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ANTIBACTERIAL ACTIVITIES OF SELECTED INDIAN MEDICINAL PLANTS

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ABSTRACT

Plants are a valuable source of natural products for maintaining health, since time immemorial. The past few decades has witnessed an overwhelming increase in the development of modern drugs from plant products. India is bestowed with medicinal plants which are widely used for folk therapeutics and in Siddha, Unani and Ayurveda systems of medicine. The present study is an attempt to evaluate the antibacterial activities of *Alpinia galanga, Ayapana triplinervis, Aegle marmelos* and *Euphorbia hirta* against a panel of bacteria. *K. pneumonia, E. coli* and *Salmonella* sp. are sensitive to all the extracts tested. *P. aeruginosa* was resistant to *A. marmelos* and *E. hirta* extracts while *Citrobacter* sp. was resistant towards *E. hirta* methanolic extract only. *S. aureus* also showed resistance against *A. triplinervis*. The study demoed that the methanolic and petroleum ether extracts of the selected plants comprise antibacterial principles as usurped by indigenous herbal healers.

KEYWORDS

INTRODUCTION

Multi-drug resistance among microbial strains and the appearance of strains with reduced susceptibility to current antibiotics are steadily increasing which is attributed to indiscriminate use of broad-spectrum antibiotics along with increasing population of immunocompromised individuals (Senka *et al.,* 2008; Levyand Marshall, 2004). Currently, the chemical and pharmacological constituents of medicinal plants are widely utilized in different traditional systems of treatment globally and are growingly inquired for human well being (Patwardhan, 2005). The Indian system of medicine has medicinal plants as its cradle which is time tested in varied geographic and climatic zones across the country. The quest for more and more drugs from botanical origin is incessantly increasing which demands sieving of medicinal plants with sufficient biological activities. The present study is an attempt to evaluate the antibacterial activities of selected medicinal plants which are traditionally used by traditional healers like *Alpinia galanga, Ayapana triplinervis, Aegle marmelos* and *Euphorbia hirta* against a panel of bacteria such as *Escherichia coli, Citrobacter* sp., *Psueudomonas aeruginosa*, *Klebsiella pneumoniae*, *Salmonella* sp. and *Staphylococcus aureus*.

MATERIALS AND METHODS

Plants selected

***Alpinia galanga* (Zingiberaceae)**

The plant is consistently used by South Indian physicians of traditional Ayurveda and Siddha medicine system to treat various ailments. The plant has uses like cardiotonic, diuretic, hypotonic, gastric lesions, antiplatelet, anti-tumor, anti-fungal, carminative, irritant action, whooping cough in children, bronchitis, anti-asthma, dyspepsia, fever and diabetes mellitus (Verma *et al*., 2011).

***Ayapana triplinervis* (Asteraceae)**

The plant is widely used in folk medicine due its analgesic, anticoagulant, antianorexic, antiparasitic, anthelmintic, sedative and antimicrobial properties. Leaves are used through infusions, decoctions, baths, and tea. It is largely used in Brazilian folk medicine as sedative, febrifuge, stimulant, tonic and anti-inflammatory (Bose *et al*., 2007).

***Aegle marmelos* (Rutaceae)**

The plant is widely used in indigenous systems in Indian medicine due to its multifarious activities. The plant is considered scared in Hindu tradition and offered in prayers to Lord Shiva and Parvathi and hence the name Shivduma (tree of Shiva). Different parts of the plant are used for various therapeutic uses like asthma, anaemia, fractures, healing of wounds, swollen joints, high blood pressure, jaundice and diarrhea. It is used as a herbal medicine for the management of diabetes mellitus in Ayurvedic, Unani and Siddha systems of medicine (Sharma *et al*., 2011)

***Euphorbia hirta* (Euphorbiaceae)**

The plant is native to India but is a pan tropical weed, found especially on roadsides and wasteland. The plant is used for female disorders, respiratory ailments, especially cough, coryza, bronchitis and asthma. The plant is commonly used to treat worm infestations in children and for dysentery, jaundice, pimples, digestive problems tumours and ethnoveternary usages (Asha *et al*., 2014).

