

Antifungal Activity of Tubers And Seeds Extracts of *Gloriosa Superba* L. Against in Human Pathogens.



Agriculture

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ABSTRACT

The present study deals with the antifungal activity of Acetone, Dichloromethane, methanol extracts of the tubers and seeds of *Gloriosa superba* L. (Liliaceae) using disc diffusion method against human pathogens, such as *Aspergillus flavus*, *Aspergillus niger*, *Candida albicans* and *Candida glabrata*. In the present investigation all the extracts were found to be effective against five human fungal species. The maximum inhibition was observed on *Aspergillus flavus* (16.50 mm) methanol extract for seed and followed by *Candida glabrata* in methanol seed extract (16.50 mm).

INTRODUCTION

Higher plants produce hundreds to thousands of diverse chemical compounds with different biological activities (Hamburger and Hostettman, 1991). Thus, they have been used in the treatment of various human diseases for thousands of years all over the world. Most of the plants used for medicinal purpose have been identified, and their uses are well documented and described by different authors (Nadkarni, 1876; Dastur, 1985; Saradamma, 1990), but the efficiency of many of these plants is yet to be verified. Several screening carried out in different plant parts of the world. There are several reports on the antimicrobial activity of different herbal extract in different region of the world (Chung *et al.*, 2004; Nair and chanda, 2004; De Boer *et al.*, 2005; Nair *et al.*, 2005).

Plants are traditionally used in the treatment of bacterial and fungal infections for its wide range of bioactive molecules. Phytochemicals are applied as natural anti pathogenic, which can be derived from leaves, stems, barks, flowers and other parts (SajadYousuf *et al.*, 2011). The traditional plant medicine is getting back with modern science all over the globe. The extracts from medicinal plants are used in the treatment of different diseases of humans, plants and animals (Nostro *et al.*, 2000).

Gloriosa superba L. (Liliaceae) is an ornamental climbing herb native of tropical Asia and Africa often been cultivated for its beautiful flowers. The seeds and tubers of this plant have been used in traditional Indian medicine for the treatment of gout, rheumatic arthritis, in disease of skin, lever and several other purposes (Finnie and Van staden, 1994). Khan *et al.* (2008) reported antimicrobial potential of *G. superba* extracts in which excellent antifungal activity was confirmed against *Candida albicans*, *C. glabrata* and *Microsporum canis*.

MATERIALS AND METHODS

Test organisms and culture media

The clinical pathogenic fungal strains were aseptically collected from Rajah Muthiah Medical College and Hospital, Annamalai University. The fungal cultures *viz.*, *Aspergillus flavus*, *Aspergillus niger*, *Candida albicans*, *Candida glabrata* were maintained in potato dextrose broth (PDB).

Extraction of plant materials

In the present study, the fresh tubers and seeds were used to evaluate their antifungal activity. Plant extracts were prepared by cold percolation method. The tuber and seeds were air dried in room temperature for 10 days. The fully dried plant materials were powdered and weighed. The powdered tubers and seeds (5 g) each was soaked in 50 ml of different solvents such as Acetone, Dichloromethane, Methanol and kept for 48 hours with intermittent shaking. The plant extract were filtrate through Whatman's No.1 filter paper. The filtrated was collected in separate clean beaker.

Antifungal Assay

The screening of the extracts for antifungal effect was carried

out by determining the zone of inhibition using disc diffusion method. Sterile potato dextrose agar plates were prepared. Then 0.1 ml of test organism was taken from the stock (broth) and swabbed on the agar medium in aseptic condition. The filter paper disc of 2 mm diameter (Whatman's No.1 Filter paper) were prepared and sterilized. The plant extracts to be tested were prepared with various concentration *viz.*, 50 µl/ml, 100 µl/ml, 150 µl/ml and 200 µl/ml and were added to each disc of holding capacity of 10 microlitres. The sterile impregnated disc with plant extracts were placed on the agar surface with framed forceps and gently pressed down to ensure complete contact of the disc with the agar surface. Positive control disc of Fluconazole were prepared and placed on the agar surface. All the plates were incubated at 37°C for 3 to 5 days. After incubation, the area of inhibition zones was measured.

