ANTIBACTERIAL EFFICACY AND PHYTOCHEMICAL OBSERVATION OF SOME INDIAN MEDICINAL PLANTS

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ABSTRACT:

The aim of the study is to evaluate the antibacterial activity of Pistacia integerrima, Cedrus deodara and Gymnema sylvestre against seven different microorganisms such as Escherichia coli, Salmonella typhi, Klebsiella pneumoniae, Proteus vulgaris, Pseudomonas, Bacillus subtilis and Staphylococcus aureus by using disc diffusion method. Preliminary studies with ethanol extract and water extract of plants indicated that the growth of test organism was markedly inhibited by ethanol extract of Pistacia integerrima and Gymnema sylvestre. But in case of Cedrus deodara, water extract was more effective. Efficacy of plant extract which showed variable inhibitory activity against each bacteria was compared to standard antibiotic (tetracyclin). The two extracts were subjected to qualitative analysis to find out phytoconstituents present. Results showed that Pistacia integerrima contained all the phytochemicals, so exhibited higher antibacterial activity.

KEY WORDS:

Anti bacterial activity, Pistacia integerrima, Cedrus deodara, Gymnema sylvestre, phytochemical activity.

INTRODUCTION:

Presently in developing countries, synthetic drugs are not only expensive and inadequate for the treatment of disease but are often adulterated and show side effects. So there is a need to search for plants of medicinal value. Through out history, plants have provided human with an essential source of medicine. The use of plants for health care in India dates back close to 5000 years. A large scale screening of Indian plants for biological activity was conducted. Essential oils from different plant species are known to exhibit various kinds of biological activities including antifungal, antimicrobial, cytostatic, insecticidal, antioxidant and many other useful properties.

According to the reports of many
researchers, antibacterial resistance is a worldwide problem. Over the last 40 years, intensive efforts have been taken to discover clinically useful antibacterial and antifungal drugs.

**Pistacia integerrima**, a member of the Anacardiaceae (Tamil Name: Karkata-Shing) is a native of the south Alpine Himalayas. On the leaves and petioles of this plant, are found peculiar gall-like excretions, which give the appearance of horns from a distance (Chopra, et al., 1985). They are used in the treatment of coughs, phthisis, asthma, dysentery, etc (Chandrasekhar, 1950). Galls are produced on the tree in response to attack by insects and are expectorant and toxic.

**Gymnema sylvestre**, a member of the Asclepiadaceae (Tamil name: Shirukarnija) is a stout, large, woody, climbing plant, which grows commonly in central Southern India, in the Western Ghats and in Goa territory (Nadkarni, 1994). The leaves of the plant is used for diabetes. The plant is bitter, astringent, acrid, thermogenic, anti-inflammatory, digestive, liver toxic, emetic, diuretic, stimulant, anthelmintic, alexipharmic, laxative, cardio tonic, expectorant, antipyretic, akrine and tonic.

**Cedrus deodara**, a member of the Pinaceae (Tamil name: Tavatara) grows throughout Western Himalayas and lives to a great age. The hard wood of plant is carminative, diaphoretic and diuretic. It is used in the treatment of fever, flatulence, pulmonary and urinary disorders, rheumatism, piles, kidney stones, insomnia and diabetes. The bark is astringent and has proved to be useful in the treatment of fever, diarrhea and dysentery.

A review of literature revealed that the various parts of **Pistacia integerrima**, **Gymnema sylvestre**, and **Cedrus deodara** have not been subjected to screening for antibacterial property. So in the present study an attempt was made to study the antibacterial activity of selected plants (**Pistacia integerrima**, **Gymnema sylvestre**, **Cedrus deodara**) in water and ethanol extract against the selected pathogenic bacterial strains (Escherichia coli, Salmonella typhi, Klebsiella pneumoniae, Proteus vulgaris, Pseudomonas species, Bacillus subtillis and Staphylococcus aureus).

**MATERIALS AND METHODS:**

**Collection of plant material:**

The useful parts of the plant- galls of **Pistacia integerrima**, leaf of **Gymnema sylvestre** and hard wood of **Cedrus deodara** were collected from Ayurvedic medical shop in Kerala and Tamilnadu, India and preserved in the Department of Biochemistry, Dr. N.G.P. Arts and Science College, Coimbatore, India.
Selection of Bacterial Strains:

The test organisms Escherichia coli, Salmonella typhi, Klebsiella pneumoniae, Proteus vulgaris, Pseudomonas species, Bacillus subtilis and Staphylococcus aureus were collected from Post Graduate and Research Department of Microbiology, Dr. N.G.P Arts and Science College, Coimbatore, India.

Preparation of plant extract:

The collected parts of medicinal plants (gall, leaf and hard wood) were dried and powdered. 10 gram of the plant powder was weighed and mixed with 100 ml of water and ethanol to obtain water and ethanol extract respectively. This mixture was filtered through Whatmann No. 1 filter paper. Then the filtered solution was used as plant extract (1% water and ethanol extract).

Preparation of medium:

Nutrient Agar is composed of 5 g of Pentose, 5 g of Sodium Chloride, 3 g of Beef extract, 2 g of Yeast extract, 2% of Agar and 1000 ml of distilled water.

