

PHYTOCHEMICAL STUDY AND IN VITRO ANTIMICROBIAL ACTIVITY OF *PISTACIA LENTISCUS* L. IN BOUMERDES MOUNTAINOUS REGION (ALGERIA)

L. Bendifallah*, A. E. Benmahfoud, Y. Hameni, S. Mameche

Department of Biology, Faculty of Sciences, University of Boumerdes, Algeria.

Received: 12 September 2014 / Accepted: 15 October 2014 / Published online: 31 December 2014

ABSTRACT

Pistacia lentiscus L. (Pistaciaceae) is among the most important medicinal plants in Algeria that is known for its antifungal and antimicrobial properties. For this study, the leaves were collected from the mountainous region of Boumerdes, in northern Algeria. In such a propitious context, the aim of this study was to enhance *Pistacia lentiscus* as a medicinal herb. For their antimicrobial activity, extracts of tannin and polyphenols were screened against three pathogenic bacterial strains and one pathogenic yeast strains. The phytochemical analysis results showed a remarkable combination of chemical components including a high content in tannins, in leucoanthocyanins, in glucosids, alkaloids, flavonoids and in saponosids. The tannins and the polyphenols have strong antimicrobial activity against some species.

Key words: *Pistacia lentiscus*; leaves; phytochemistry; biological activity.

1. INTRODUCTION

Algerian flora is characterized by its floral diversity: Mediterranean, Saharan and tropical paleo flora, estimated at more than 3000 species belonging to several botanical families, 15% are endemic[1]. This floral diversity gives the traditional pharmacopoeia invaluable wealth. The mountainous area is rich in endemic species. Among the floral resources, the pistachio. The genus *Pistacia* belongs to the family Anacardiaceae, is widespread in the Mediterranean basin[2].

Author Correspondence, e-mail: bendif_1@yahoo.fr

[ICID: 1124398](https://doi.org/10.17359/jfas.1124398)

Among 15 known species of pistachios, only 3 endemic species grow in Algeria, including the Atlas pistachio (*Pistacia atlantica* Desfontaines), the terebinth (*Pistacia terebinthus* Linnaeus) and mastic (*Pistacia lentiscus* Linnaeus)[3]. However, in the field of pistachio production in Algeria certainly still far behind compared to other Mediterranean countries.

Pistacia lentiscus L. commonly known as Mastic tree or lentisk, is known for its virtues in the treatment of ulcers, wounds and slight burns. Algerian traditional medicine mainly uses fatty oil obtained by expression of the fruits of mastic in the treatment of eczema, diarrhea, throat infections, jaundice, asthma, stomach pain, intestinal inflammation, kidney stones[4,5] minor burns, small wounds and rashes[6].

They have various biological activities, anti-ulcer, anti-atherogenic, hypoglycemic, antioxidant, anti-inflammatory, antispasmodic, antiseptic, hepatoprotective, analgesic, antipyretic, antimicrobial, insecticidal, healing[7-12] Anti-ulcer and anti-ulcer duodenal of *Pistacia lentiscus* has been reported by several authors[13], chewing mastic resin pistachio eliminates bacteria *Helicobacter pylori*[14].

Given the limitations of therapeutic chemical drugs[15], the development of research on medicinal plants has been directed towards the achievement of herbal medicines. These are presented in various dosage forms, responding to specific regulations for the evaluation of safety, therapeutic efficacy and stability[16].

Each plant has specific constituents such as tannins, glycosids, flavonoids and anthocyanins that can be used to make drugs if they are not toxic. This plant has garnered very little scientific interest so far, particularly in Algeria. For this study, the leaves were collected in spring (in March) from the mountainous region of Boumerdes, in northern Algeria. By its geographical position and its micro-climate, this region exhibits an important ecological and floristic diversity, giving rise to a very strong tradition in herbal medicine practices. In such a propitious context, the aim of this study was evaluate the antimicrobial activity of some tannins and polyphenolic compounds of extracts leaves of *Pistacia lentiscus* collected from the mountainous region of Boumerdes against a diverse range of microorganisms comprising bacteria and yeasts.

