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Research Article

EVALUATION OF ANTIMICROBIAL ACTIVITIES OF *ARISTOLOCHIA INDICA* (LINN)

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ABSTRACT

The present study was designated to evaluate the antimicrobial activities of ethanolic extract of *Aristolachia indica* L. which was a creeper used as traditional folk medicine for the treatment of different infectious diseases and disorders. The antimicrobial activities of the extract against 12 strains belong to bacterial and fungi species were tested by using agar diffusion method. The results showed that ethanolic extract of *Aristolachia indica* had moderately significant antibacterial and significant antifungal activity. It inhibited the growth of both bacterial and fungal species dose dependently. The inhibition of growth was highest at 100mg/ml as compared to the controls. Ethanolic extract showed stronger antimicrobial activity against the fungi than that of the bacteria's. Thus we can conclude that *Aristolochia indica* Linn was a potent antimicrobial agent which can be tried as a Novel anti-fungal agent.

Keywords: Antimicrobial, *Aristolachia indica* and Agar diffusion assay.

INTRODUCTION

Traditional medicine is an important source of potentially useful new compounds for the development of chemotherapeutic agents. The first step towards this goal is the screening of plants used in popular medicine. Thus, antimicrobial research is geared towards the discovery and development of novel antibacterial and antifungal agents[1.] Herbal medicine represents one of the most important fields of traditional medicine in India. These herbal medicines have been utilized in developing countries not only as a way to rescue the traditional medicine but also an alternative solution to health problems. The world health organization (WHO) also reports that 80% of worlds population depend mainly on traditional medicine and the traditional treatment involve mainly the use of plant extracts[2.]

Plant based antimicrobials represent a vast untapped source for medicines and further exploration of plant antimicrobials needs to occur[3.] *Aristolochia indica*, commonly known as Ishwari, Nakuli and Gandhanakuli have enormous therapeutic potential and it was found to be effective in the treatment of intermittent fever, malaria, parasitic infestations, various skin diseases, as an aphrodisiac, an anthelmintic and it is also used in oedema, intestinal disorders[4.], fungal and bacterial infections[5-6]. The plant is widely distributed throughout the tropical and subtropical regions of India. This paper reports the antimicrobial activity of *Aristolochia indica*. This particular plant and its leaves were selected on the basis of its traditional and medicinal uses and it is found to be very effective against microbes.

MATERIAL AND METHODS

Plant Collection

The plant materials (leaves) were freshly collected in various parts of trichy in the beginning of September month and were officially authenticated by Dr. N. Ravichandran. The voucher speciemen of the same was submitted to the herbarium of CARISM, SASTRA University and Thanjavur.

Preparation and selection of plant extract

The collected leaves were shade dried, coarsely powdered and extracted by using hot continuous extraction technique in a soxhlet extractor using various solvents such as ethanol, petroleum ether, ethyl acetate, chloroform and acetone until the extracts were colourless in the siphon tube. The extracts were concentrated and dried under vacuum. Preliminary phytochemical studies revealed that the ethanolic extract was found to possess higher number of phytoconstituents than any other extracts. So, for the present study ethanolic extract was selected for the antimicrobial evaluation.

Test microorganisms

The following bacterial strains and fungi strains were used for screening the antimicrobial activity on a broad spectrum basis. All microbial human pathogens used were procured from IMT, Chandigarh. Gram negative bacteria such as *Escherichia coli* (MTCC 724), *Pseudomonas aeruginosa* (MTCC 741), *Salmonella typhi* (MTCC 733), Vibrio *vulnificus* (MTCC 1145), Proteus *Vulgaris* (MTCC 426), Gram Positive bacteria's such as Bacillus *subtilis* (MTCC 441), *Staphylococcus aureus* (MTCC 96) *Streptococcus pneumoniae* (MTCC 655), and Fungi such as Aspergillus *niger, Aspergillus flavus*, *Aspergillus fumigatus* (MTCC 1344) and *Candida albicans* (MTCC 227) were utilized for the study.

Media used

Nutrient agar (NA) and potato Dextrose agar (PDA) were used respectively for testing the antibacterial and antifungal activity and their compositions were given below.