Extract preparation

Leaves of the plants (*Ayapana triplinervis, Aegle marmelos* and *Euphorbia hirta*) and rhizome (*Alpinia galanga*) were collected from different regions in Kottyam District and taxonomically identified by using standard taxonomic keys and expert consultations. The collected plants were dried under shade, crushed and subject to soxhlet extraction with petroleum ether and methanol. The extract was filtered and concentrated.

Antibaterial susceptibility testing

Pure cultures of *Escherichia coli, Citrobacter* sp., *Psueudomonas aeruginosa*, *Klebsiella pneumoniae*, *Salmonella* sp. and *Staphylococcus aureus* were used for the study. The strains were maintained on nutrient agar slants at 4°C. A loopful of each bacterial strain was added to a 50 ml sterile nutrient broth in a 100 ml conical flask. The flasks were then incubated for 24 h to activate the test strain.

Agar diffusion method

The antibacterial activity of the selected plant extracts was obtained with two different solvents - petroleum ether and methanol which was assessed by the agar diffusion method (Balouiri et al., 2016). The agar plate surface is inoculated by spreading of the microbial inoculum uniformly over the entire agar surface. Then, a hole with a diameter of 6 to 8 mm is punched aseptically with a sterile cork borer and a volume (50 µL) of the extract was introduced into the well. Then, agar plates are incubated overnight. The antimicrobial agent diffuses in the agar medium and inhibits the growth of the microbial strain tested. The antibacterial activity was determined by measuring the zone of inhibition and expressed as millimeter (mm). Five sets of plates are used for the antimicrobial studies. Control plates were also maintained for the study.

RESULTS

The antibacterial activities of *Alpinia galanga, Ayapana triplinervis, Aegle marmelos* and *Euphorbia hirta* against a bacterial pathogens such as *Escherichia coli, Citrobacter* sp., *Psueudomonas aeruginosa*, *Klebsiella pneumoniae*, *Salmonella* sp. and *Staphylococcus aureus* was conducted. The obtained results are given Table 1. In an overall analysis *K. pneumonia, E. coli* and *Salmonella* sp. are sensitive to all the extracts tested. *P. aeruginosa* was resistant to *A. marmelos* and *E. hirta extracts while Citrobacter sp. was resistant towards E. hirta methanolic extract only. S. aureus* also showed resistance against *A. triplinervis*. *K. pneumoniae* and *E. coli* are more sensitive towards the tested extracts. The highest zone of inhibition was exhibited by *A. triplinervis* methanol extract against *Citrobacter* sp. and *E. hirta* methanol extract against *S. aureus*.

Table 1: Sensitivity pattern of tested extracts

Bacteria tested	Plants tested							
	A.triplinervis		*A. marmelos*		*E. hirta*		*A. galanga*	
	Zone of inhibition (mm)							
	PE	M	PE	M	PE	M	PE	M
Citrobacter sp.	16±1.03	27±2.21	26±1	20	19±2.14	--	16	16
P. aeruginosa	19±3.44	24±2.17	--	--	--	--	20±1.67	17±2.01
E. coli	20±1.76	19±1.64	16	27±3.12	20±1.98	25±2.94	25±2.09	18±1.09
S. aureus	--	--	19±1	21±2.82	23±3.54	27±1.98	16±2.26	19±1.98
K. pneumoniae	24±1.67	24±3.27	20±1	18±2	18±1.84	20±2.61	26±1	24±1.99
Salmonella sp.	16±2	27±2.90	17	16±1.29	21±2.09	24±1.57	20±3.93	18

(PE-Petroleum ether; — Methanol)