2. MATERIALS AND METHODS

2.1. Plant material

The leaves of *Pistacia lentiscus* were collected in March 2013 from the region of Boumerdes at the Lakhdaria Mountain situated in the eastern part of Algeria (Fig. 1, 2) and were identified

by Dr. Abelkrim, Professor at National School of Agronomy, Department of Botany. The leaves were shade dried at room temperature for fifteen days.

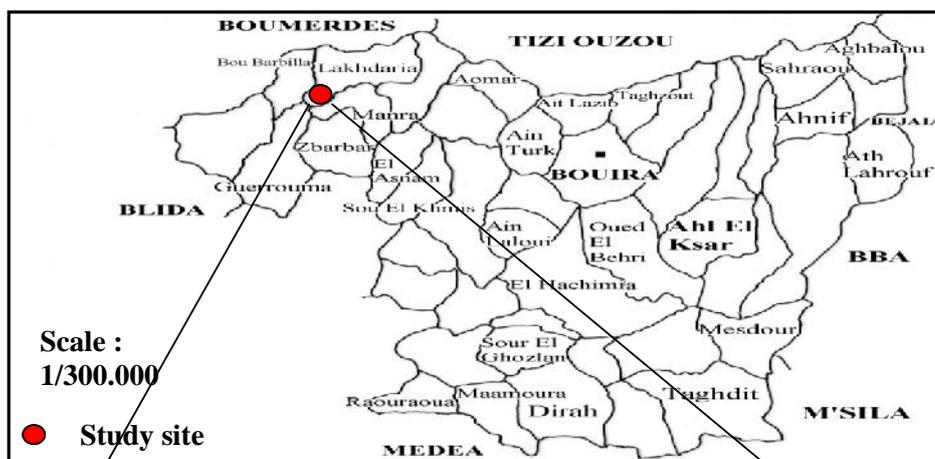


Fig.1. Geographical situation of collection station of *Pistacia*



Fig. 2- Lakhdaria mountain collection area of *Pistacia lentiscus*

2.2. Phytochemical screening

The phytochemical screening methods used are those described by Paris and Northis[17]. The phytochemical components analysed are: tannins (catechin, gallic), free quinones, coumarins, flavonoids, alkaloids, saponins, glycosids, anthocyanins, leucoanthocyanins, starch and iridoids.

2.3. Preparation of plant extracts

2.3.1. Polyphenols extract

The powdered of *Pistacia lentiscus* (30g) were extracted with methanol (100 ml) for 72 hours. After this step, the decoction was filtered. The filtrate was concentrated by evaporation in vacuo at 40 ° C. using a rotavapor and the residue was kept at 4° C[18].

2.3.2. Tannins extract

Tannins extract obtained by extraction of 15 g of powdered for 24 h in about 100 ml of solvent used (Acetone). This extract was concentrated to dryness and the residue was kept at 4° C[18].

2.3.3. Microorganisms used

The test organisms used included: three bacteria strains: *Staphylococcus aureus*, *Escherichia coli* and *Klebsiella pneumoniae* and one yeast strain *Candida albicans* these strains were collected from the Pasteur Institute (Algiers - Algeria).

2.3.4. Culture media and Antimicrobial assay

Mueller-Hinton agar (MH) and Sabouraud Dextrose agar (SD) were respectively used for bacteria and yeasts growth. Microbial cultures, freshly grown at 37°C/30°C were appropriately diluted in sterile normal saline solution to obtain the cell suspension at 10⁵ CFU: ml.

To evaluate antimicrobial activity, an agar well diffusion method was used as described by Nongpanga et al. (2008). The organisms were spread on MH and SD agar plates by cotton swab. Wells of 6 mm diameter were punched into the agar medium and filled with 50 µl of plants extracts. The plates were incubated for 24 h at 37°C for bacteria and 48h at 28°C for yeast. Determining the sensitivity of the microbial strains to the polyphenols and tannins *Pistacia lentiscus* is done using the antibiogram.

