Phytochemical investigation and antimicrobial studies on the leaf extracts of *Psoralea corylifolia* Linn.

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**INTRODUCTION**

Medicinal plants are of great importance to the health of individual and communities. The medicinal value of these plants lies in some chemical active substances that produce a definite physiological action on the human body. The most important of these chemically active (bioactive) constituents of plants are alkaloids, tannin, flavonoid and phenolic compounds. Many of these indigenous medicinal plants are also used for medicinal purposes (1).

The genus *psoralea* Linn. contains approximately 120 species distributed worldwide. These plants are mainly found in South Africa, North Africa, Australia, South America, North America, Asia and the Mediterranean region. Among the different species of *Psoralea* plants known worldwide *Psoralea corylifolia* (known as *Cullen corylifolia*) is a rare and endangered herbaceous medicinal plant, native to India. In India, it is distributed in Tamil Nadu, Uttar Pradesh, Rajasthan, Bihar, Gujarat and Andhra Pradesh (2).

*Psoralea corylifolia* Linn. (family: Fabaceae; Papilionaceae) is an erect, annual plant, upto 30–180 cm high and found throughout India. Leaves are stalked, simple, 1–3 inches long, 1–2 inches broad, firm in texture, covered with numerous black dots, hairs few, stalks up to 1 inch long hairy gland dotted. Flowers are dense, 10–30 in a bunch arising in the axils of leaves; fruits are small, subglobose, without hairs, slightly compressed. Single seeded pods are kidney shaped, 2–5 mm long, 2–3 mm broad and 1–1.5 mm thick, consisting of a sticky oily pericarp, a hard seed coat and kernel.

Various parts of this plant have been used in the folk, Siddha and Ayurvedic systems of medicine. In Ayurvedic medicine, the plant is described as stomachic, deobstruent, anthelmintic, diuretic, diaphoretic and aphrodisiac (3). Earlier studies have shown significant antibacterial activity of aqueous, alcohol, petroleum ether extracts and essential oil obtained from the seeds of *P. corylifolia* (4). The essential oil of *P. corylifolia* has shown moderate antifungal activity (5). Alcohol extracts of both leaves and seeds of *P. corylifolia* exhibited potential antifilarial activity on cattle filarial parasite *Setaria cervi* (6). No attempt has been made to study the antimicrobial activity of *P. corylifolia* extracts. Hence the present work was aimed to study the phytochemical properties and antimicrobial activity of different leaf extracts of *Psoralea corylifolia* Linn.

**ABSTRACT**

Aqueous and alcoholic extracts from *Psoralea corylifolia* leaves were screened for the presence of chemically active compounds by standard methods and evaluated for their antimicrobial activity *in vitro* by disc diffusion method. The results revealed the presence of saponins, tannins, flavonoids, glycosides, carbohydrates, tannins and phenolic compounds, gums and mucilages, fixed oils and fats. Alkaloids were not detected from any of the leaves extract under study. Aqueous and alcoholic extracts exhibited broad-spectrum antibacterial and antifungal activity against *Eschericia coli*, *Pseudomonas aeruginosa*, *Staphylococcus pyogenes* and *Candida albicans*. Alcoholic extract is better than that of aqueous extract of *P. corylifolia* leaves in respect to their antimicrobial activity and the broad spectrum of activity makes it a promising indigenous drug.

**Keywords:** Alcoholic extract, Antimicrobial activity, Aqueous extract, Leaves, *Psoralea Corylifolia*.
MATERIALS AND METHODS

Plant materials

The plant material of *P. corylifolia* was collected from the survey of medicinal plant unit, Regional Research Institute of Unani Medicine, Aligarh (U.P.), India. The plant was authenticated by Dr Athar Ali Khan, Department of Botany, A.M.U., Aligarh, where the voucher specimen has been deposited (Voucher No. 1122).

Preparation of the plant extract

Dried and powdered leaves of *P. corylifolia* were extracted with ethanol and water, separately. The crude ethanol and aqueous extracts were dried and dissolved in 95% ethanol and distilled water before use (6).

Phytochemical screening

The leaves extract of *P. corylifolia* were analyzed for the presence of alkaloid, saponin, tannins and phenolic compounds, glycosides, flavonoids, lignins, carbohydrates, gums and mucilages, phytosterols, fixed oil and fat, proteins and free amino acids according to standard methods (7, 8).