RESULT AND DISCUSSION

To investigate the antifungal activity of *Gloriosa superba* tubers and seeds. Using three different solvents such as Acetone, Dichloromethane and Methanol. The plant extracts were prepared with various concentration *viz.*, 50 µl/ml, 100 µl/ml, 150 µl/ml and 200 µl/ml were tested their antifungal effect against *Aspergillus flavus*, *Aspergillus niger*, *Candida albicans* and *Candida glabrata*. Positive control disc of fluconazole were used. The results are presented in (Table 1 to 4) (Figure 1 to 4).

The methanol extract exhibited pronounced inhibition against all tested organisms. The maximum inhibition was observed on *Aspergillus flavus* seed extract 16.50 mm (Table-1) and *Candida glabrata* in methanol seeds extract 16.50 mm, (Table-4). The maximum inhibition of dichloromethane seeds extract in 200µl against *Aspergillus flavus* (16 mm) (fig-1), *Aspergillus niger* (13 mm), *Candida albicans* (15 mm), *Candida glabrata* (15 mm). The inhibition of acetone extract of seeds contain (15.50mm) for *Aspergillus niger* (Table-2) (fig-2), (14 mm) for *Candida albicans* (fig-3), (14.50 mm) for *Candida glabrata*. The positive control of fluconazole recorded the inhibition of 11.00 mm, 11.00 mm, 12.00 mm and 11.00 mm in *Aspergillus flavus*, *Aspergillus niger*, *Candida albicans* and *Candida glabrata* respectively. The methanol extract of tuber exhibited maximum antifungal activity against 200µl in *Aspergillus flavus* (15.50 mm), *Aspergillus niger* (15.00 mm), *Candida albicans* (15.50 mm) (Table-3) and *Candida glabrata* (16.00 mm) (fig-4). Similar observations were recorded in methanol extracts of *Gloriosa superba* showed high antifungal activity against *Aspergillus flavus* and *Candida glabrata*.

CONCLUSION

From the study it can be concluded that the extracts prepared from the tubers and seeds of *G. superba* are a source of different secondary metabolites which may act in synergy to produce an increased activity against microbes. The result may justify the use of plant in treatment of certain skin diseases.

TABLE-1Antifungal activity of plant extracts of *Gloriosa su-*

perba against *Aspergillus flavus*

Plant	Extract	Solvents used	Area of inhibition zone (mm)				
			*C	50µl	100 µl	150 µl	200 µl
Gloriosa superba	Tuber	Acetone	11.00	10.00	12.00	13.00	13.50
		Dichloromethane	10.00	11.00	12.00	13.00	14.00
		Methanol	11.00	12.00	12.50	13.00	15.50
	Seed	Acetone	10.00	11.50	13.00	15.00	15.50
		Dichloromethane	11.00	12.50	13.00	15.00	16.00
		Methanol	11.00	11.00	13.00	14.00	16.50

TABLE -2 Antifungal activity of plant extracts of *Gloriosa superba* against *Aspergillus niger*

Plant	Extract	Solvents used	Area of inhibition zone (mm)				
			*C	50µl	100 µl	150 µl	200 µl
Gloriosa superba	Tuber	Acetone	10.00	10.50	11.00	12.00	13.00
		Dichloromethane	11.00	11.50	12.00	13.00	14.00
		Methanol	10.00	11.00	12.50	13.00	15.00
	Seed	Acetone	10.00	12.50	13.00	14.00	14.50
		Dichloromethane	10.00	11.00	11.50	12.00	13.00
		Methanol	11.00	12.00	12.00	12.50	13.00

TABLE - 3 Antifungal activity of plant extracts of *Gloriosa superba* against *Candida albicans*

