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Research Article

EVALUATION OF ANTIMICROBIAL ACTIVITY OF LEAVES OF *GISEKIA PHARNACEOIDES*

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ABSTRACT

The aim of the present research was focused on the antimicrobial properties of leaves of *Gisekia pharnaceoides* by *in vitro* approach. Chloroform, ethanol and aqueous extracts of leaves were evaluated against two Gram positive (*Staphylococcus aureus, Staphylococcus epidermidis)*, two Gram negative (*Escherichia coli, Klebsiella pneumoniae*) bacterial strains and two fungal strains (*Aspergillus niger*, *Aspergillus fumigatus*) by agar disc diffusion method. Minimum Inhibitory Concentration (MIC) required for cessation of microbial growth was evaluated by agar streak dilution method. The study also includes preliminary phytochemical screening of the taxon. All the extracts showed concentration dependent activity against the microorganisms investigated. It may be concluded that the aqueous extract exhibited significant activity against the test organisms than the other two extracts.

Keywords: *Gisekia,* Antibacterial, Antifungal, Minimum Inhibitory Concentration, Leaves

INTRODUCTION

Gisekia pharnaceoides Linn, commonly known as Manalikeerai in Tamil [1], belongs to the family Molluginaceae [2]. It is one of the sources for controversial drug *Elavaluka* used in Ayurvedic system of Medicine [3]. It is a diffuse, somewhat succulent herb [4]. The plant cures scabies, rhinitis, bronchitis, loss of appetite, heart troubles, leprosy, leucoderma and urinary diseases. It is also given for chest disorders, worm infestation and mental disorders [5]. The plant is reported to contain oxalic, succinic, tartaric, citric acids, triacontane, dotriacontane, myristone and tetracosanol. 50% ethanolic extract of the plant showed CNS depressant activity [6]. Chloroform extract of this plant exhibited a strong anthelmintic, and antimicrobial activities [7]. In the present work, an attempt was made to study the antimicrobial screening of the extracts (Chloroform, ethanol and aqueous extracts) of leaf of *G. pharnaceoides,* which could be useful for the development of new tools as antimicrobial agents for the control of infectious diseases.

MATERIALS AND METHODS

The plant specimens of *Gisekia pharnaceoides* were collected from Udangudi, Tuticorin district, Tamilnadu, India. The specimens were identified and authenticated by Prof. P. Jayaraman, Director of Plant Anatomy Research Centre, West Tambaram, Chennai. The voucher specimen (No: PARC/2010/521) has been deposited in the same Institution. The coarsely powdered leaves were extracted successively with chloroform, ethanol and water by cold maceration process. The extracts were reduced to a molten mass by rotary vacuum evaporator and the respective yields of chloroform, ethanol and water extracts were 0.7%w/w, 1.2%w/w and 1.1 %w/w respectively. The preliminary phytochemical screening was carried out by using standard procedure.

Antimicrobial activity study

The microbes used to determine the antimicrobial activity are *Staphylococcus aureus* ATCC 6538P, *Staphylococcus epidermidis* ATCC 155, *Escherichia coli* ATCC 25922, *Klebsiella pneumoniae* ATCC 29665, *Aspergillus niger* ATCC 9029, *Aspergillus fumigatus* ATCC 46645. All the bacterial and fungal cultures were procured from the Institute of Microbial Technology, IMTECH, Chandigarh, India.

Screening of Antimicrobial Activity

The antimicrobial screening was performed by agar diffusion method using a paper disc [8]. Nutrient agar and Sabouraud's dextrose agar media were used for the antimicrobial screening. 1ml suspension of the microorganisms (matched with McFarland barium sulphate standard) was inoculated with 100ml of the sterilized (autoclaved at 120˚C for 30 min) medium (40-50˚C). The paper impregnated with the extracts (100, 200 and 300 µg/ml) was placed on the solidified medium. The plates were preincubated for 1h at room temperature and incubated at 37˚C for 24h and 48h for antibacterial and antifungal activities respectively. Ciprofloxacin (100µg/disc) and Ketaconazole (100µg/disc) was used as standard for antibacterial and antifungal activity respectively.

The Minimum Inhibitory Concentration (MIC) for the above organisms was found by agar streak dilution method [9]. Stock solutions of the chloroform, ethanol and water extracts were mixed with the known quantity of molten sterile agar media aseptically to provide the required concentrations. About 20 ml of the media containing the extract was poured into each sterile petridish and allowed to solidify. Microorganisms were then streaked one by one on the agar plate aseptically. After streaking, all the plates were incubated at 37±1˚C for 24hr and the plates were observed for the growth of microorganism. The lowest concentration of the plant extract required for inhibiting the growth was considered as the MIC of the extracts against bacterial and fungal strains.

