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Research Article

IN VITRO EVALUATION OF ANTI MYCOTIC ACTIVITY OF ETHANOLIC EXTRACT OF *GLYCYRRHIZA GLABRA*

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ABSTRACT

Objective: The aim of the present study was to assess the anti fungal activity of ethanolic extract of *Glycyrrhiza glabra*. *Glycyrrhiza glabra* commonly called as liquorice or sweet root is one of the oldest known herbal remedies famous for its vast variety of therapeutic properties. Ethanolic extract of *Glycyrrhiza glabra* was tested for antifungal activity against *Candida albicans, Aspergillus niger, Aspergillus fumigates, Mucor spp and Penicilium marneffei.*

Method: Disc diffusion technique was followed for screening anti fungal activity. The discs were loaded with 100µl of ethanolic extracts at different concentrations [500ug/disc,1000ug/disc and 2000µg/disc]. Positive controls used were fluconazole (10 mcg/disc) and amphotericin B (100 units/disc).

Result : After incubation at 28oC for 48 hours, the zone of Inhibition was measured.

Conclusion: The extract at different concentrations showed varying degree of antifungal activity against the micro organisms tested compared to standard**.**

Keywords: *Glycyrrhiza glabra*, disc diffusion, zone of inhibition, anti mycotic activity.

INTRODUCTION

The very old folk medicine is based on the use of plant and plant extracts. There are innumerable types of indigenous plants that have been used by people for centuries in the treatment of many ailments. The use of herbs to treat disease is almost universal and is often more affordable than expensive modern pharmaceuticals.[1] Many of the phytochemicals found in the herbs have beneficial effects and can be used to treat human diseases. It has become increasingly more in recent years as scientific evidence about the effectiveness of herbal medicine has become more widely available.[2] There are herbal remedies that have been tested for their use as a treatment for many fungal diseases.

Glycyrrhiza glabra, also known as liquorice and sweet wood, is native to the Mediterranean and certain areas of Asia. It is a perennial herb which possesses sweet taste. The main taproot, which is harvested for medicinal use, is soft, fibrous, and has a bright yellow interior. It was one of the most widely known medicines in ancient history, and records of its use include Assyrian tablets of around 2000 BC and cortisone has been found useful for arthritis and allergies.[3] In addition licorice has been used for mild Addison's disease and other adrenal insufficiencies, such as hypoglycemia. [4] Licorice also acts like the hormone, ACTH, causing sodium retention, potassium depletion, and water retention. The herb contains glycyrrhizin, glycyrrhetinic acid, flavonoids, asparagine, iso-flavonoids, and chalcones. The glycoside, glycyrrhizin has a similar structure and activity as the adrenal steroids.[5.6] It also possess good anti bacterial, [7] anti fungal,[8] anti oxidant,[9] antitussive,[10] hepatoprotective [11] and anti inflammatory activity.[12] Historically, the dried rhizome and root of this plant were employed medicinally as an expectorant and carminative. It is used for treating upper respiratory ailments including coughs, hoarseness, sore throat and bronchitis.

TEST MICROORGANISMS

Fungal strains used were *Candida albicans, Aspergillus fumigates, Aspergillus niger, Mucor sps,* and *Penicillium marneffei.* The organisms were obtained from Department of Microbiology, Saveetha Medical College and maintained in SDA slope at 4°C.

METHODOLOGY

The extracts were prepared in the following concentrations in sterile water. 5mg/ml and 10mg/ml and 20mg/ml. 100µl of extract of different concentrations were loaded on sterile filter paper discs measuring 6mm in diameter, so that the concentration of the extract on each disc was 500µg, 1000 µg and 2000µg respectively. The discs were dried and kept aseptically.

SCREENING OF ANTIFUNGAL ACTIVITY [DIS DIFFUSION TECHNIQUE]

The ethanolic extract of *Glycyrrhiza glabra* was screened for antifungal activity by disc diffusion method. Activated cultures of fungal strains in Sabouraud's broth were adjusted to 0.5 McFarland standards [108 cfu/ml].[13-17] 100 µl of the inoculum was introduced to molten Sabourauds dextrose agar and poured in the sterile petri plates and allowed to set. Sterile filter paper discs (6.0 mm diameter) impregnated with 2000µg/disc, 1000 µg /disc and 500 µg /disc of the plant extract dissolved in sterile water were placed on fungal seeded plates and incubated at 28oC for 48 hrs. As a positive control, Fluconazole (10 mcg /disc) and Amphotericin B (100 units /disc) were used. Following an incubation period of 48 hrs, plates were removed from the

incubator and antifungal activity was evaluated by measuring zones of inhibition of fungal growth. Clear zones within which fungal growth was absent were measured and recorded as the diameter (mm) of complete growth inhibition. The whole experiment was performed three times to minimize error.

RESULTS AND DISCUSSION

Effect of three different concentrations (2000, 1000, 500,µg /disc) of the ethanolic extract of *Glycyrrhiza glabra* was tested against the fungal strains using disc diffusion technique. All the concentrations of the test solution inhibited the fungal species with varying degree of sensitivity. The antifungal activity of the extract against the fungal strains is shown in Table 1. The extract showed good antifungal activity against the two *Aspergillus Spp*, *Aspergillus niger* and

Aspergillus fumigates at all concentrations with a maximum zone of inhibition of 21mm and 20mm diameter respectively at 2000 µg

/disc concentration. The lowest concentration of the extract showed least activity against *Mucor sps* and P*enicillium morneffei,* while the higher concentrations showed antifungal activity with a maximum zone diameter of 18mm and 17mm respectively at 2000 µg/disc concentration. With *Candida albicans*, the lowest concentration of the extract showed a zone of inhibition of 9mm while the highest concentration showed an inhibitory zone of 20mm. From the results, it was evident that the lower concentration showed very weak while the higher concentration of the extract showed good antifungal activity against all the fungal strains tested.

Table 1: Antifungal activity of ethanolic extract of the *Glycyrrhiza glabra*

Extract	Conc [µg]	Zone of inhibition [in mm diameter]				
		B1	B2	B3	B4	B5
Ethanolic	500	09	10	12	07	08
	1000	15	20	21	15	13
	2000	20	21	20	18	17
Fluconazole	10mcg /disc	24	21	22	22	23
Amphotericin B	100mcg/disc	25	23	20	24	25

B1-*Candida albicans,* B2-*Aspergillus fumigates,* B3-*Aspergillus niger*, B4-*Mucor sps,* B5-*Penicillium marneffei.*

Over the past two decades fungal infections have evolved into important cause of morbidity and mortality in modern medicine. The prevalence of resistance to antifungal agents has significantly increased. So it makes necessary to discover new classes of antifungal compounds to treat fungal infections. The research on natural products derived compounds has accelerated in recent years due to their importance in drug discovery.[18] Plants are rich source of bioactive secondary metabolites of wide variety such as tannins, terpenoids, alkaloids, and flavonoids, which are reported to have in vitro antifungal properties. A series of molecules with antifungal activity against different strains of fungus have been found in plants, which are of great importance. The results obtained from our study shows that ethanolic extract of *Glycyrrhiza glabra* has got a very good anti mycotic activity against the selected fungal species

CONCLUSION

The results of present investigation clearly indicate the antifungal activity of ethanolic extract of the *Glycyrrhiza glabra*. The anti-fungal activities could be enhanced if active components are purified and adequate dosage determined for proper administration. Further studies on their cytotoxicity or toxicity will be beneficial in providing data on the possible harmful effects of this extract commonly used by the numerous local communities. Thus, the study ascertains the value of plants used in Ayurveda, which could be of considerable interest to the development of new drugs.

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