



Search for Antimicrobial Efficacy of Certain Indian Medicinal Plants

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Received on: 24/02/2012

Accepted on: 29/02/2012

ABSTRACT

*Pet ether, benzene, chloroform, ethyl acetate, methanol and distilled water extracts of two Indian medicinal plants *Alpinia galanga* and *Embelia ribes* were examined for their antimicrobial potential against selected bacteria and fungi. The purpose of screening is to justify and authenticate the use of Indian medicinal plants in ethnomedicinal or folklore as traditional treasure to cure various ailments. In present investigations attempts were made to screen the Indian medicinal plants as antimicrobial agent. The extracts were tested against selected test bacteria and fungi through disc diffusion assay where Tetracycline and Mycostatin were used as standard. Indian medicinal plants have a traditional background that they have potentials to use as antimicrobial agents. The results showed that all the extracts possess good antimicrobial activity against selected test bacteria and fungi. The present results therefore offer a scientific basis for traditional use of the various extracts of *Alpinia galanga* and *Embelia ribes*.*

Key Words: *Alpinia galanga*, *Embelia ribes*, Antimicrobial, Indian Medicinal Plants, Disc diffusion assay

INTRODUCTION

The use of medicinal plants as a source for aid from illness can be traced back over five millennia to written documents of the early culture in China, India and the Near east, but it is, without a doubt, an art as old as mankind. The prospective of higher plants as basis of new drugs is still largely uncharted¹. Among the anticipated 250,000-500,000 plant species, only a small percentage has been investigated phytochemically and the fraction submitted to biological or pharmacological screening is even smaller. Historically pharmacological screening of compounds of natural or synthetic origin has been the source of inestimable therapeutic agents. Casual screening as tool in discovering new biologically active molecules has been most productive in the area of antibiotics². Medicinal plants signify a rich source of antimicrobial agents³.

The microorganisms have developed resistance to many antibiotics because of arbitrary use of antimicrobial drugs that create a big dilemma in the treatment of infectious diseases⁴. With the augment in the resistance of many microorganisms to the presently used antimicrobials and the high cost of production of synthetic compounds; in addition to many side effects; there is a need to look for the alternatives. Plants have provided a good source of antiinfective agents; emetine, quinine, berberine, tannins, terpenoids, alkaloids and flavonoids continue to be highly efficient instruments in the fight against microbial infections⁵.

Therefore, in present study an attempts have been made to evaluate antimicrobial potential of two medicinal plants *Alpinia galanga* and *Embelia ribes* [Fig. 1(a) and (b)] each belonging to different families.

MATERIALS AND METHODS

Collection

Plant samples (*Alpinia galanga* and *Embelia ribes*) were collected from various tribes living in tribal pockets of Mt. Abu, arid zone of Rajasthan. These plants were used by these tribes in their daily lives to cure various ailments and few from Chunnilal Attar Ayurvedic Store, Ghat Gate, Jaipur in the month of May, 2011.

Identification

All the samples were authenticated and were given identification number *Alpinia galanga* and *Embelia ribes*. These samples were authenticated and submitted in Ethnomedicinal Herbarium, Centre of Excellence funded by DST, MGias, Jaipur (Rajasthan).

Sources of Test Organisms

Pure culture of all test organisms, bacteria namely *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Escherichia coli*, *Proteus vulgaris*, *Klebsiella pneumoniae*, *Shigella sonnei* and *Trichophyton rubrum* and fungi *Candida albicans*, *Aspergillus niger* and *Aspergillus flavus* were obtained through the courtesy of Mahatma Gandhi Institute of applied Sciences(MGias), Jaipur, which were maintained on Nutrient broth media.