DISCUSSION

In the present investigation, *in vitro* antibacterial efficacy of the soxhlet extracts of *Alpinia galanga, Ayapana triplinervis, Aegle marmelos* and *Euphorbia hirta* was evaluated on the basis of zone of inhibition. All the plants tested in the present study exhibited commendable inhibitory effect against the selected human bacterial pathogens. Finding healing powers in plants is an ancient idea and people from all geographic realms still rely on indigenous system to cure various ailments. Plants have provided a source of inspiration for novel drug compounds as plant derived medicines have made significant contribution towards human health remedies. These compounds can be the base for the development of medicine, a natural blueprint for the development of novel drugs (Phillipson, 2001; Kennedy and Wightman, 2002).

The present study reveals that there is a potential for obtaining potent antibacterial compounds from all the four tested plants, either as botanical antibacterials by themselves, or as models for the synthesis of antibacterial analogues. The biologically active plants showing the highest activity need to be validated more precisely and formulated with other natural compounds to increase the ambit of effect. The obtained results are in tune with the available reports. Lopes *et al*. (2015) evaluated the antimicrobial effect of methanol extract of *Eupatorium triplinervis* with promising results. The effect of the plant extract on naso-pharyngeal pathogens - *Streptococccus pyogenes* and *Pseudomonas aeruginosa* was also reported (Krishnan and Nair, 2016). The plant is also rich in Coumarins which are components of the general defense response of plants to abiotic and biotic stresses and it has been proved antimicrobial effects (Murray *et al*., 1982).

The antibacterial effects of *A. galanga* against *Bacillus subtilis*, *Enterobacter aerogene, Enterobacter cloacae, Enterococcus faecalis, Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Salmonella typhimurium, Staphylococcus aureus* and *Streptococcus epidermis* using Agar well diffusion method was well reported (Rao *et al*., 2010). The GC–MS analysis of methanol extracts also yielded compounds like 5-hydroxymethyl furfural, benzyl alcohol, 1,8 cineole, methylcinnamate, 3-phenyl-2-butanone and 1,2 benzenedicarboxylic acid which could be responsible for its broad spectrum activity.

Poonkothai and Saravanan (2008) assessed the antibacterial activity of the methanol, chloroform and aqueous extracts from the leaves, bark and fruit of *A. marmelos* using disc diffusion method against *Bacillus subtilis, Staphylococcus aureus, Klebsiella pneumoniae, Proteus mirabilis, Escherichia coli, Salmonella paratyphi A* and *Salmonella paratyphi B*. The methanolic extract has significant antibacterial activity and justifies the uses of the plant in the traditional system of medicine to cure diseases. Joglekar *et al*. (2012) also attested the spectrum of antibacterial activity of *A. marmelos*.

The antimicrobial activities of the methanolic extracts of *Euphorbia hirta* leaves, flowers, stems and roots were evaluated with promising results (Rajeh *et al*., 2010). *E. hirta* is a very popular herb among the practitioners of traditional medicine around the world. Moreover, it is used as an antidote and pain relief of scorpion stings and snakebites. The use of the latex to facilitate removal of thorns from the skin is also common.

The differences in activity pattern of the tested extracts are due to the type of solvent used in the extraction procedure and in addition to their intrinsic bioactivity (Dey *et al.,* 2010). The tested plants are rich in various phytochemicals and the synergistic activity of phytochemicals may be the reason for the detected antibacterial activity. Failure of modern medicine, side effects of chemical drugs and alarmingly rising cost of treatment has resulted in an explosion of researches with medicinal plant extracts.

CONCLUSION

Phytochemicals are cheap and serving not only as functional food but also as nutraceuticals. It should be noted that herbal medicine is still the mainstay of more than 80% of the whole population, especially in developing countries. The results of the study ponder the presence of effective plant based antimicrobials with motleying inhibition patterns. Further research in this line is the need of the hour, as drug resistance and disease causalities are on the rise.

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