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Original Article

Antibacterial Potential of some medicinal plants

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Abstract

Five medicinal plant species, *Abrus precatorius* (Fabaceae), *Amaranthus spinosus* (Amaranthaceae), *Argyeria nervosa* (Convolvulaceae), *Vernonia cinerea* (Asteraceae), *Zizyphus nummularia* (Rhamnaceae) were screened for potential of antibacterial activity against four medically important human pathogen namely *Staphylococcus aureus, Streptococcus pyogene, Bacillus subtilis, Pseudomonas aeruginosa*. The antibacterial activity of aqueous and ethanol extracts were determined by agar disc diffusion method. The ethanol extracts was more active than the aqueous extract for all five plants studied. The most susceptible bacteria were *Staphylococcus aureus,* followed by *Bacillus subtilis*.
Key words: Medicinal plants, antibacterial activity, agar disc diffusion method.

Introduction

The uses of plants for curing various human aliments are figured in ancient manuscripts such as the Rigveda and the Samhitas etc. In early ages, man use raw drugs, isolated or obtained from the plants leading to information about the inter-relationship between primitive man and plants. Aboriginal people have used plants traditionally as medicine since long back. Herbal medicine is still the mainstay of about 75-80% of the population, mainly in developing countries for primary health care because of better cultural acceptability, better compatibility with the human body and fewer side effects. However the last few years have seen a major increase in their use in the developed countries. A major part of the population in developing countries still uses traditional folk medicine obtained from plant resources (Farnsworth, 1994; Srivastava, 1996).

India is a varietal emporium of medicinal plants and it is one of the richest countries in the world with regard's to genetic resources of medicinal plants. It exhibits a wide range of topography and climate, which has a bearing on its vegetation and floristic composition (Martins *et al.,* 2001). It is one of the 12 mega biodiversity centers having 45,000 plant species; its diversity is unmatched due to the 16 different agro climatic zones, 10 vegetative zones and 15 biotic provinces. The country has a rich floral diversity (Perumal Samy and Gopalakrishnakone, 2007).The medicinal action of plants are unique for particular plant species or groups, consistent with the concept that the combination of secondary products in a particular plant is taxonomically distinct (Wink *et al.,*1999).

Rai (1988) worked on *Abrus precatorius*,used against skin and Nisha and Sivadasan (2007) described that the leaf of *Vernonia cinerea* is used in the treatment of skin disease like ringworm and eczema.

In the present work five different medicinal plants were evaluated for their antibacterial properties.

Material and Methods

Ethno botanical Survey

Ethno botanical survey was carried out in some tribal inhabited localities of Gwalior district. The information on medicinal uses of plants was collected. Emphasis was laid on uses of plant species for the treatment of skin infections like, cuts, wound, ringworm, eczema etc.

Collection of Plant materials

Selected plants were identified with the help of flora and other authentic literature available in the department. Identification was finally confirmed at National Botanical Research Institute, Lucknow (U.P.).

Fresh plant part (leaves) were collected from field. Leaves were washed under running tap water, air dried and then homogenized to fine powder and stored an airtight bottles.

Extract preparation

For aqueous extraction, 10 gm of leaves of selected species were placed in distilled water and boiled for 6 hours. Later it was filtered

through 8 layers of muslin cloth and centrifuged at 5000rpm for 15min. The supernatant was collected (brown colour). After 6 hours, the supernatant was concentrated to make the final volume one-fourth of the original volume. Finally 10 gm of material was extracted in 25 ml of distilled water giving a concentration of 40 mg/0.1 ml. It was then autoclaved at 121° C and 15 lbs pressure and stored at 4°C (Nair *et al.*, 2005).

For alcoholic extraction 10 gm of air dried powder was placed in 100 ml of organic solvent (ethanol) in a conical flask, plugged with cotton and then kept on a rotary shaker at 190-220rpm for 24 hours. Later it was filtered through 8 layers of muslin cloth and centrifuged at 5000 rpm for 15min. The supernatant (dark green colour) was collected and the solvent was evaporated to make the final volume one-fourth of the original volume, giving a concentration of 40 mg/0.1 ml. It was stored at 4° C in air-tight bottles for further studies (Nair *et al.*, 2005).

