Collection: bacteria (*Staphylococcus aureus* ATCC25923 and *Bacillus subtilis* ATCC6633). Muller Hinton agar was used for bacteria culture for yeast.

Anti-bacterial Activity

Agar disc diffusion method was employed for the determination of antibacterial activities of the extract [14, 15]. Briefly, a suspension of the tested microorganism (10^8 CFU / mL) was spread on the solid media plates. Filter paper discs (6 mm in diameter) were impregnated with 10 µL (100 mg/mL) of the extract and placed on the inoculated plates. These plates were incubated at 37 °C for 24 hours. Gentamicin (10 µg/disc) was used as a standard and dimethylsulfoxide DMSO as a control.

The antibacterial activity was determined by measuring of inhibition zone diameters (mm) and was evaluated according the parameters suggested by Alves *et al*. [16]:

- <9 mm, inactive ;
- 9–12 mm, less active ;
- 13–18 mm, active;
- >18 mm, very active.

Statistical analysis

Results were expressed as the mean ± standard deviation. Data was statistically analysed using one-way ANOVA and Newman-Keuls Multiple Comparison to determine whether there were any significant with the criterion of P values < 0.05 between methanol extracts of the two species and standards, using Graphpad prism 5 Demo Software.

RESULTS AND DISCUSSIONS

The content of phenolic compounds was 58.61±19.22mg EAG/GE for methanolic extract of *Pistacia atlantica* Desf. and 56.91±7.08mg EAG/GE for aqueous extract of *Pistacia atlantica* Desf. However, the total flavonoid content of extracts were 53.92 ± 1.54mg EQ/GE extract methanolic of *Pistacia atlantica* Desf. and 52.20 ± 2.32mg EQ/GE for extract aqueous of *Pistacia atlantica* Desf.

The phenolic compound inhibits lipid oxidation by scavenging free radicals, chelating metals, activating antioxidant enzymes and inhibiting enzymes that cause oxidation reactions.

IC_{50} for DPPH radical-scavenging activity was in the order: methanolic extract (0.56 ± 0. µg/ml) > aqueous extract (5.73 ± 1.04 µg/ml l) respectively in table 1. More active than our positive control, BHT4.47 ± 0.37 µg/ml

Table-1: **DPPH free radical scavenging activity of different IC_{50} of two extract *Pistacia atlantica* Desf**

	IC_{50} (µg/ml)	$\pm SD$
BHT	4.47 ±	0.37***
méthanolic Extract	0.56 ±	0.74***
Aqueous extract	5.73 ±	1.04***

***: highly significant difference; ** very significant difference; * significant difference with P < 0,001

The role of antioxidants in the inhibition of antioxidant processes occurring in living organisms consists of: scavenging free radicals and quenching singlet oxygen, disconnection of radical reactions, chelate metals which catalyze the oxidation process, inhibition of certain enzymes Flavonoids are active in all these processes [17].

Results obtained in the present study relieved that the tested metabolite group possess a low potential microbial activity against *Bacillus Bacillus subtilis* and *Pseudomonas aeruginosa* (Tables2).

Table-2: Antimicrobial activity of standards and extracts of *Pistacia atlantica* Desf

	Bacillus subtilis	*Pseudomonas aeruginosa*
methanolic extract	7.00±0.47 ***	8,53±0.25 ***
aqueous extract	6,85±1.12 ***	8,62±0.68 ***
Standards	24.83±0,62 ***	25.950,41 ***
Control	NI	NI

***: highly significant difference; ** very significant difference; * significant difference with P <0,001

The inhibition of microorganisms by phenolic compounds may also be due to iron deprivation or hydrogen binding with vital proteins such as microbial enzymes [18].

CONCLUSION

In this study, the results indicate that the total polyphenol contents and antioxidant activity are highly dependent on the nature of solvent. *Pistacia atlantica* Desf excellent plant candidate for further investigation of individual phenolic compounds, there in vivo antioxidant activity and the different antioxidant mechanisms and also appears to be a most promising candidate from which specific antioxidant bioactive products could be developed

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