Antimicrobial activity was evaluated by measuring the inhibition zone diameter against the test organism according Durafour and colleagues (1990).

3. RESULTS

The phytochemical results (Table 1) show that *Pistacia lentiscus* is very rich in in total tannins, gallic tannins and glucosids. It is moderately rich in flavonoids, anthocyanins, leucoanthocyanins, alkaloids and saponins. Finally, we must point out that *Pistacia lentiscus* has no catechic tannins, starch and iridoïds and free quinone, it is also richer in total polyphenols than in tannins, figure 3.

The extracts leaves of *Pistacia lentiscus* have inhibitory action on all microbial strains tested, but the diameter of the zone of inhibition varies from one strain to another. The screening antibacterial activity shows that *Staphylococcus aureus* is extremely sensitive to the methanol

extract and acetone extract with a zone of inhibition (17.5 ± 3.54) mm for polyphenols (9.33 ± 1.15) mm for tannins, table 2.

Table 1. Phytochemical screening results of *Pistacia lentiscus* L.

| Substances | <i>Pistacia lentiscus</i> |
|-------------------|---------------------------|
| Total tannins | +++ |
| Catechic tannins | - |
| Gallic tannins | +++ |
| Flavonoids | ++ |
| Anthocyanins | ++ |
| Leucoanthocyanins | ++ |
| Alkaloids | ++ |
| Starch | - |
| Glucosids | +++ |
| Saponins | ++ |
| Iridoids | - |
| Free quinons | - |

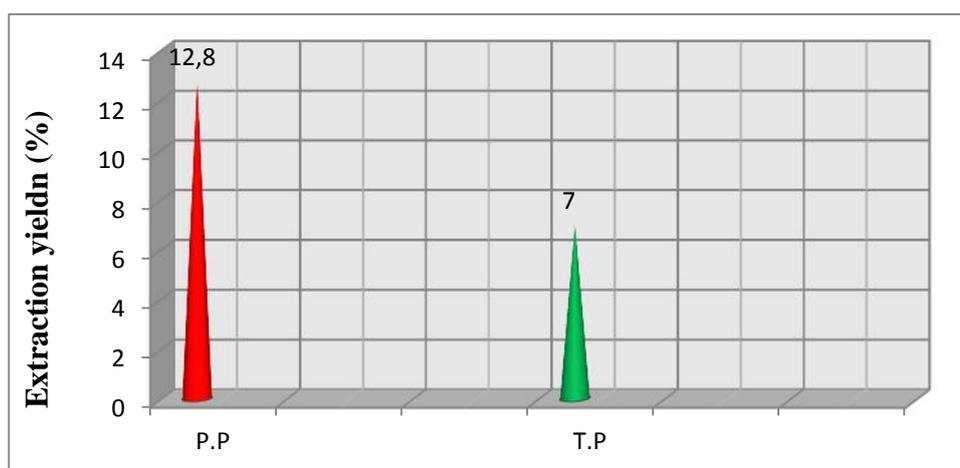


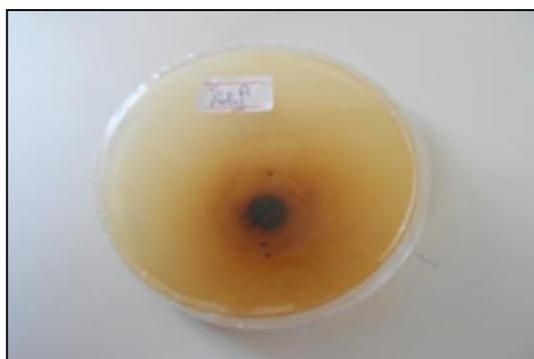
Fig.3. Graphical representation extraction of total polyphenols and tannins of *Myrtus Communis* (P.P.: Polyphenols of *Pistacia lentiscus*; T.P.: Tannins of *Pistacia lentiscus*)

The results of sensitivity testing of the microbial strains to polyphenols *Pistacia lentiscus* by the method of Antibiogram are shown in figure 4.

