SCREENING FOR ANTIBACTERIAL ACTIVITY OF TWELVE MEDICINAL PLANTS USED IN SOUTH INDIA FOLKLORIC MEDICINE

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Summary

Twelve medicinal plants were screened namely *Sesbania aegyptica* (Poir) Pers, *Eupatorium glandulsom* (Michx.), *Vitex trifolia* (L.), *Asteracantha longifolia* (Ness), *Berberis tinctoria* (Lesch), *Passiflora edulis* (Sims), *Spheranthus indicus* (L.), *Solanum trilobatum* (L.), *Delonex elata* (L.) Gamble, *Gymnema sylvestre* R.Br, *Spathodea campanulata* (Beauv) and *Euphorbia tirucalli* (L.) for potential antibacterial activity against 5 medically important bacterial strains *Escherchia coli* MTCC734, *Staphylococcus aureus* MTCC737,*Salmonella typhi* MTCC734, *Pseudomonas aeruginosa* MTCC737 and *Bacillus subtilis* (MTCC2423). The antibacterial activity of aqueous and methanol extracts were determined by agar disc diffusion method. The methanol extracts of all 12 medicinal plants showed pronounced antibacterial activity than aqueous extracts. *Solanum trilobatum* was found to possess significant antibacterial activity against *Staphylococcus aureus.*

Key words: Medicinal plants, Antibacterial activity, aqueous extracts, Methanol extracts

Introduction

The use of traditional medicine is wide spread in India especially within poor and tribal population. This may be due to lack of modern doctors, medicine and as well as the high expenses associated with such treatments (1). A major part of the total population in developing countries still uses traditional folk medicine obtained from plant resources (2).There are many approaches to the search for new biologically active principles in medicinal plants (3). India is the largest producer of medicinal herbs and is appropriately called the botanical garden of world (4). Practioners of Ayurveda and Unani system of medicine regularly employ a large number of Indian medicinal plants as therapeutic agents. There are about 45,000 plant species in India with concentrated hot spot in the region of Himalayas, Western Ghats & Andaman and Nicobar islands.

The officially documented plants with medicinal potential are 3000, but traditional practioners use more than 6000 plants. Emergence of drugs resistant strains due to indiscriminate use of commercial synthetic drugs is always concerned as a growing global problem. Hence, there is a pressing need to search for new antimicrobial substances alternative to commercial chemotherapeutic agents. The resurgence of Ethanomedicines has gained greater importance to overcome the drug resistant strains under infectious status. Herbal medicine represents one of the most important fields of traditional medicine all over the world. Contrary to the synthetic drugs, antimicrobials of plant origin are not associated with many side effects and have an enormous therapeutic potential to heal many infectious diseases. The secondary metabolites of plants may exert their action by resembling endogenous metabolites, ligands, hormones, signal transduction molecules or neurotransmitters and thus have beneficial effect on humans due to similarities in their potential target sites. For example Vincristine (an antitumor drug), digitalis (heart regulators) and ephedrine (a bronchodilator used to decrease respiratory congestion) were all originally discovered through research on plants. Over the last four decades, intensive efforts have been made to discover clinically used antibacterial and antifungal agents (2) (5) (6) (7) (8) (9) (10) (11) (12) (15). Considering above facts this paper supports a systematic attempt to study the antibacterial properties of twelve medicinal plants

Materials and Methods

Plant material

The plant materials used for the study were collected from Coimbatore region and shade dried at room temperature for 15 days.

Extraction

For solvent extraction, 50 g of air-dried powder was taken in 250 ml of organic solvent (Methanol and Hot water) in a conical flask, plugged with cotton wool and then kept on a rotary shaker at 190-220 rpm for 24 h. After 24 hours the supernatant was collected and the solvent was evaporated at room temperature. The extracts were stored at 4^0C in airtight bottles.