Culture of Test Microbes

For the cultivation of bacteria, Nutrient Agar Medium (NAM) was prepared by using 20 g Agar, 5 g Peptone, 3 g beef extract and 3 g NaCl in 1 L distilled water and sterilized at 15 lbs pressure and 121°C temperature for 25-30 min. Agar test plates were prepared by pouring approximately 15 mL of NAM into the Petri dishes (10 mm) under aseptic conditions. A saline solution was prepared (by mixing 0.8% NaCl) in distilled water, followed by autoclaving and the bacterial cultures were maintained on this medium by regular sub-culturing and incubation at 37°C for 24-48 h. To prepare the test plates, in bacteria, 10-15 mL of the respective medium was poured into the Petri plates and used for screening. For assessing the bactericidal efficacy, a fresh suspension of the test bacteria was prepared in saline solution from a freshly grown Agar slant.

Preparation of Test Extracts

Crushed powder (50 g) of all the species were successively soxhlet extracted with ethanol. Later, each of the homogenates was filtered and the residue was re-extracted twice for complete exhaustion, the extracts were pooled individually. Each filtrate was concentrated to dryness in vitro and redissolved in respective solvents, out of which 80 mg/10 disc i.e. 8 mg/disc concentration were stored at 4°C in a refrigerator, until screened for antibacterial activity.

Antimicrobial Assay by Disc Diffusion Method

For both, bactericidal in vitro Disc diffusion method was adopted (Gould and Bowie, 1952), because of reproducibility and precision. The different test organisms were proceeded separately using a sterile swab over previously sterilized culture medium plates and the zone of inhibition were measured around sterilized dried discs of Whatman No. 1 paper (5 mm in diameter), which were containing 1mg, 5mg and 10mg of the test extracts and reference drugs (tetracycline and mycostatin for bacteria and fungi, respectively) separately. Such treated discs were air-dried at room temperature to remove any residual solvent, which might interfere with the determination, sterilized and inoculated. These plates were initially placed at low temperature for 1 h so as to allow the maximum diffusion of the compounds from the test disc into the agar plate and later, incubated at 37°C for 24 h in case of bacteria and °C in case of fungi, after which the zones of inhibition could be easily observed. Five replicates of each test extract were examined and the mean values were then referred. The Inhibition Zone (IZ) in each case were recorded and the Activity Index (AI) was calculated as compared with those of their respective standard reference drugs (AI = Inhibition Zone of test sample/Inhibition zone of standard).

RESULTS AND DISCUSSION

The profile of the medicinal plants used in the present investigation. The results of antimicrobial activity of the crude extracts of Selected Indian Medicinal Plants (*Alpinia galanga* and *Embelia ribes*) showed good antimicrobial activity against selected test bacteria and fungi (Table-1 and 2). Overall, these extracts showed appreciable activity against selected test bacteria and fungi and hence, it justify their use in our traditional system of medicine to cure various diseases (Fig.2 and 3).

Alpinia galanga

While screening the extracts of *Alpinia galanga* good antimicrobial activity against the selected bacteria and fungi was observed. The various extracts were found active against all the bacteria and fungi tested. Results, comparable to the standards, were found against *P.vulgaris*, *E.coli*, *K.pneumoniae*, *S.sonnei* (pet ether extract), *E.coli* (benzene extract), *P.vulgaris*, *E.coli* (chloroform extract), *A.niger* (ethyl acetate extract), *P.aeruginosa*, *E.coli*, *S.sonnei*, *C.albicans*, *A.niger* (methanol extract), *P.aeruginosa*, *E.coli* and *C.albicans* (aqueous extract). Antimicrobial activity greater than the activity of the standard was observed against *P.aeruginosa*, *T.rubrum* (pet ether extract), *P.vulgaris*, *P.aeruginosa*, *T.rubrum*, (benzene extract), *P.aeruginosa*, *C.albicans*, *A.niger*

(chloroform extract), *P.vulgaris*, *P.aeruginosa*, *S.sonnei*, *C.albicans* (ethyl acetate), *S.sonnei* and *A.niger* (aqueous extract).

