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# Preliminary Phytochemical Screening and Antibacterial Activity of *Pistacia atlantica* and *Prunus persica* Plants of Libyan Origin

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**Abstract:** ***Pistacia atlantica and Prunus persica are the useful plants used in traditional medicines for the treatment of various diseases. In the present investigation we have examined the preliminary phytochemicals screening of aqueous and ethanlolic leaves extracts and antibacterial activity of ethanolic extracts of both plants of Libyan origin. The qualitative phytochemical studies of aqueous and ethanolic extracts of the plants leaves were carried out by using standard testing procedures for different metabolites viz. tannins, saponins, phlobatanins, flavonoids, terpenoids, cardiac glycosides and alkaloids. The aqueous and ethanolic extracts of Pistacia atlantica showed the common presence of tannins, phlobatanins, flavonoids, terpenoids and cardiac glycosides metabolites, however, the aqueous and ethanolic extracts of Prunus persica exhibited the common presence of tannins, saponins, phlobatanins , and flavonoids metabolites . The antibacterial screening of ethanolic leaves extract of both plants was performed against Gram positive bacterial strains (Staphylococcus epidermidis ,Staphylococcus saprophyticus) and Gram negative bacterial strains (Proteus vulgaris, Eschericia coli, Citrobactor freundii). Present findings suggest that Pistacia and Prunus leaves extracts possess antibacterial effect against both gram positive and gram negative bacteria. In conclusion, phytochemicals present in both plants may be responsible for displaying different antibacterial actions. Thus these plants seem to be considered for further detailed study in an attempt to investigate the bioactive chemical entities responsible for antibacterial action against different pathogenic microbes.***

**Keywords:** Libyan plants, *Pistacia atlantica*, *Prunus persica*, Phytochemicals screening, Antibacterial activity.

## 1. Introduction

The traditional use of plants in medical practice has a long drawn history starting from ancient times and has produced excellent results in primary health care in most of the third world. Recent reports has indicated that more than 60% of world population is adopting traditional medicines as therapeutic agents for the treatment of diseases in both developed and developing countries [1]. At present medicinal plants are believed to be an important source of new remedial agents with potential therapeutic effects [2][3]. .Microbial infections are well known to a major cause of morbidity and mortality in humans and animals. Medicinal plants are to be believed as important source of obtaining antimicrobial drugs[4][6][7] [8].

*Pistasia atlantica Desf* L. (family anacardiaceae) is a deciduous tree adapted to drought and the Mediterranean climate with pinnate leaves each with seven to 9 lance-shaped leaflets and branches often have galls.. It is a common tree in mountain forests in Iran, and in some north African countries. Pistasia genus has been reported for various biological actions like antioxidant, antibacterial, antifungal, anti-diarrheal, anti-ulcer, hypoglycemic activities and many important secondary phytochemicals metabolites were evaluated for different biological actions [9],[10],[11].

The *Prunus persica* (L) Batsch (family Rosaceae) is a deciduous tree or large shrub with lanceolate tapering leaves and pink flowers native to China, Iran and in African and south American countries. Batsch tree parts have reported for its useful medicinal properties like antioxidant[12] anti-acetylcholinesterase [13], anti-inflammatory [14][15], hypermenorrhea, dysmenorrhea, leiomyoma, infertility, anti-tumour promoter and anti-oketsu syndrome (stagnation of blood circulations)[16], anthelmintic, laxative, sedative, antimalarial, hepatoprotective, antiasthmatic, anticoagulant, antifungal, cholinomimetic, calcium antagonist [17] and anti-allergic inflammatory properties via controlling calcium influx and NF-kB signaling[18]. Many important phytochemicals compounds were isolated and characterized for various biological actions.

The use of plant extracts and phytochemicals, both with known antimicrobial properties, can be of great significance in therapeutic treatments. In the last few years, there are very few reports available about the characterisation of the antimicrobial activity of crude extracts of *P. atlantica and P. persica.* So far no specific study has been carried out previously in these Libyan origin plants. This prompted us to perform pre-phytochemical testing and evaluation of the potential of extracts of two plants *viz. Pistacia atlantica* and *Prunus perscica* on standard Gram positive and Gram negative microorganism strains to better understand their antibacterial properties and efficiency.

## 2. Materials and Methods

### 2.1. Collection of Plants Materials

*Pistacia atlantica* and *Prunus persica* plants were collected from different parts of the Al-khoms city (Libya) in May/ June months of 2012. Identification of both these plants was confirmed by Plant taxonomist of the Department of Biology

of Al- Mergheb University, Al-Khoms, Libya. Leaves of each plant were collected, washed under running tap water and dried under shade and ground into fine powder in the electronic grinder., The powder was stored in plastic bags at room temperature under low humidity condition.