Screening for alkaloids

A small portion of solvent free alcohol extract and aqueous extracts were stirred separately with few drops of dilute hydrochloric acid and filtered. The filtrate may be tested carefully with various alkaloidal reagents such as Mayer’s reagent, Dragendorff’s reagent, Hager’s reagent and Wagner’s reagent. Precipitation in any of the 4 test indicates the presence of alkaloids.

Screening for saponin

About 0.5 g of the plant extract was shaken with water in a test tube. Frothing, which persist on warming was taken as a preliminary evidence for the presence of saponin.

Screening for tannins and phenolic compounds

Extract of the sample was treated with 5% ferric chloride test solution. The resultant color was noted. A violet color indicated the presence of hydrolysable tannin. Or into 1% solution of gelatin containing 10% sodium chloride in a beaker, 0.5 g of the extract was added and shaken to dissolve. A white precipitate observed indicates the presence of tannin. Or into 10% lead acetate solution, 0.5 g of the extract was added and shaken to dissolve. A white precipitate observed indicates the presence of tannins and phenolic compounds.

Screening for phytosterol

*(Libermann Burchard test)*

1 g of extract was dissolved in few drops of dry acetic acid and 3 ml of acetic anhydride was added followed by few drops of concentrated sulphuric acid. A bluish green color indicates the presence of phytosterol.

Screening for flavonoid: Shinoda’s test

Extract is dissolved in alcohol. To that piece of magnesium followed by concentrated hydrochloric acid is added. Red color indicates the presence of flavonoids. Or 5 ml of 20% sodium hydroxide was added to equal volume of the sample extract. A yellow solution indicates the presence of flavonoid.

Screening for glycosides

*(Borntrager’s test)*: Extract was treated with chloroform and the chloroform layer was separated. To this equal quantity of dilute ammonia solution was added. A layer of pink, red or violet color indicates the presence of glycosides.

Screening for carbohydrates:

Minimum amount of extracts were dissolved in 5 ml distilled water and filtered. The filtrate was subjected to test for carbohydrates.

a. *Molisch’s test*: The filtrate was treated with 2–3 drops of 1% alcoholic α-naphthol and 2 ml of concentrated sulphuric acid was added along the sides of the test tube. A purple color indicates the presence of carbohydrates.

b. *Fehling’s test*: Filtrate was treated with 1 ml Fehling’s solution and heated. An orange-red precipitate indicates the presence of carbohydrates.

c. *Legal’s test*: Extract was treated with chloroform and the chloroform layer was separated. To this equal quantity of dilute ammonia solution was added. Purple color in ammoniacal layer indicates the presence of carbohydrates.

Screening for fixed oil and fat:

Small quantity of various extracts was separately pressed between two filters. Appearance of oil stain on the paper indicates presence of fixed oil.

Few drops of 0.5 N alcoholic potassium hydroxide were added to small quantity of various extracts along
with phenolphathlein. The mixture was heated on a water bath for 1–2 h. Formation of soap or partial neutralization of alkali indicates the presence of fixed oil and fats.

**Screening for proteins**

Dissolve small quantities of various extracts in few ml of water and treated with

a. *Millon’s reagent*: Appearance of red color indicates presence of proteins.

b. *Ninhydrin’s reagent*: Appearance of purple color indicates presence of proteins.

c. *Biuret test*: Equal volume of 5% solution of sodium hydroxide and 15% solution of copper sulphate were added. Pink color indicates presence of proteins.

**Screening for lignins:**

Extract was treated with alcohol followed by addition of phloroglucinol and hydrochloric acid. Appearance of red color indicates presence of lignins.

**Screening for gums and mucilages:**

About 10 ml of various extracts were treated with absolute alcohol and filtered. Occurrence of precipitation indicates the presence of gums and mucilages.

**Antimicrobial activity on various extracts of *Psoralea corylifolia* leaves**

*Microorganism used*: *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus pyogenes* and *Candida albicans* are the microorganisms were used for present *in vitro* antimicrobial assay. All the organisms were obtained from the National Collection of Industrial Microorganism (NCIM), National Chemical Laboratory, Pune, India.

**Determination of antimicrobial activity**

Disc diffusion assay method was used to screen the extracts for antimicrobial activity against *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus pyogenes* and *Candida albicans*. The organisms to be tested were inoculated on Muller hinton agar. After an incubation period of 24 h at 37°C, three or four colonies were isolated from these media and inoculated into 4 ml of Muller hinton agar broth. After an incubation period of 2 h at 37°C, the cultures were adjusted with sterile solution to obtain turbidity comparable to that of Me Farland No. 0.5 standard. Muller hinton agar plates were swabbed with the respective broth culture of the organisms and incubated over night at 37°C. The diameters of the inhibition zone were measured in millimeter.