RESULTS AND DISCUSSION

Medicinal plants have been used since long for mankind against various infections and non-infectious diseases because they contain natural bioactive components for therapeutic value. According to the World Health Organisation, medicinal plants are the best source to obtain a variety of drugs and 80% of population from developed countries uses traditional medicines, which have bioactive compounds derived from medicinal plants for their primary health care needs [10]. Many approaches were made to search the antimicrobial compounds with a novel chemical structure from the medicinal plants. The development of new antimicrobial compounds against different microorganisms is becoming critically important, as infectious diseases are still one of the main causes of death in the world [11]. Currently, there has been an increased interest in antimicrobial agents from the plants origin due to the resistance that microorganisms have developed against traditional antibiotics [12]. Therefore, there is a need for the investigation of new source of potential antimicrobial agents.

The results of the antimicrobial activities of chloroform, ethanol and water extracts from the leaves of *G. pharnaceoides* showed different degree of activity against the tested bacterial and fungal strains. The higher concentrations of all extracts had inhibitory effects towards the tested microorganisms. In chloroform extract, the maximum inhibition against *A. niger* (28mm), for *K. pneumoniae* (27mm) (Table 1) and in ethanol extract, the maximum inhibition against *S. aureus* and *A. niger* (29mm) (Table 2) whereas in aqueous extract, the maximum inhibition against *E. coli* (29mm) and *A. niger* (28mm) at a concentration of 300 µg/ml (Table 3). The MIC values of each

extract against the tested bacterial and fungal strains were tabulated (Table 4). The lowest MIC values were observed for chloroform (90 - 93 µg/ml), ethanol (90 - 94 µg/ml) and aqueous (89 - 95 µg/ml) extracts against the bacteria and fungi tested.

The preliminary phytochemical screening indicates the presence of alkaloids, carbohydrates, glycosides, phenolic compound, saponins, flavonoids, proteins and amino acids in the extracts of *G. pharnaceoides.*

Table 1: Zone of Inhibition of chloroform extract of *G. Pharnaceoides*

S. No.	Organisms	Zone of Inhibition			
		Standard (mm)	100µg (mm)	200µg (mm)	300µg (mm)
1.	*Staphylococcus aureus*	39	18	20	24
2.	*Staphylococcus epidermidis*	38	17	23	26
3.	*Escherichia coli*	39	15	19	25
4.	*Klebsiella pneumoniae*	39	18	23	27
5.	*Aspergillus niger*	38	16	23	26
6.	*Aspergillus fumigatus*	39	19	24	28

Table 2: Zone of Inhibition of ethanol extract of *G. Pharnaceoides*

S. No.	Organisms	Zone of Inhibition			
		Standard (mm)	100µg (mm)	200µg (mm)	300µg (mm)
1.	*Staphylococcus aureus*	39	19	23	29
2.	*Staphylococcus epidermidis*	38	18	21	25
3.	*Escherichia coli*	38	17	20	25
4.	*Klebsiella pneumoniae*	39	19	22	27
5.	*Aspergillus niger*	40	19	23	29
6.	*Aspergillus fumigatus*	40	17	21	26

Table 3: Zone of Inhibition of water extract of *G. Pharnaceoides*

S. No.	Organisms	Zone of Inhibition			
		Standard (mm)	100µg (mm)	200µg (mm)	300µg (mm)
1.	*Staphylococcus aureus*	39	16	22	26
2.	*Staphylococcus epidermidis*	39	18	23	27
3.	*Escherichia coli*	38	20	26	29
4.	*Klebsiella pneumoniae*	39	15	21	25
5.	*Aspergillus niger*	40	20	26	28
6.	*Aspergillus fumigatus*	40	18	25	27

Table 4: Minimum Inhibition Concentration of various extracts of *G. Pharnaceoides*

S. No.	Organisms	MIC (µg/ml)		
		Chloroform	Ethanol	Aqueous
1.	*Staphylococcus aureus*	93	94	94
2.	*Staphylococcus epidermidis*	92	93	93
3.	*Escherichia coli*	90	92	91
4.	*Klebsiella pneumoniae*	93	93	95
5.	*Aspergillus niger*	91	90	89
6.	*Aspergillus fumigatus*	90	91	90

CONCLUSION

It was concluded that the leaves of *G. pharnaceoides* have a stronger and broader spectrum of antimicrobial activity against the tested pathogens, and the extracts may be used to discover bioactive natural products that may serve as basic source for the development of new antimicrobial compounds to overcome the problem of increasing resistance to known traditional antibiotics.

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