Embelia ribes

While screening the extracts of *E. ribes* the, good antimicrobial activity against the selected bacteria and fungi was observed. The various extracts were found active against all the bacteria and fungi tested. Results, comparable to the standard, were obtained against *P.vulgaris*, *E.coli*, *K.pneumoniae*, *S.sonnei* (pet ether extract), *E.coli* (benzene extract), *P.vulgaris*, *E.coli* (chloroform extract), *A.niger* (ethyl acetate extract), *P.aeruginosa*, *E.coli*, *S.sonnei*, *C.albicans*, *A.niger* (methanol extract), *P.aeruginosa*, *E.coli* and *C.albicans* (aqueous extract). Antimicrobial activity greater than the activity of the standard was observed against *P.aeruginosa*, *T.rubrum* (pet ether), *P.vulgaris*, *P.aeruginosa* (benzene extract), *P.aeruginosa*, *C.albicans*, *A.niger* (chloroform extract), *P.vulgaris*, *P.aeruginosa*, *S.sonnei*, *C.albicans* (ethyl acetate extract), *S.sonnei* and *A.niger* (aqueous extract of the plant).

CONCLUSION

The versatile medicinal plants are the unique source of various types of compounds having diverse chemical structures. Very little work has been done on the biological activity and plausible medicinal applications of these compounds and hence extensive investigation is needed to exploit their therapeutic utility to combat diseases.

The present results therefore offer a scientific basis for traditional use of the various extracts of *Alpinia galanga* and *Embelia ribes*. These results explain that Indian Medicinal Plants have potentials as antimicrobials. Further, more or less both the selected Indian Medicinal Plants have also possessed antimicrobial potential against all test bacteria and fungi which explains that their use in daily life will generate a resistance or immunity to fight against microorganisms.

ACKNOWLEDGEMENT

Author acknowledge with thanks the financial support from Department of Science and Technology, Government of Rajasthan, in the form of Centre with Potentials for Excellence in Biotechnology, sanction no

Table 1: Antibacterial efficacy in terms of inhibition zone of *Alpinia galanga* against selected bacteria and fungi

<i>Alpinia galanga</i> Extracts	Measure	Bacteria							Fungi		
		<i>P. vulgaris</i>	<i>P. aeruginosa</i>	<i>T. rubrum</i>	<i>S. aureus</i>	<i>E. coli</i>	<i>K. pneumoniae</i>	<i>S. sonnei</i>	<i>C. albicans</i>	<i>A. niger</i>	<i>A. flavus</i>
	Standard I.Z.	9.1	9.9	10.9	17.9	9.8	12.3	11.9	11.4	9.7	11.8
Pet ether	I.Z. (mm)	7.6	10	12	7.6	9.6	12	11.5	9	6.5	6.5
	A.I.	0.835	1.010	1.100	0.424	0.979	0.975	0.966	0.789	0.670	0.550
Benzene	I.Z. (mm)	11	11	4	7	9.3	8.6	9	8.6	5.6	7.6
	A.I.	1.208	1.111	0.366	0.391	0.948	0.699	0.765	0.754	0.577	0.644
Chloroform	I.Z. (mm)	7.6	10.3	8	6.5	9.6	8	8.3	21.5	11.6	8
	A.I.	0.835	1.040	0.733	0.363	0.979	0.650	0.697	1.88	1.195	0.677
Ethyl Acetate	I.Z. (mm)	10.3	10.6	8	7.5	7.6	8.5	16.5	18	8.3	8
	A.I.	1.131	1.070	0.733	0.418	0.775	0.691	1.386	1.578	0.855	0.677
Methanol	I.Z. (mm)	7	8.6	7	7	9	8.6	11	10.3	9.3	9
	A.I.	0.769	0.868	0.642	0.391	0.918	0.699	0.924	0.903	0.958	0.762
Distilled Water	I.Z. (mm)	6.3	8	6.5	8.6	9.3	8.5	12.5	9.5	19	9
	A.I.	0.692	0.808	0.596	0.480	0.948	0.691	1.050	0.833	1.95	0.762

I.Z. = Inhibition zone, A.I. = Activity index *I.Z. in mm are the mean value of the triplicates

Table 2: Antibacterial efficacy in terms of inhibition zone of *Embelia ribes* against selected bacteria and fungi