Selection of microorganisms

The microbial strains are identified strains and were obtained from Cancer Hospital and Research Institute, Gwalior. The bacterial strains studied are *Staphylococcus aureus* (MTTC 740), *Streptococcus pyogenes* (MTTC 442), *Bacillus subtilis* (MTTC 121), and *Pseudomonas aeruginosa* (MTTC 741).

Antibacterial assay

The antibacterial activity of different plant species was evaluated by Agar disc diffusion method-for aqueous and solvent extract using Mueller Hinton Agar medium for the assay. The micro-organism was activated by inoculating a loopful of the strain in the nutrient broth (25ml) and incubated at room temperature on a rotary shaker. Then 0.2ml of inoculum (inoculum size was 10^8 cell/ml as per Mac Farland standard) was inoculated into the media.

For the agar disc diffusion, the leaf extract (3.2ppm) was introduced on to the disk (5mm) and then allowed to dry. Thereafter, the disc was impregnated on the seeded agar plate and all the plates were incubated at 37°C for 24 hours in triplicates. The experiment was performed under strict aseptic conditions (Bauer *et al.*, 1966). Microbial growth was determined by measuring the diameter of the zone of inhibition (mm).

Result and Discussion

The plant material is largely dependent on the type of solvent used in the extraction procedure. Traditional healers use primarily water as the solvent but in our studies we found that plant extracts in organic solvent (ethanol) provided more consistent antimicrobial activity compared to those extracted in water. The work which was carried out in triplicate but the results are the mean of triplicate readings. The results of screening are presented in table-1.

Table-1: Antibacterial activity of aqueous and ethanol extract of screened medicinal plants

Plant species	Extract Type	Zone of inhibition (including disc 5mm)			
		S.aureus	*S.pyogen*	*B. subtilis*	*P.aureginosa*
A.brus precatorius	Aqueous	10mm	-	8mm	-
	Alcoholic	11mm	6mm	6mm	6mm
Amaranthus spinosus	Aqueous	-	-	-	-
	Alcoholic	6mm	7mm	8mm	7mm
Argyeria nervosa	Aqueous	-	-	10mm	9mm
	Alcoholic	13mm	6mm	9mm	7mm
Vernonia cinerea	Aqueous	8mm	6mm	10mm	-
	Alcoholic	15mm	7mm	10mm	9mm
Zizyphus nummularia	Aqueous	7mm	6mm	-	6mm
	Alcoholic	10mm	7mm	8mm	8mm

The aqueous and ethanol extracts of five plants were tested against gram positive and gram negative bacteria using disk diffusion method (Vincent and Vincent, 1944). *Abrus precatorius* was active against *S.aureus* in both extracts, while against *B. subtilis* aqueous extract was effective. Ross (2003) and Saxena (1986) have also described the antimicrobial effect of ethanolic extract of *Abrus precatorius.* The aqueous extract of *Amaranthus spinosus* was totally inactive while as alcoholic extract was

effective against *B.subtilis*. This is in conformity to Mahato and Chaudary, (2005). The alcoholic extract of *Argyeria nervosa* was found effective against *S. aureus* which is in similarity with the earlier observation of Chansakaow *et al.,* (2003) and Parekh *et al*., (2007). *Vernonia cineria*, inhibit the bacterial growth in both alcoholic and aqueous extracts against *S. aureus* and *B.subtilis*. Similar observations have been recorded by Valsaraj *et al.,* (1996) in leaf and stem of *Vernonia cinerea* against *S. aureus, B. subtilis, E.coli* and *P. aeruginosa*. *Zizyphus nummularia* shown inhibition against *S.aureus* and almost same in *B.subtilis* and *P.aureginosa*. Adamo *et al.,*(2000) have observed the antibacterial activity of *Zizyphus mucronata* leaves against *P. aeruginosa* and *S. aureus.*

Out of the five plant species, *Vernonia cineria* showed significant antibacterial activity and both the extracts (aqueous and ethanol) were active against the investigated bacterial strains. The plant extracts were more active against the gram positive micro organisms. This is in agreement with previous report that plant extract are more active against gram positive bacteria than gram negative bacteria Valsaraj *et al.* (1996). *S. aureus* was the most susceptible bacteria amongst all bacterial strains investigated in the present work.

The results of the present study support the folkloric usage of the studied plants and suggest with antibacterial agents in new drugs for the therapy of infectious diseases caused by pathogens.

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