### 2.2. Preparation of Extract

The crude powdered plants leaves were extracted in water and ethanol separately at a 40% (w/v) concentration (20g leaves powder in 500 ml water) by using Soxhlet method for 6 - 8 h and filtered. The extracts were stored in refrigerator at 4°C until further use.

### 2.3. Phytochemicals Screening

Chemical tests were performed for the water and ethanolic leaves extracts of both the plants using standard procedures to identify the presence of various phytochemicals as described by Sofowora, 1993 [4] and Raman, 2006 [5].

#### 2.3.1. Tannins
A small quantity of each extracts was heated on water bath and filtered. A few drops of ferric chloride were added to the filtrate. A dark green solution indicated the presence of tannins.

#### 2.3.2 Saponins
Small amount of filtered plant extract was shaken and heated to boil. Frothing (appearance of creamy mass of small bubbles) shows the presence of saponins.

#### 2.3.3 Phlobatanins
Small amount of filtered extract was boiled with 2% HCl solution. Red precipitate shows the presence of phlobatanins.

#### 2.3.4. Flavonoids
Filtered extract **of** each plant was mixed with diluted NaOH and HCl was added. A yellow solution that turns colorless indicates the presence of flavonoids.

#### 2.3.5. Terpenoids
Small amount of each extract was filtered and mixed with 2 ml of chloroform ($CHCl_3$ ) and concentrated $H_2SO_4$ (3 ml) was carefully added to form a layer. A reddish brown coloration of the interface was formed to indicate positive results for the presence of terpenoids.

#### 2.3.6. Cardiac glycosides
Small amount of each extract was filtered and shaken with 1ml of glacial acetic acid. A drop of ferric chloride and a drop of concentrated sulfuric acid was added. Green blue color to upper layer and reddish brown color at the junction of two layers indicates the presence of cardiac glycosides.

#### 2.3.7. Alkaloids
Small amount of the extracts was filtered and warmed with 2% $H_2SO_4$ for two minutes. It was filtered and few drops of Dragendorff's reagent were added. Orange red precipitate indicated the presence of alkaloids.

### 2.4. Antibacterial screening

#### 2.4.1. Test Organisms
Two Gram positive strains (*Staphylococcus epidermidis ,Staphylococcus saprophyticus*) and three Gram negative bacterial strains (*Proteus vulgaris, Eschericia coli, Citrobactor freundii* ) from standard cultures were used as test strains. Standard bacterial strains were procured from Microbiology Medical Laboratory, Central Hospital, Al-Khoms, Libya.

#### 2.4.2. Determination of Antibacterial activity by disc diffusion method:
Ethanolic leaves extracts of both plants were screened for antibacterial activity against two Gram positive bacterial strains and three Gram negative bacterial strains by the paper disc diffusion method [22]. From the 50 mg/ml stock solution of ethanolic extracts of both plants, 40 µl aliquots were tranferred onto blank sterile paper discs (6 mm diameter). Dried discs were placed onto nutrient agar medium (UK) previously inoculated with a bacterial suspension and incubated at 37ºC for 24 h. Ethanol solvent was used as control to determine the sensitivity of the tested strains. After incubation, plates were examined for the presence of zones of growth inhibition, and the diameters of these zones were measured in mm . Tests were performed in duplicate under sterile conditions.

## 3. Results

### 3.1. Phytochemicals Screening

Table 1 shows the results of phytochemicals screening of aqueous and ethanolic leaves extracts of *Pistacia atlantica* and *Prunus persica* plants. *Pistacia atlantica* leaves extracts revealed the presence of tannins, saponins & alkaloids ( both absent in aqueous extract), phlobatanins, flavonoids, terpenoids and cardiac glycosides , however, *Prunus persica leaves extracts* revealed the presence of tannins, saponins, flavonoids , terpenoids and cardiac glycosides.

**Table 1:** Phytochemical screening of aqueous and ethanolic leaves extracts of *P. atlantica and P. perscica* :

| Chemical - Component | *P. atlantica* | | *P. perscica* | |
|---|---|---|---|---|
| | aq. extr. | eth. extr. | aq. extr. | eth. extr. |
| Tannins | + | + | + | + |
| Saponins | - | + | + | + |
| Phlobatanins | + | + | + | + |
| Flavonoids | + | + | + | + |
| Terpenoids | + | + | - | - |
| Cardiac glycosides | + | + | - | - |
| Alkaloids | _ | + | _ | _ |

### 3.2. Antibacterial Activity

Table 2 shows the results of antibacterial activity of both of plants leaves ethanolic extracts.The ethanolic extract of *Pistacia atlantica* exhibited antibacterial activity against a Gram positive bacterial strains (*S. saprophyticus ) among the two tested Gram positive bacterial strains* and a Gram

negative bacterial strain (*Citrobactor freundii* ) among the three tested Gram negative bacterial strains. While the ethanolic extract of *Prunus persica* revealed antibacterial activity against both tested Gram positive bacterial strains (*S. epidermidis; S.saprophyticus*) and two Gram negative strains (*Proteus vulgaris Citrobactor freundii* among the three tested Gram negative bacterial strains. Both, *Prunus persica* and *Pistacia atlantica* exhibited maximum antibacterial action against Gram positive strain *S. saprophyticus* with zone of inhibition 8 mm and 9 mm, respectively.

