

ANTIBACTERIAL ACTIVITY OF PLANT EXTRACT *PLUMBAGO ZEYLANICA* AGAINST CLINICAL BACTERIA

K.S.M. Karthika

Department of Microbiology, Dr. Lankapalli Bullayya PG College and Research Centre,
Visakhapatnam, Andhra Pradesh, India
E-mail: lkarthika@yahoo.co.in

Abstract: The present study was carried out to screen the antibacterial potentials of plant extract *Plumbago zeylanica* L. Plant extract was prepared in methanol, hexane and chloroform by Soxhlet extractor and tested for its antibacterial activity against clinical pathogens i.e. *Enterococcus*, *E.coli*, *Klebsiella pneumonia*, *Lactobacillus acidophilus* and *Staphylococcus aureus* by well diffusion method. Streptomycin was used as the standard drug respectively. The maximum antibacterial activity was determined by zone of inhibition in methanol extract against the tested organisms.

Keywords: *Plumbagozeylanica*, Streptomycin, Antibacterial screening.

Introduction

Owing to the realization of the toxicity associated with the use of antibiotics and synthetic drugs, Western countries are increasingly aware of the fact that drugs from natural sources are safer. Therefore, an upsurge in the use of products based on plants is exposed, especially in the field of health care products. Plants have always been the source of food, medicine and other necessities of life since the origin of human beings. Plants containing medicinal properties have been known and used in some form or other, even by primitive people. In India, the oldest record of the use of plants as medicine is given in the Rigveda, Charaka Samhita and Susruta Samiha. The role of traditional medicines in the solution of health problems is invaluable on a global level. World Health Organisation (WHO) has estimated that at least 80% of the world's population relies on traditional systems of medicine, for their primary health needs. According to WHO over 21,000 plant species are useful in the preparation of medicines. Nowadays even the developed countries are turning to herbal remedies. Besides, modern scientific medicine also depends on plants for some essential drugs. Modern therapies are far too costly to be practical for the majority diseases caused by microorganisms.

Plumbago zeylanica L. is a multipurpose medicinal herb of family Plumbaginaceae commonly called as Doctorbush or Ceylon Leadwort. It is a semi climbing shrub that

grows widely in Australia, Asia, Africa and Ceylon and widely used in ethanomedicine. In Ayurvedic and Unani system of medicines the whole plant has been described for significant effective against anaemia, rheumatic pain, sprains, dysmenorrhoea, carbuncles, scabies, leprosy, inflammation, ulcers and elimination of intestinal parasites. Pharmacological studies have indicated that *P. zeylanica* extract has antiplasmodial (Simonsen et al., 2001), antimicrobial (Ahmad et al., 2000), antifungal (Mehmood et al., 1999), anti-inflammatory (Oyedapo, 1996), antihyperglycemic (Olagunju et al., 1999), hypolipidaemic and antiatherosclerotic activities (Sharma et al., 1991).

The present investigation is an attempt to test the antibacterial activity of *Plumbago zeylanica* L. against the selected pathogenic bacteria.

Materials and Method

Materials Used

Plant Material Used: The plant material of *Plumbago zeylanica* was collected from local market of Visakhapatnam. The identification was confirmed at Department of Botany, Andhra University.

Organisms Used: Clinical pathogens like *Enterococcus*, *Escherichia coli*, *Klebsiella pneumonia*, *Staphylococcus aureus* and *Lactobacillus acidophilus* purchased from MTCC, Chandigarh.

Solvents, Chemicals and Medias Used: Methanol, Chloroform and Hexane were purchased from Qualigens fine chemicals, Mumbai and S.D. Fine chemicals, Mumbai. Nutrient Agar, Nutrient Broth were bought from HiMedia, Mumbai, India.

Instruments Used: Soxhlet apparatus, Autoclave, Laminar Airflow and Incubator.