Plant	Extract	Solvents used	Area of inhibition zone (mm)				
			*C	50µl	100 µl	150 µl	200 µl
Gloriosa superba	Tuber	Acetone	10.00	12.00	13.00	13.50	14.00
		Dichloromethane	11.00	11.00	12.50	13.00	15.00
		Methanol	11.00	12.00	13.00	13.50	15.50
	Seed	Acetone	10.00	11.00	12.50	13.00	14.00
		Dichloromethane	12.00	12.00	13.50	14.00	15.00
		Methanol	11.00	11.00	13.00	14.00	16.00

TABLE - 4 Antifungal activity of plant extracts of *Gloriosa superba* against *Candida glabrata*

Plant	Extract	Solvents used	Area of inhibition zone (mm)				
			*C	50µl	100 µl	150 µl	200 µl
Gloriosa superba	Tuber	Acetone	11.00	11.00	12.00	12.50	13.00
		Dichloromethane	10.00	11.00	12.50	13.00	14.00
		Methanol	11.00	12.00	13.00	15.50	16.00
	Seed	Acetone	10.00	11.00	12.00	13.00	14.50
		Dichloromethane	11.00	12.50	13.00	13.50	15.00
		Methanol	10.00	12.00	14.00	15.00	16.50

Fig- 1 Antifungal activity of plant extracts of *Gloriosa superba* against *Aspergillus flavus*

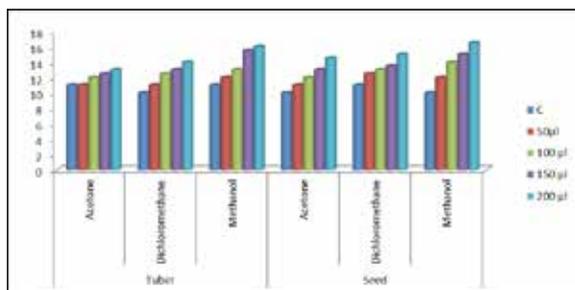


Fig- 2 Antifungal activity of plant extracts of *Gloriosa superba* against *Aspergillus niger*

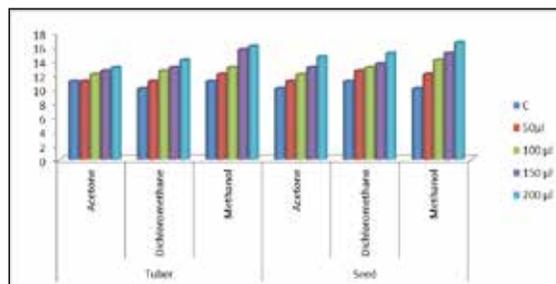


Fig- 3 Antifungal activity of plant extracts of *Gloriosa superba* against *Candida albicans*

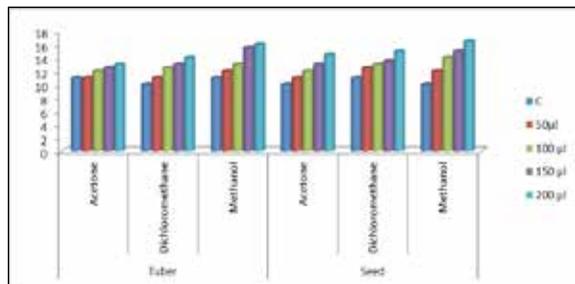
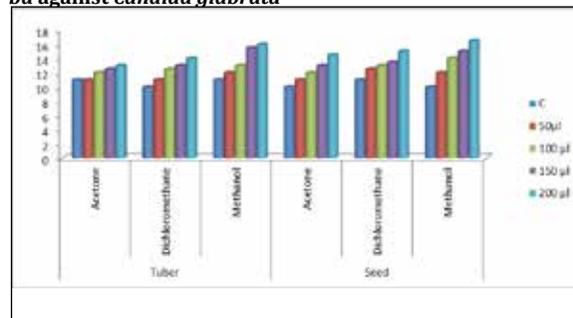


Fig- 4 Antifungal activity of plant extracts of *Gloriosa superba* against *Candida glabrata*



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