**Table 2:** Antibacterial activity of leaves of ethanolic extracts (50 mg/ml) of *Pistacia atlantica* and *Prunus persica* :

| Bacterial strains | *P. atlantica* | *P. perscica* |
|---|---|---|
| | Zone of inhibition (mm) | |
| *S. epidermidis* | - | 8 |
| *S. saprophyticus* | 8 | 9 |
| *Proteus vulgaris* | - | 7 |
| *Eschericia coli* | - | - |
| *Citrobactor freundii* | 7 | 7 |

**Note :** Discs injected with 40µl of ethanol solvent served as controls against each bacterial strain and showed nil zone of inhibition.

## 4. Discussion

The results of the screened phytochemicals (tannins, saponins, phlobatanins, flavonoids, terpenoids, cardiac glycosides and alkaloids) of aqueous and ethanolic extracts of *Pistacia* showed the common presence all the screened phytochemicals except saponins and alkaloids which were absent in aqueous extract, however, common presence of. tannins, saponins,flavonoids, phlobatanins was found in aqueous and ethanolic extracts of *Prunus* leaves extract. Further, The use of ethanol like solvent allows the extraction of phenolic compounds from the leaves of plants and ethanol extraction is preferentially used to obtain crude extracts of phytochemicals from plant materials in the herbal medicine industry for therapeutic applications and most of the previous studies have indicated that ethanolic leaves extracts of the plants contain richer concentrations of secondary metabolites related to microbiological actions than the aqueous and other organic solvents . Therefore, ethanolic leaves extracts of both the plants have been selected in present study for the antibacterial screening . Previous reports have indicated that the aqueous and alcoholic extract of *Pistacia* and *Prunus* are known to possess remarkable biological activities and therapeutic effects including antibacterial potential [9][10][11][12][13] [14][15][16] [17] [18] **.** No related studies was carried out previously in these plants of Libyan origin which prompted us to perform first time the present study. Good antibacterial activity was produced by both of the plants leaves ethanolic extracts (50 mg/ml concentration) against standard strains of Gram positive and Gram negative bacterial strains. Zone of inhibition of bacterial growth was observed in range of 7 to 9 mm diameter produced by both the plants extracts. It was interesting to note that ***S. saprophyticus*** (Gram positive bacterial strain) indicated maximum sensitivity to the investigated plants leaves extract of *Prunus persica* .Antibacterial action of leaf extract of *Pistacia atlantica* of different source of origin was also reported in previous findings [20]. The cause of antibacterial action might be due to the. presence of tannins, flavanoids, saponins and alkaloids in these plant extracts. Some previous phytochemical and biological screenings revealed the presence and antimicrobial action of alkaloids, flavanoids and tannins in *Pistacia* and other plants extracts[21][22][23][24]. Some studies have also shown that plants with antimicrobial activity contain bioactive constituents such as tannins, flavonoids, alkaloids and saponins [25].

It is apparent from our results that the antibacterial activity of the extracts might be attributed to the presence of tannins, flavanoids, saponins in *Prunus* and tannins, flavanoids, saponins and alkaloids in *Pistacia* plants extracts. The purified components of both these plants extracts may produce even more potency with respect to inhibition of microbes. The result of the present study signifies the potential of both plants as a source of therapeutic agents, which may provide leads in the ongoing search for antibacterial agents from plants. Further detailed studies on the isolation and purification of bioactive chemical components can reveal the exact potential(s) of the plants to inhibit growth of Gram positive and Gram negative pathogenic microbes.

## 5. Conclusion

The present investigation revealed antibacterial activity of ethanolic leaves extracts of *Prunus* and *Pistacia* plants of Libyan origin .It is concluded that ethanolic leaves extracts of both these plants showed the presence of phytochemicals( tannins, flavanoids, saponins in *Prunus* and tannins, flavanoids, saponins and alkaloids in *Pistacia),* these appear to responsible for the individual/synergistic antibacterial action against standard Gram positive and Gram negative bacterial strains. Further detailed investigation on the isolation of bio-active phyto-constituents from both these plants extracts and evaluation of their individual or synergistic role of antibacterial action against pathogenic microbes might helpful in revealing the exact potential of these plants.

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