Methodology

Preparation & Extraction of crude plant extract

The total plant material of *Plumbago zeylanica* were washed under running tap water and blotted with filter paper then shade dried on laboratory benches by putting newspapers. After complete drying the plant material was then ground into powder by using hand mill. The dried and powdered plant material (100 g) was extracted successively with 600 ml methanol, chloroform and hexane with a Soxhlet extractor for 48h at temperature not exceeding the boiling point of the solvent. The extracts were filtered through Whatman No. 1 filter paper and then concentrated in a vacuum at 40°C using a rotary evaporator. Each extract was transferred to glass vials and kept at 4°C before use.

Preparation of Inoculum

Bacterial strains preserved in nutrient agar at 4°C were revived in nutrient broth (liquid medium) and incubated at 37±1°C overnight, and the suspensions were checked to provide-10⁵cfu/ml.

Preparation of Media

For testing of the anti-bacterial activity Nutrient Agar medium (NAM) medium was used. The medium was sterilized at 15 lbs for 20 min at 121°C.

Antibacterial Assay of the Extract by Agar Well Diffusion Method

To test the antimicrobial activity on agar plates, NAM was prepared using the ingredients mentioned above. The medium was sterilized at 121°C for 30 min's. The agar test plates were prepared by pouring about 15ml of the medium into 10cm Petri dishes under aseptic condition and left undisturbed for 2hrs to solidify the medium. 0.1ml of *Enterococcus*, *Escherichia coli*, *Klebsiella pneumonia*, *Staphylococcus aureus* and *Lactobacillus acidophilus* were spread on the distinct plates with the help of a sterile glass spreader. The wells were prepared using alcohol sterilized borer (8 mm). Then 20 µl of various concentrations 100mg/ml and 250mg/ml plant extract were added into each well using micropipette. After 24 hours of incubation at 37°C, inhibition zone diameter was calculated.

Results and Discussion

The use of different parts of several medicinal plants to cure specific ailment, has been in continuous practice since ancient times. Herbal medicine represents probably the first and certainly the oldest system of human health care. Almost all civilization and cultures have employed plants in treatment of human sickness. A plant used in this study was selected for its importance in ethanobotanic. Results obtained in the present study revealed that plant extracts possess potential antibacterial activity against the tested organism such as *Enterococcus* spp, *E.coli*, *Klebsiella pneumonia*, *Lactobacillus acidophilus* and *Staphylococcus aureus*. Among the extract assayed, methanol extract of *Plumbago zeylanica* showed more inhibitory effect than the other plant extracts. This tends to show that the active ingredients of the plant parts are better extracted with methanol than Chloroform and Hexane. *P. zeylanica* exhibited good activity against *Staphylococcus aureus* at 250mg/ml i.e, 18 mm was recorded as diameter zone of inhibition. This was followed by 15mm *Klebsiella pneumonia* and *Lactobacillus acidophilus*, 12mm *Escherichia coli* and *Enterococcus* spp 10 mm respectively. Activities of the various extracts were comparable to those of standard antibacterial drug Streptomycin.

Results of antimicrobial activity of Soxhlet extract of used plants against the test microorganism were given in the following Table 1.

Table 1. Antibacterial Activity of *Plumbagozeylanica*

S.No.	Clinical Pathogens	ZONE OF INHIBITION (mm)						Streptomycin 5µg/ml
		Methanol Extract		Chloroform Extract		Hexane Extract		
		100mg/ml	250mg/ml	100mg/ml	250mg/ml	100mg/ml	250mg/ml	
1	<i>Enterococcus sps</i>	10	10	NA	12	NA	10	13
2	<i>Escherichia coli</i>	10	12	NA	14	NA	10	10
3	<i>Klebsiella pneumonia</i>	12	15	12	15	10	16	12
4	<i>Lactobacillus acidophilus</i>	10	15	10	13	NA	10	14
5	<i>Staphylococcus aureus</i>	15	18	13	13	10	17	13

Vol per well- 20µl, Bore size used – 8mm, NA- No Activity

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