**Activity Index:**

The zone of inhibition in extract and the standard antimicrobial agent were used to calculate the activity index.

\[
AI = \frac{\text{Zone of inhibition by extract}}{\text{Zone of inhibition by standard antimicrobial agents}}
\]

**Proportion Index:**

Number of positive results obtained for aqueous and alcoholic extract of plant part was against all the

<table>
<thead>
<tr>
<th>Sl. No.</th>
<th>Phytochemical constituents</th>
<th>Aqueous extract</th>
<th>Alcoholic extract</th>
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</thead>
<tbody>
<tr>
<td>1.</td>
<td>Alkaloids</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>2.</td>
<td>Carbohydrates</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>3.</td>
<td>Fixed oils and fats</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>4.</td>
<td>Saponins</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>5.</td>
<td>Tannins and phenolic compounds</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>6.</td>
<td>Proteins and amino acid</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>7.</td>
<td>Gums and mucilage</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>8.</td>
<td>Flavonoids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>9.</td>
<td>Lignin</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>10.</td>
<td>Phytosterol</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>11.</td>
<td>Glycosides</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

+= Present, = Absent
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**RESULTS AND DISCUSSION**

The study on *P. corylifolia* leaves extract revealed the presence of saponins, tannins, flavonoids, glycosides, carbohydrates, tannins and phenolic compounds, gums and mucilages, fixed oils and fats. The leaves did not show the presence of alkaloids in any of the extracts that were tested for its presence (Table 1).

The results of the antimicrobial activity presented in table 2 shows that the alcoholic extract exhibited appreciable antimicrobial property by inhibiting the growth of all microorganisms, whereas the aqueous extract inhibited the growth of all microorganisms except *E. coli*. Generally, proportion index of antimicrobial activities of *P. corylifolia* extracts shows the highest activity in alcoholic extract as presented in table 2.

The various phytochemical compounds detected are known to have beneficial importance in industrial and medicinal sciences. There are records that show the benefits of these compounds detected from *P. corylifolia* for example:

Saponin is used as a mild detergent and in intracellular histochemistry staining to allow antibody access to intracellular proteins. In medicine, it is used in hyperchloles-trolaemia, hyperglycaemia, antioxidant, anticancer, antiinflammatory, weight loss and antifungal etc. It is also known to have antimicrobial properties.

Tannin is reported to exhibit antiviral, antibacterial, antitumor and antimicrobial activities. It was also reported that certain tannin are able to inhibit HIV replication selectivity and is also used as diuretic plant tannin have been recognized for their pharmacological properties and are known to make trees and shrubs a difficult meal for many caterpillars (9).

**Flavonoid** have been referred to as nature’s biological response modifiers because of strong experimental evidence of their inherent ability to modify the body’s reaction to allergies, virus and carcinogens. They show antiallergic, anti-inflammatory, antimicrobial and anticancer activity.

**CONCLUSION**

Our observations confirm that the alcoholic extract is better than that of aqueous extract of *P. corylifolia* leaf in respect to their antimicrobial activity. Phytochemical analysis revealed the presence of saponins, tannins, flavonoids, glycosides, carbohydrates, tannins and phenolic compounds, gums and mucilages, fixed oils and fats. We recommend further research on this plant leaves for possible isolation and characterization of the various chemical active substances.

**REFERENCES**


### Table 2: Antimicrobial activity of *P. corylifolia* extracts

<table>
<thead>
<tr>
<th>Sl. No.</th>
<th>Plant extract</th>
<th>Zone of inhibition (mm) (± SEM)</th>
<th>Proportion Index (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>SAA</td>
<td>17.3 ± 0.2</td>
<td>14.1 ± 0.5</td>
</tr>
<tr>
<td>2</td>
<td>Aqueous extract</td>
<td>----</td>
<td>8.0 ± 0.4</td>
</tr>
<tr>
<td>3</td>
<td>Alcoholic extract</td>
<td>10.1 ± 0.2</td>
<td>9.4 ± 0.2</td>
</tr>
</tbody>
</table>

SAA- Standard antimicrobial agent (Riboflavin)