Composition of Nutrient Agar: (gm/ litre)

Peptone	: 5.0
Beef extract	
Yeast extract	: 3.0
Sodium chloride	
Agar	: 3.0
Distilled water	
pH	: 5.0

Composition of potato Dextrose Agar (PDA) (gm/l)

Infusion from potatoes	: 200 g
Dextrose	: 20 g
Agar	: 15 g
Distilled water	: 1 litre
pH	: 5.6 ± 0.2

Inoculation

Inoculation of each bacterial and fungi strain were suspended in nutrient broth and incubated for 8 hrs at 37^{0}C.

Determination of anti-microbial activity

Agar well diffusion method[7-11] was followed to determine the anti-microbial activity. Nutrient Agar (NA) and potato dextrose agar (PDA) were inoculated with the organisms and allowed to solidify. Four wells of 10mm diameter were made in each of this plate using sterile cork borer. About 0.3 ml of different concentration of plant extract were added using sterilized dropping pipette into the wells and allowed to diffuse at room temperature for 2 hrs. The

plates were incubated at 37^{0}C for 18 – 24 hrs for bacteria pathogens and 3 days for fungal pathogens. Diameter of inhibition zones were recorded for determining the anti-microbial activity.

RESULTS AND DISCUSSION

The results of antimicrobial activity of alcoholic extract of *Aristolachia indica* Linn against various human pathogens were given in the Table.01.

In the present study, the effect of ethanolic extract of *Aristolachia indica* Linn against various microbes and their potency were qualitatively and quantitatively assessed by the presence or absence of inhibition zones and zone diameters. The results were tabulated in table 01. The results showed that the ethanolic extract of *Aristolachia indica* dose dependently inhibit the growth of microorganisms such as bacteria's and fungi. The extract showed poor antimicrobial activity against, *Salmonella typhi* (MTCC 733), *Staphylococcus aureus* (MTCC 96), *Streptococcus pneumoniae* (MTCC 655), *Escherichia coli* (MTCC 724), *Vibrio vulnificus* (MTCC 1145) and *Candida albicans* (MTCC 227). The extract exhibited moderate antimicrobial activity against *Bacillus subtilis* (MTCC 441), *Proteus vulgaris* (MTCC 426) and *Aspergillus flavus* (MTCC 1344). *Aristolachia indica* exhibited significant antimicrobial activity against *Pseudomonas aeruginosa* (MTCC 741), *Aspergillus niger* (MTCC 1344) and *Aspergillus fumigatus* (MTCC 1344) .

Table 1: *In vitro* antimicrobial activity of *Aristolachia indica* Linn

Microorganism	Inhibition zones in mm			
	Std 10µg/ml	100 mg/ml	50 mg/ml	25 mg/ml
Bacteria				
	Clotrimaxazole			
Salmonella typhi (MTCC 733)	35	22	19	18
Bacillus subtilis (MTCC 441)	33	26	23	17
	Ampicillin			
Staphylococcus aureus (MTCC 96)	38	25	22	18
Streptococcus pneumoniae (MTCC 655)	39	20	19	14
Pseudomonas aeruginosa (MTCC 741)	25	24	22	16
Proteus vulgaris (MTCC 426)	32	24	23	23
	Kanamycin			
Escherichia coli (MTCC 724)	36	18	16	15
Vibrio vulnificus (MTCC 1145)	32	22	20	19
Fungi				
	Penicillin			
Aspergillus niger (MTCC 1344)	22	20	18	15
Aspergillus flavus (MTCC 1344)	36	28	22	21
Aspergillus fumigatus(MTCC 1344)	25	22	20	20
Candida albicans (MTCC 227)	37	26	25	20

The present results showed that medicinal plants which were used in traditional medicine against infections may have some antimicrobial activity. This is true for *Aristolachia indica* Linn ethanolic extract. These results were consistent with traditional uses of the plant leaves *Aristolachia indica* Linn[4] and pharmacological actions of its leaves[6] and essential oil[5]. The concentrations of ethanolic extract needed for bacteristasis were 10000 times higher than the concentrations of usual antibiotics. However, the Soxhlet extracts were very crude preparations, and further purifications may yield more potent compounds[12.] Furthermore, the detection of antimicrobial activities—albeit to varying extents— indicates that the plants may be sources for bactericidal and fungicidal drugs. The present results and similar results in the past[12-14] showed that there may be plenty of unexploited natural sources of compounds in higher plants, which can be used to control microorganisms. Bioassay-guided research could reveal new, renewable and more potent compounds in these plants.

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