The results of sensitivity testing of the microbial strains to tannins *Pistacia lentiscus* by the method of Antibiogram are shown in figure 5.

Table 2. Screening antibacterial activity of *Pistacia lentiscus* leaves extracts collected from Boumerdes mountainous region (Algeria)

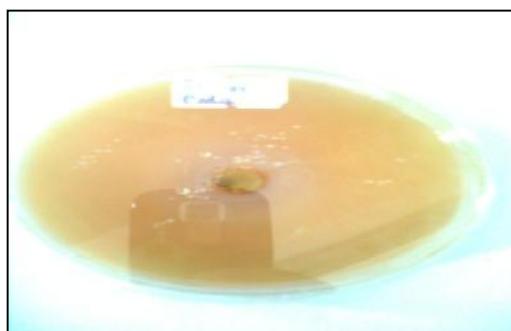
| Plant extracts | Polyphenols | Tannins |
|---------------------------------|-------------|------------|
| Microbial strains tested | | |
| Bacterial strains | | |
| <i>Escherichia coli</i> | 8± 1.41 | 5.33± 0.58 |
| <i>Staphylococcus aureus</i> | 17± 3.54 | 9± 1.15 |
| <i>Klebsiella pneumoniae</i> | 2± 0.58 | 0 |
| Yeast strains | | |
| <i>Candida albicans</i> | 5± 1.17 | 19±0.31 |



Staphylococcus aureus



Escherichia coli



Candida albicans

Fig.4. Photos showing the zones of inhibition caused by polyphenols *Pistacia lentiscus* against microbial strains