Bacteria Used

The bacterial strains such as *Escherichia coli* (MTCC734), *Staphylococcus aureus* (MTCC737), *Salmonella typhi* (MTCC734), *Pseudomonas aeruginosa* (MTCC737) and *Bacillus subtilis* (MTCC2423) were procured from Institute of Microbial Technology (IMTECH), Chandigarh. The bacterial strains were maintained in nutrient agar slants in the Department of Microbiology, RVS Hospital, Sulur, Coimbatore, Tamilnadu.

Inoculums

The test organisms were inoculated into nutrient broth medium (5% Peptone,5%Sodium chloride,3% Yeast extract,2% Beef extract, pH 7.0) and incubated at 37^0 C for overnight. The bacterial cells were harvested by centrifuging at 5000rpm for 15 minutes. The pellet formed was washed twice with PBS (Phosphate Buffer Saline), (10Mm Sodium chloride, pH 7.4) and the cells were diluted to approximately 10^5 CFU ml^{-1} before use (13).

Determination of antibacterial activity

Using diluted bacterial cells lawn plates were prepared for each microorganism to perform antibacterial assay (14). A 3mm discs (Himedia) were impregnated in the leaf extracts of each plant. The discs were then placed on the lawn plates and the plates were incubated at 37^0 C for 18h. The antibacterial activity was assessed by measuring the diameter of the zone of inhibition (mm). The relative antibacterial potency of the given preparation was calculated by comparing its zone of inhibition with that of the standard drug chloramphenicol.

1.Botanical Name : *Asteracantha longifolia* Ness.
Family : Acanthaceae
Parts used : Leaves
Medicinal properties : leaves are used for rheumatism, gonorrhoea, genitourinary tract, hepatic obstruction with dropsy, jaundice.

2.Botanical Name : *Berberis tinctoria*. Lesch
Family : *Berberidaceae*
Parts used : Root
Medicinal properties : The root infusion of this plant is used to cure jaundice and stomachache.

3.Botanical Name : *Solanum trilobatum*(L.)
Family : Solanaceae
Parts used : Leaves
Medicinal properties : Respiratory disorders, antioxidant activity, hepatoprotective activity

4. Botanical Name : *Delonex elata* (L.) Gamble
Family : Fabaceae - Caesalpinioideae
Parts used : Leaves
Medicinal properties : The leaves are anti inflammatory, rheumatic problems like pain and stiffness of the joints, especially the knees, dysmenorrhoea

5.Botanical Name : *Spathodea campanulata* Beauv.
Family : Bignoniaceae
Parts used : Leaves
Medicinal properties : The leaves are antimalarial, antimicrobials, antidiabetic

6.Botanical Name : *Sphaeranthus indicus* L.
Family : *Asteraceae*
Parts used : Whole plant
Medicinal properties : The leaves are used to treat tuberculosis, indigestion, bronchitis, spleen diseases, elephantiasis, anaemia

7.Botanical Name :*Gymnema sylvestre* R.Br.
Family : Asclepiadaceae
Parts used : Leaves
Medicinal properties : The herb is useful for stimulating the heart; it increases urine Secretion and good for the treatment of Diabetes 2 type. The leaves are useful for the ayurvedic herbal medicine.

8.Botanical Name : *Passiflora edulis* Sims
Family : *Passifloraceae*
Parts used : Leaves
Medicinal properties : The leaves are Antiasthamatic

9. Botanical Name : *Vitex trifolia* (L.)
Family : *Lamiaceae*
Parts used : Leaves
Medicinal properties : The leaves are used to treat female ailments in the Cook Islands, and used to relieve fever in Samoa. Additionally in Samoa, the dried leaves are burned to deter mosquitos.

10.Botanical Name : *Sesbania aegyptiaca* (Poir.) Pers
Family : Fabaceae
Parts used : Leaves
Medicinal properties : The leaves are used to treat Cough , Gynecology, Hemoptysis, Vermifuge

11.Botanical Name : *Euphorbia tirucalli*.L
Family : Euphorbiaceae
Parts used : Whole plant
Medicinal properties : It has been used to treat cancers, excrescences, tumors, and warts in such diverse places as Brazil, India, Indonesia, Malabar and Malaysia. It has also been used as an application for asthma, cough, earache, neuralgia, rheumatism, toothache in India.