<i>Embelia ribes</i> Extracts	Measure	Bacteria							Fungi		
		<i>P. vulgaris</i>	<i>P. aeruginosa</i>	<i>T. rubrum</i>	<i>S. aureus</i>	<i>E. coli</i>	<i>K. pneumoniae</i>	<i>S. sonnei</i>	<i>C. albicans</i>	<i>A. niger</i>	<i>A. flavus</i>
	Standard I.Z.	9.1	9.9	10.9	17.9	9.8	12.3	11.9	11.4	9.7	11.8
Pet ether	I.Z. (mm)	9.3	8.3	8	9	8.6	9	8.3	9	8	19.6
	A.I.	1.021	0.838	0.733	0.502	0.877	0.731	0.697	0.789	0.824	1.661
Benzene	I.Z. (mm)	8	11	6.3	9.6	9.6	9	9.6	8	10.3	10.6
	A.I.	0.879	1.111	0.577	0.536	0.979	0.731	0.806	0.701	1.061	0.898
Chloroform	I.Z. (mm)	9.3	9.3	7	8.3	9.6	9.6	8.6	9	14	7.5
	A.I.	1.021	0.939	0.642	0.463	0.979	0.780	0.722	0.789	1.443	0.635
Ethyl Acetate	I.Z. (mm)	9	11	10	8.6	7.6	8.5	9	8	7.5	11
	A.I.	0.989	1.111	0.917	0.480	0.775	0.691	0.756	0.701	0.773	0.932
Methanol	I.Z. (mm)	8.5	8.6	7	7	9	8.6	11	10.3	9.3	9
	A.I.	0.934	0.868	0.642	0.391	0.918	0.699	0.924	0.903	0.958	0.762
Distilled Water	I.Z. (mm)	8.3	13.3	7.5	8.6	10.6	9.5	8.5	19.3	11.3	12.6
	A.I.	0.912	1.343	0.688	0.480	1.081	0.772	0.714	1.692	1.164	1.067

I.Z. = Inhibition zone, A.I. = Activity index *I.Z. in mm are the mean value of the triplicates