In the present study, 7 g of nutrient agar was used directly and dissolved in 250 ml of sterile distilled water. The pH was adjusted to 7.2 and the solution was autoclaved at 121°C for 15 minutes at 15 lbs.

Antibiotic Sensitivity Assay (Disc diffusion method) (Kirby-Bauer method)

100 µl of 5 hour culture was poured into nutrient agar media by a micropipette. The culture mixed nutrient agar media was poured into the petridish and allowed to solidify. Antibiotic discs were placed on the medium with the help of sterile forceps. These plates were incubated at 37°C for a period of 24 hours. The sensitivity zone around the discs of each organism plate was measured using zone diameter scale. The antibacterial sensitivity of the antibiotic Tetracycline (standard) was also measured.

Phytochemical Screening:

To determine the presence of phytochemicals in selected plants the extracts were subjected to tests as per the method of Kokate, Trease and Evans.

RESULTS AND DISCUSSION:

The antibacterial efficacy of both water and ethanol extract of selected plants is given in table I. All the plants exhibited different degree of antibacterial activity which is compared with the reference standard antibiotic, tetracycline.

Depending on the measured value of the complete inhibition diameter of the zone (including the disc in millimeter), the antibacterial activity can be classified into the following types, such as > 12 mm zone of inhibition.
– highly sensitive, 9-12 mm zone of inhibition – moderate sensitive, 9-6 mm zone of inhibition resistant 22.

The ethanol and water extract of Pistacia integerrima was highly sensitive to the selected pathogen. In case of Cedrus deodara, water extract showed more antibacterial activity when compared to that of ethanol extract.

Among the water extract and ethanol extract of 3 medicinal plant used in this efficacy test, Pistacia integerrima showed more significant antibacterial activity when compared to others because it contains almost all of the phytochemicals. Gymnema sylvestre showed lesser activity against the selected pathogen because of absence of some phytochemicals. These results suggest that the flavanoids found in the tested plants may be responsible for antibacterial activity. Several types of alkaloids, steroids and proteins have been reported to have antibacterial activity 23, 24. When compared to the antibacterial activity of control (tetracycline) the activities of the plant extract were less effective. Tetracycline showed higher sensitivity to selected pathogen.

Phytochemical constituents like alkaloids, flavanoids, phenol, carbohydrates, steroids, proteins, tannins, saponins, glycosides and resins were analysed quantitatively and are reported in table II. Alkaloids, phenols, glycosides, resins and tannins were detected in all the selected plant extracts (both in water and alcohol). Steroids were absent in all the screened extracts. Flavanoids were absent in extract of Gymnema sylvestre while proteins in extract of Cedrus deodara. Water extract of Pistacia integerrima showed the presence of all phytochemical constituents except steroids.

CONCLUSION:

The selected plants (Pistacia integerrima, Cedrus deodara and Gymnema sylvestre) showed significant antibacterial activity and so we can use these medicinal plants to control disease and infections caused by the selected pathogens.
| S.No | Organism                      | Pistacia integerrima | Cedrus deodara | Gymnema sylvestre | C   |
|------|------------------------------|----------------------|----------------|-------------------|-----|
|      |                              | W.E   | E.E | W.E   | E.E | W.E | E.E |     |
| 1    | Escherichia coli             | 16    | 18  | 6     | 8   | 7   | 7   | 19  |
| 2    | Salmonella typhi             | 15    | 15  | 7     | 12  | 9   | 8   | 21  |
| 3    | Klebsiella pneumoniae        | 16    | 17  | 5     | 8   | 10  | 8   | 19  |
| 4    | Proteus vulgaris             | 14    | 16  | 6     | 9   | 9   | 7   | 21  |
| 5    | Pseudomonas                  | 15    | 15  | 5     | 10  | 7   | 6   | 20  |
| 6    | Bacillus subtilis            | 14    | 16  | 7     | 8   | 10  | 8   | 25  |
| 7    | Staphylococcus aureus        | 13    | 17  | 6     | 9   | 8   | 7   | 17  |

C - control (tetracycline), W.E - water extract, E.E - ethanol extract
TABLE II
ANALYSIS OF ACTIVE PHYTOCHEMICAL CONTITUENTS IN 3 MEDICINALLY IMPORTANT PLANTS

| S No | Tests          | Pistacia integerrima | Cedrus deodara | Gymnema sylvestre |
|------|----------------|----------------------|----------------|-------------------|
|      |                | W.E | E.E | W.E | E.E | W.E | E.E |
| 1    | Alkaloids      | +   | +   | +   | +   | +   | +   |
| 2    | Flavanoids     | +   | +   | +   | +   | -   | -   |
| 3    | Saponins       | +   | -   | -   | -   | +   | +   |
| 4    | Carbohydrates  | +   | +   | -   | -   | -   | -   |
| 5    | Proteins       | +   | +   | -   | -   | +   | +   |
| 6    | Phenols        | +   | +   | +   | +   | +   | +   |
| 7    | Steroids       | -   | -   | -   | -   | -   | -   |
| 8    | Glycosides     | +   | +   | +   | +   | +   | +   |
| 9    | Resins         | +   | +   | +   | +   | +   | +   |
| 10   | Tannins        | +   | +   | +   | +   | +   | +   |

(+)- Phytochemical present, (-)- phytochemical absent
WE - Water Extract, EE - Ethanol Extract

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