



## STUDIES ON THE ANTIMICROBIAL ACTIVITY OF AQUEOUS AND ALCOHOLIC EXTRACTS OF *ICHNOCARPUS FRUTESCENS* LEAVES

**B. L. ANOOP<sup>a</sup>, I. J. KUPPAST<sup>\*</sup>, H. G. PRASHANTHA KUMAR<sup>b</sup>,  
R. ONKARAPPA<sup>b</sup> and S. A. GOUTHAM<sup>b</sup>**

Department of Pharmacology, National College of Pharmacy, SHIMOGA – 577201 (K.S.) INDIA

<sup>a</sup>Balekodlu, SHIMOGA – 577201 (K.S.) INDIA

<sup>b</sup>Sahyadri Science College, SHIMOGA – 577201 (K.S.) INDIA

### ABSTRACT

The aqueous and ethanol extracts of leaves of the *Ichnocarpus frutescens* were screened for their antimicrobial activity by well-in-agar method. The extracts were tested against bacteria like *Bacillus subtilis*, *staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa* and *Vibrio cholerae* and fungi like *Saccharomyces cervisiae*, *Cryptococcus neoformans*, *Candida albicans* (unicellular fungi), *Aspergillus niger* and *Fusarium sps* (multicellular fungi). The susceptibility of the microorganisms to the extracts of the leaves of plant was compared with selected standard antibiotics (Chloramphenicol and fluconazol). The water extracts gives the inhibition zone of 10 mm on *Fusarium sps* (fungi). For *Vibrio choerae*, the water extract gives the inhibition zone of 11 mm. The rest of organisms tested were resistant to both extracts.

**Key words:** Microbial activity, *Ichnocarpus frutescens*, Well-in-agar.

### INTRODUCTION

The use of plants and their extracts to treat infections is an old age practice of man. The traditional medical practice has been known for centuries in many parts of the world. Medicinal plants represent a rich source of antimicrobial agents. Plants are used medicinally in different countries and are source of many potent and powerful drugs<sup>1</sup>. A wide range of medicinal plant parts are used as raw drugs and they possess varied medicinal property<sup>2</sup>.

*Ichnocarpus frutescens* (apocyanaceae) is medicinally used for asthma, fever, inflammatory diseases, headache and snakebite etc.<sup>3,4</sup> by the local Vaidyas and in various

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\* Author for correspondence; E-mail: [ijkuppast@gmail.com](mailto:ijkuppast@gmail.com), [kuppastij@yahoo.com](mailto:kuppastij@yahoo.com)

Ayurvedic formulations. In the present study, the literature reveals that no antimicrobial activity of leaf extract has been carried out so far. In view of this, the present investigations were therefore carried out to evaluate the efficacy of aqueous and ethanol extracts of *Ichnocarpus frutescens* leaves against some microbes.

## EXPERIMENTAL

### Plant material

The fresh leaves of *Ichnocarpus frutescens* were collected in Balekodlu village, Shimoga Dist. Karnataka, India. The plant was identified by Department of Botany, Sahyadri Science College, Shimoga. The collected plant material was air dried under shade, and dried leaves were crushed into powder by the mechanical blender.

### Ethanol extract

The dried powdered material was extracted with ethanol (85-90°C) in a Soxhlet apparatus. The extract was filtered and the solvent was removed by distillation under vacuum (yield 12.8%). 100 mg of dried extract was dissolved in 15 mL of ethanol and diluted by 85 mL of sterile distilled water.

### Water extract

The dried powder was subjected for the cold extraction. Then it was filtered and dried by distillation under vacuum and (yield 10.29 %). This dried water extract of 100 mg was dissolved in 100 mL of sterile distilled water.

### Phytochemical analysis

Qualitative phytochemical studies of water and ethanol extracts of leaves were done to identify its alkaloid, carbohydrate and glycoside, saponin, protein and amino acid, phenolic compound, flavonoids and phytosterols by using suitable chemicals and reagents<sup>5</sup>. The qualitative phytochemical screening showed that presence of alkaloids, glycosides, phenols and flavonoids.

### Screening for antimicrobial activity

*In vitro* antimicrobial activity of plant extract was tested by well-in-agar method.<sup>6,7</sup> The pure cultures of bacteria and Fungi used were obtained by the National Centre for Industrial Microbiology, Pune (NCIM), where as *Fusarium sps*, *vibrio cholerae* and *Bacillus*

*subtilis* were extracted and identified by the experts in the Department of Microbiology, Sahyadri Science College, Shimoga. Sabourand dextrose agar media (SDA), potato dextrose agar media and nutrient agar media were prepared and used for the activity. Water extract solution (0.1 mL) was filled into well in the plates of agar medium being inoculated with microorganisms (0.1 mL of 15 mL of alcohol in 85 mL of water was used as the solvent control). The standard antibiotics (Chloramphenicol and fluconozol) of 1 mg/mL were used for this antimicrobial activity activity study. The plates were incubated at 37°C for 24 hr (bacteria) and 72 hr at 28°C for fungi. The diameters of the inhibition zones were measured in mm.

**Table 1: Zones of inhibition (mm) of aqueous and ethanolic extracts on the microorganisms**

Organism	Y <sub>1</sub>	Y <sub>2</sub>	Y <sub>3</sub>	F <sub>1</sub>	F <sub>2</sub>	B <sub>1</sub>	B <sub>2</sub>	B <sub>3</sub>	B <sub>4</sub>	B <sub>5</sub>
Standard antibiotic	10	-	11	12	-	25	27	26	19	27
Solvent	12	-	M	10	-	-	10	-	-	-
Water extract	M	-	-	10	-	-	11	-	-	-
Ethanol extract	12	-	M	-	-	-	-	-	-	-

– : No Inhibition; M: Marginal; Y<sub>1</sub>: *Saccharomyces cerevisiae*; Y<sub>2</sub>: *Cryptococcus neoformans*; Y<sub>3</sub>: *Candida albicans*; F<sub>1</sub>: *Fusarium sps*; F<sub>2</sub>: *Aspergillus niger*; B<sub>1</sub>: *Staphylococcus aureus*; B<sub>2</sub>: *Vibrio cholerae*; B<sub>3</sub>: *Escherichia coli*; B<sub>4</sub>: *Bacillus subtilis* and B<sub>5</sub>: *Pseudomonas aeruginosa*

## RESULTS AND DISCUSSION

The plant extracts were tested for both; antifungal and antibacterial activity. The water extract gives the zone of inhibition of 11 mm against *Vibrio cholerae*. For *Fusarium sps*, zone of inhibition was 10 mm for water extract. The rest of the organisms were resistant to both extracts of *Ichnocarpus frutescens*. The water extract of the plant having slight efficacy against *Vibrio cholerae* and *Fusarium sps*. The zone of inhibition for alcoholic extracts is almost nil except *Saccharomyces cerevisiae* (12 mm).

Most of the flavonoids have been found to exhibit antibacterial and antifungal activity<sup>8</sup>. These flavonoids act as antioxidants and they exhibit lot of pharmacological activity by their antioxidant nature. They also inhibit lot of enzymes in cell and micro-

organisms, which are required to carry out normal metabolic reaction. Flavonoids claims to reduce the infectivity and cellular replication of various bacteria and viruses. This could be probable mechanism of action of antimicrobial activity of *Ichnocarpus frutescens*. However, further investigations are required to find out, which active ingredient is responsible for this antimicrobial activity.

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