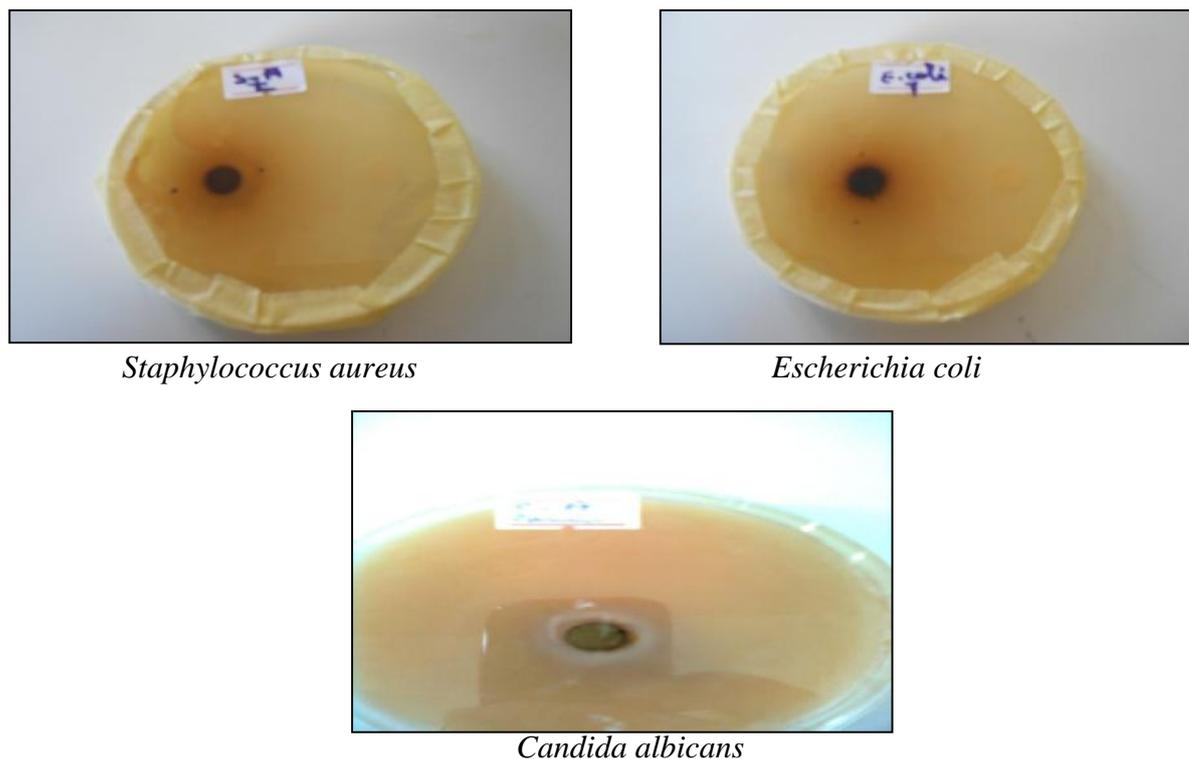


Fig.5. Photos showing the zones of inhibition caused by tannins *Pistacia lentiscus* against microbial strains

4. DISCUSSION

4.1. Phytochemical screening of *Pistacia lentiscus*

These results are similar to those Hamad *et al.* (2011). These authors reported that the leaves of *Pistacia lentiscus* are very rich in tannins, glycosids, in flavonoïds and in alkaloids. Also, Kawashty *et al.* (2000) reported the glycosids and flavonoïds are present in aerial parts of the pistachio. *Pistacia lentiscus* contains 12.8% (128 mg / g) of total polyphenols and 7% of total tannins while Hamad *et al.* (2011) note 8.9% (89mg / g) for the performance of polyphenols leaves *Pistacia lentiscus*. The difference in yield of total polyphenols and tannins of plant species is a function of intrinsic and extrinsic factors: genotype, environment, geographical origin, sunshine duration, humidity, rainfall, soil type, harvest time, temperature and drying time[18-21].

4.2. Antimicrobial activity of *Pistacia lentiscus*

The two extracts myrtle (polyphenols and tannin) have strong antimicrobial activity against the majority tested strains. These results are in agreement with those found by Hamad *et al.* (2011) that evaluated the antimicrobial activity of methanol extract of *Pistacia lentiscus* against 10 microorganisms including *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas*

aerogenosa. Also, Paiva et al (2010) show that the antimicrobial activity of *Pistacia lentiscus* against six pathogenic bacteria including *S. aureus*, *E. coli*, *P. aerogenosa*.

We found that the methanol extract (polyphenol) of leaves of *Pistacia lentiscus* have the highest antimicrobial activity against Gram + strains, especially against *S. aureus*. According our results, the tannin has antimicrobial activity against all strains tested, specially *S. aureus* and *Candida albicans*. The sensitivity of microorganisms to polyphenols, depending on the species itself and the structure of polyphenols.

5. CONCLUSION

The extracts leaves of *Pistacia lentiscus* could be good antimicrobial agents. In perspective, it would be desirable to complete this study by the extraction of active ingredients from different parts of the plant (roots, stems, leaves, flowers and seeds) and evaluation of their antimicrobial activity of several strains of pathogen.

6. ACKNOWLEDGEMENT

We sincerely thank Professor Abdelkrim from National school of Agronomy for identifying the plant of *Pistacia lentiscus*.