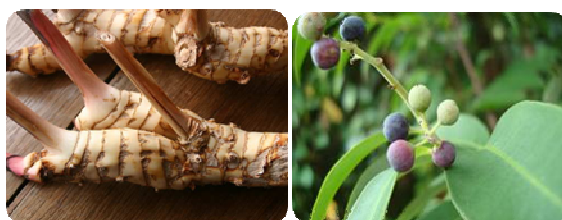


Fig.1: (a) *Alpinia galanga* and (b) *Embelia ribes*

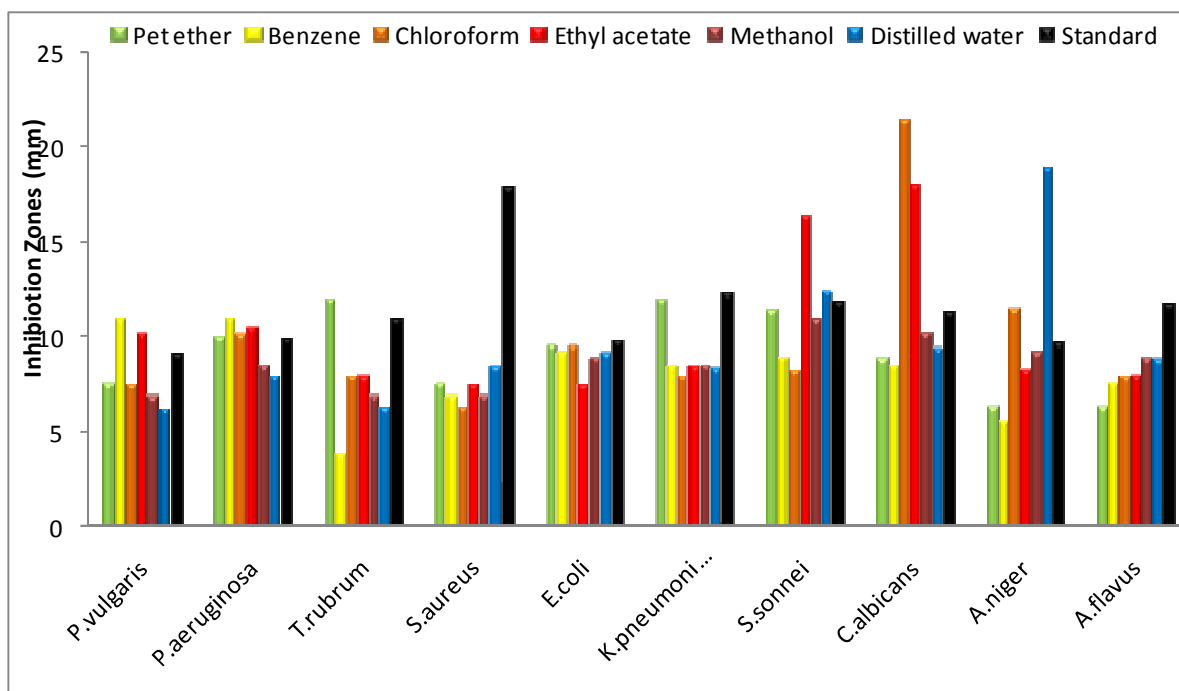


Figure 2: Antimicrobial potential of *Alpinia galanga* against selected test microorganisms in terms of inhibition zone

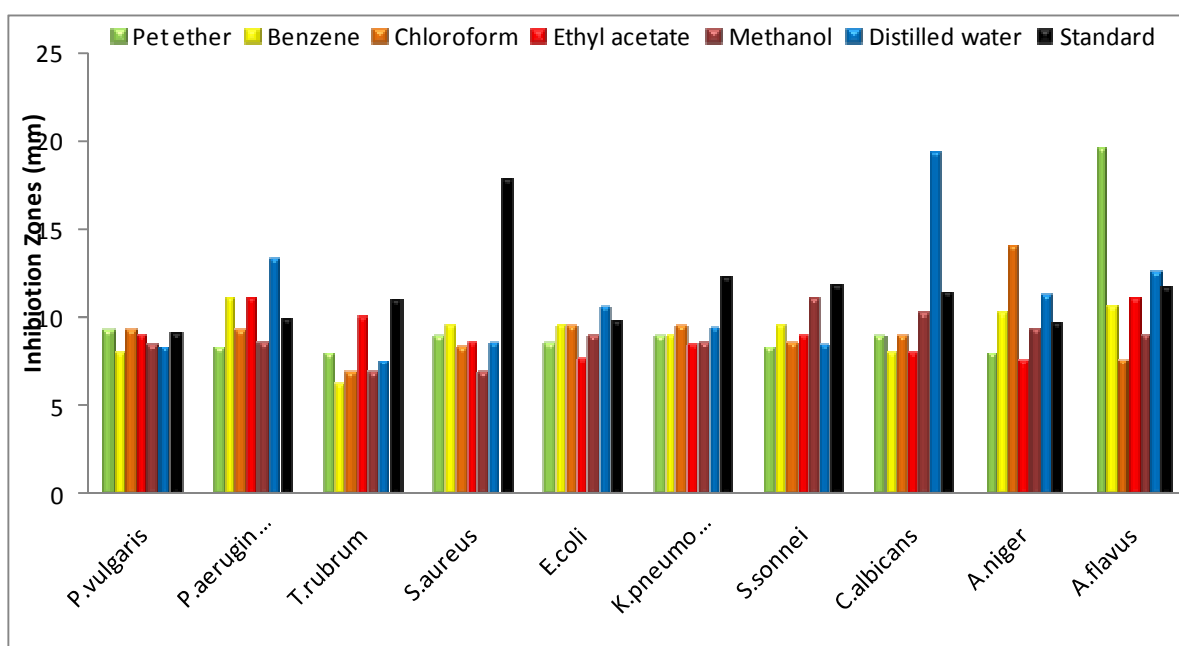


Figure- 3: Antimicrobial potential of *Embelia ribes* against selected test microorganisms in terms of inhibition zone

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