12.Botanical Name : *Eupatorium glandulosum* (Michx.)
Family :Asteraceae
Parts used : Leaves
Medicinal properties : The leaves are used to treat cuts and wounds.

Table.1.Antibacterial activities of 12 medicinal plant extracts and Streptomycin

Plant Name	**Extracts** (100mg/ml)	**Zone of Inhibition (mm)**				
		a	**b**	**C**	**d**	**e**
Asteracantha longifolia	Met.	9	10	10	9	10
	Aq.	7	7	8	8	9
Sphaeranthus indicus	Met.	8	8	8	10	-
	Aq.	7	-	7	7	-
Passiflora edulis	Met.	8	10	9	8	9
	Aq.	-	-	7	8	7
Delonex elata	Met.	8	12	8	11	7
	Aq.	-	-	-	-	-
Euphorbia tirucalli	Met.	12	14	12	10	12
	Aq.	-	-	-	-	-
Solanum trilobatum	Met.	15	14	18	16	12
	Aq.	12	10	11	10	14
Gymnema sylvestre	Met.	10	17	7	8	-
	Aq.	7	9	-	8	-
Spathodea campanulata	Met.	10	15	8	13	10
	Aq.	7	-	-	-	-
Vitex trifolia	Met.	12	14	11	14	12
	Aq.	9	10	10	8	8
Euphatorium glandulsom	Met.	10	12	14	12	10
	Aq.	9	7	-	8	-
Sesbania aegyptica	Met.	12	10	10	12	10
	Aq.	-	9	7	8	9
Berberis tinctoria	Met.	12	10	11	14	12
	Aq.	9	7	10	8	8
Streptomycin		21	16	20	22	17

Met. – Methanol, Aq. – Aqueous

a. *Escherichia coli* (MTCC734)

b. *Salmonella typhi* (MTCC734)

c. *Staphylococcus aureus* (MTCC737)

d. *Bacillus subtilis* (MTCC2423)

e. *Pseudomonas aeruginosa*(MTCC737)

Results and Discussion

Medicinal plants have been used for centuries to cure human diseases. It has been proved that many of these plants exhibit general antibacterial and antifungal activities. Extracts from 12 medicinal plants from Western Ghats were tested against 5 clinically important standard strains to determine their antibacterial activity. All the bacterial strains tested in the growth inhibition assay showed various degrees of sensitivity to the 12 plant extracts obtained (Table.1).The methanolic extract was found to be the most effective antimicrobial agent as compared to the aqueous extract. The methanolic extract of *Solanum trilobatum* (L.) was active (85 %) against microorganisms investigated while its aqueous extract showed only moderate activity. The methanolic as well as the aqueous extract of *Vitex trifolia* showed 60 % antibacterial activity respectively. Amongst all the plant species, *Solanum trilobatum* exhibited comparable antibacterial activity against tested strains.

The traditional healers or practitioners make use of water primarily as a solvent, but our studies showed that methanol extracts of these plants were certainly much better and powerful. This may be due to the better solubility of the active components in organic solvent (16). These observations can be rationalized in terms of the polarity of the compounds being extracted by each solvent and, in addition to their intrinsic bioactivity, by their ability to dissolve or diffuse in the different media used in the assay. The growth media also seem to play an important role in the determination of the antibacterial activity. The previous reports showed that Muller-Hinton agar appears to be the best medium to explicate the antibacterial activity and the same was used in the present study (17). Amongst the gram-positive and gram-negative bacteria, gram-positive bacterial strains were more susceptible to the extracts when compared to gram negative bacteria. This is in agreement with previous reports that plant extracts are more active against gram-positive bacteria than gram-negative bacteria (3).

From our investigation of screening different plant species, the results obtained confirm the therapeutic potency of some plants used in traditional medicine. The most active extracts can be subjected to isolation of the therapeutic antimicrobials and undergo further pharmacological evaluation.

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