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## Research Article

## ANTIBACTERIAL ACTIVITY OF ROOT EXTRACT OF THEVETIA PERUVIANA (PERS.) SCHUM

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#### ABSTRACT

The present investigation has been evaluated to find out the antibacterial activity and phytochemical screening of the root extract of *Thevetia peruviana* (Pers.) K Schum. The antibacterial efficacy of root extracts of *Thevetia peruviana* was performed in different solvents like Petroleum ether, Ethanol, Methanol, Acetone and Distilled water by using the Soxhlet apparatus. The antibacterial activity was observed against test organisms like *Escherichia coli, Salmonella typhi, Klebsiella pneumonae, Pseudomonas aeruginosa, Staphylococcus aureus* and *Shigella flexinari*. The results show that the inhibitory action of petroleum ether extract was relatively higher than that of other extracts against *Salmonella typhi*. The Methanol and Acetone extract also shows marked inhibition activity against all test organisms. The Amoxicillin was used as a standard. The phytochemical screening reveals the presence of Alkaloids, Flavonoids, Saponins, Tannins, Carbohydrates and Terpenoids.

Keywords: Thevetia peruviana, antibacterial activity, Phytochemical screening.

## INTRODUCTION

Herbal remedies with antibacterial activities are prepared from a variety of plant parts such as stem, root, bark, leaf etc. by various pharmacological industries to overcome the infectious diseases. Scientists were screening plants for their antimicrobial usefulness as an alternative source to the antibiotics. The plant drugs proves relatively non-toxic safe and even free from serious side effects1. The medicinal importance of plants is because of the presence of various important bio chemicals like alkaloids, flavonoids, glycosides, tannins, steroids, carbohydrates etc. These photo chemicals are stored in the various organs of plants and could be the possible source of new and effective herbal medicine to treat the dreadful infectious diseases. However, it is necessary to screen the phytochemicals and their efficacy to generate safe and effective drugs<sup>2</sup>. The yellow oleander i.e. Thevetia peruviana is an evergreen plant oftenly wild grow or as an ornamental tree grows up to 10-15 feet high, belonging to the family Apocynaceae. Its leaves are linear lanceolate about 13-15 cm in length and glossy green in color. The leaves covered with waxy coating. Yellow oleander flowers are bright yellow and funnel-shaped with 5 petals. The fruits are somewhat globular, fleshy having 4-5 cm diameter and green in color and become black on ripening. Each fruit contains a nut, which is longitudinally and transversely divided<sup>3</sup>. The entire plant contains milky juice which is poisonous. The leaves, fruits, seeds and roots are considered as potential source of biologically active compounds<sup>4</sup>, such as insecticides, fungicides and bactericides<sup>4-10</sup>. The root paste is used against tumours. The plant has Cardiac glycosides and is considered to be a promising drug for congestive heart failure<sup>11</sup>. The present study reveals the antibacterial assay and phytochemical screening of root extracts of Thevetia peruviana.

# MATERIALS AND METHODS

#### **Collection of plant material**

The fresh roots of *Thevetia peruviana* were collected from Udgir Tahshil of latur district. The collected sample was

washed with tap water to remove soil particles and dirt. The material was dried in shade and powdered by using mechanical grinder.

### Preparation of extracts

About 25-30 g powder of root was extracted with different solvents by using Soxhlet apparatus until the extractive was clear. The extracts were concentrated in water bath at 50°C then extracts were stored in refrigerator at 4°C for further use.

# **Phytochemical Screening**

All the five extracts were subjected for phytochemical screening by dissolving them in respective solvents (1 g/ml). The extracts show the presence of Alkaloids, Flavonoids, Saponins, Tannins, Carbohydrates, Terpenoids and Amino acids<sup>12-13</sup>.

### **Detection of Alkaloids**

To the test solution, add 1 % HCl and any of Mayers reagents, Wagners reagent, or Dragendroff reagent added. A creamish or brownish red or orange precipitate indicates the presence of alkaloids.

## **Detection of Flavonoids**

To the test solution of each extracts added 5 ml of dilute ammonia solution followed by addition of conc.  $H_2SO_4$ . A yellow color in each extract indicates the presence of flavonoids.

## **Detection of Tannins**

To the test solution added few drops of ferric chloride solution. Formation of brownish green or blue black coloration indicates the presence of tannins.

# **Detection of Saponins**

Extracts were diluted in 5 ml of distilled water and shaken vigorously; formation of a stable persistent froth indicates the presence of saponins

## **Detection of Terpenoids**

Extracts were mixed with 2 ml of chloroform, and 3 ml of conc. H<sub>2</sub>SO<sub>4</sub> was carefully added. Formation of a reddish brown color at the interface indicates the presence of terpenoids.