7. REFERENCES

- [1] Ozenda P., Flore du Sahara Ed. CNRS. PARIS, France, 1977, p 250-259.
- [2] Baily L.H., Manual of cultivated plants, 4th, edn, p. 2648, Macmillan, New York, 1958.
- [3] Boudy P. Guide du forestier. Ed .La maison rustique, Paris, 1952.
- [4] Chief R., Les plantes médicinales. ED. Solor, 1982, pp. 2276-2277.
- [5] Mouhajir F., Hudson J. B., Rejdali M., Towers G.H.N., Multiple antiviral activities of endemic medicinal plants used by Berber peoples of Morocco. Pharm. Biol. 39, 2001, p 364.
- [6] Iserin P., Encyclopédie des plantes médicinales, édition Larousse, 2001, 246 p.
- [7] Demo A., Petrakis C., Kefalas, P., Boskous D., Nutrient antioxidants in some herbs and Mediterranean plant leaves. Food Res. Int.31, 1998, p 351-354.
- [8] Pascual-Villalobos M.J. et Robledo A., Screening for anti-insect activity in Mediterranean plants. Ind. Crops Prod. 8, 1998, p 183-194.
- [9] Giner-Larza E.M., Manez, S., Giner-Pons R.M., Recio M.C., Rios J.L., On the anti-inflammatory and anti-phospholipase A2 activity of extracts from lanostane-rich species. Journal of Ethnopharmacology 73, 2000, p 61-69.

- [10] Dedoussis G.V.Z., Kaliora, A.C., Psarras S., Chiou, A., Mylona A., Papadopoulos, N.G., Andrikopoulos N.K., Antiatherogenic effect of *Pistacia lentiscus* via GSH restoration and downregulation of CD36 mRNA expression. *Atherosclerosis*, 2004, p. 174, 293- 303.
- [11] Hamdan I.I., Afifi F.U., Studies on the in vitro and in vivo hypoglycemic activities of some medicinal plants used in treatment of diabetes in Jordanian traditional medicine. *Journal of Ethnopharmacology* 93, 2004, p. 117-121.
- [12] Al-said M.S., Ageel A. M., Parmar, N. S., Tarik, M., Evaluation of mastic a crude drug obtained from *Pistacia lentiscus* for gastric and duodenal anti-ulcer activity. *Journal of Ethnopharmacology* 15. 1986.
- [13] Iauk L., Ragusa S., Rapisarda A., Franco S., Nicolosi V.M., In vitro antimicrobial activity of *Pistacia lentiscus* L. extracts: preliminary report. *Journal of Chemotherapy* 8, 1996, p 207.
- [14] Chemli R., Plantes médicinales et aromatiques de la flore de Tunisie. Faculté de pharmacie, laboratoire de pharmacognosie-phytothérapie, Monastir, Tunisia, 2004, 120-125p.
- [15] Famsworth N.R., Arkererele O., Bingel A.S., Soejarto D.D., Guo Z., *Bull. WHO* 63, 1985, p 965-981.
- [16] Paris R. and Nothis A., *Plantes médicinales, phytothérapie*. Ed. Masson, Paris, 1978, 245.
- [17] Harborne J.B., *Phytochemical methods. A guide to modern techniques of plants analysis*. Third Edition. ISBN: 0-412-57260-5 (HB) and 0-412-57270-2 (PB).1998, P 521- 530.
- [18] Lahlou M., Methods to study the phytochemistry and bioactivity of the essential oils. *Phytotherapy research*, 2004, 18: 435 – 448.
- [19] Sokmen A., Gulluce M., Akbulat H.A., Daferera D., Tepe, B., Polission, M., Sokmen M. and Sahim F., The invitro antimicrobial and antioxidant activities of the essential oils and methnol extracts of endemic *Thymus spathulifolius*. *Food control*, 2004, 15: 627 – 634.
- [20] Fellah S., Romadhane M. and Abderraba M., Extraction et étude des huiles essentielles de *Salvia officinalis* L. cueillie dans deux régions différentes de Tunisie. *J. Soc. Alger. Chim*, 2006, 16: 193 – 202.
- [21] Oussala M., Caillet S., Saucier L. and Lacroix M., Antimicrobial effects of selected plant essential oils on the growth of a *Pseudomonas putida* strain isolated from meat. *Meat Sciences*, 2006, 73: 236 – 244.

How to cite this article:

Bendifallah L. Benmahfoud A. E. Hameni Y. Mameche S. Phytochemical study and in vitro antimicrobial activity of *pistacia lentiscus* l. (pistaciaceae) in Boumerdes mountainous region (Algeria). *J Fundam Appl Sci*. 2014, 6(2), 229-237.