# **Detection of Carbohydrates**

The extract was boiled on water bath and 1 ml of Fehlings solutions A and B were added respectively. A red precipitate indicates the presence of sugar.

#### **Detection of Proteins**

To the extract few drops of conc. Nitric acid added. Formation of yellow color indicates the presence of proteins.

### **Test Micro-organisms**

The microbial strains *Escherichia coli* MTCC (443), *Salmonella typhi* MTCC (734), *Klebsiella pneumonae* MTCC (2653), *Pseudomonas aureginosa* MTCC (424), *Shigella Flexinari* MTCC (1457), *Staphylococcus aureus* MTCC (96) were obtained from Institute of Microbial Technology (IMTECH) Chandigarh, India.

#### Preparation of discs

10 mm discs were prepared and sterilized in autoclave. These discs were soaked in crude different extracts like Petroleum ether, Ethanol, Methanol, Acetone and distilled water. The standard drug Amoxicillin was used as control.

#### **Determination of Antibacterial Activity**

Antibacterial activity of the *Thevetia peruviana* root extracts were determined by the agar disc diffusion method<sup>14</sup>.

Nutrient agar was prepared for the study. Each plate of Nutrient agar was swabbed with each bacterial strain by using sterile glass spreader. The soaked dried discs were placed on the surface of each inoculated plate. The plates were allowed for diffusion for ½ an hour and then transferred to incubator at 37°C for 24 hours. Standard discs of Amoxicillin were also placed as positive control and Acetone was used as negative control. The antibacterial activity of root extracts was determined by measuring the diameter of zone of inhibition in mm.

#### RESULTS

The preliminary phytochemical screening of Thevetia peruviana root extracts revealed that the presence of phytochemical compounds like Alkaloids, Flavonoids, Saponins, Tannins, Terpenoids, Carbohydrates, Amino acids in all the extracts. The Saponins are not found in Distilled water extract (Table 1). All the extracts show marked inhibition activity against all test organisms. Of all the five extracts the Petroleum ether extract shows significant activity against all the test micro-organisms which shows maximum zone of inhibition (32 mm) against S. typhi; (28 mm) against S. aureus: (26 mm) against E. coli: (24 mm) against K. pneumonae; (23 mm) against Sh. flexinari; and (22 mm) against P. aureginosa. The standard drug Amoxycillin shows (30 mm) zone of inhibition (Table 2). The methanol extract is effective against S. typhi (24 mm) and K. pneumonae (19 mm) and the acetone extract is effective against K. pneumonae (20 mm). The ethanol root extracts of Thevetia peruviana shows moderate inhibitory activity against all test organisms (Table 2).

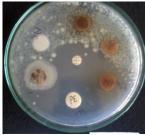
Table 1: Preliminary Phytochemical screening of root extracts T. peruviana

	Alkaloids	Flavonoids	Saponins	Tannins	Amino acids	Carbohydrates	Terpenoids
Petroleum ether	+	+	+	+	+	+	+
Ethanol	+	+	+	+	+	+	+
Methanol	+	+	+	+	+	+	+
Acetone	+	+	+	+	+	+	+
Distilled water	+	+	-	+	+	+	+

<sup>+:</sup> Present, -: Absent

Table 2: Antibacterial Assay of root extracts T. peruviana

Extracts	Zone of		inhibition in mm			
	E. coli	S. typhi	K. pneumonae	P. auriginosa	S. aureus	S. flexinari
Petroleum ether	26	32	24	22	28	23
Ethanol	16	1	18	12	12	12
Methanol	18	24	19	14	12	12
Acetone	17	16	20	14	14	13
Distilled water	-	-	-	-	-	-
Standard	28	30	30	24	26	24



1. S.typhi



2. S. aureus

Figures 1-2: Antibacterial assay of petroleum ether extracts of T. peruviana

## DISCUSSION

The antibacterial assay of present study reveals that the petroleum ether root extract shows maximum inhibitory activity against all test organisms. Earlier, studies exhibited similar observation from root and bark extract of *Nerium oleander*<sup>15</sup>. The antibacterial efficacy is due to the presence of important secondary metabolites such type of results were obtained, in methanolic extracts of *Thevetia peruviana*<sup>16</sup>. This indicates that the medicinal efficacy of a plant is because of the presence of important secondary metabolites present in them. The medicinal potential of a plant is unique which is based on the concept that the combination of secondary products in a particular plant species<sup>17</sup>.

#### CONCLUSION

The petroleum ether root extract of *Thevetia peruviana* shows maximum inhibitory activity against all test organisms as compared to standard antibiotic Amoxicillin. Therefore, these results are useful in to treat the infectious diseases created by test microorganisms. Further, pharmacological and phytochemical investigation may useful in to generate new effective antibacterial drugs in future.

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