

Short Communication

Anti-fungal potential of tridhara (*Tridax procumbens*) leaves

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Abstract: The antifungal activity of methanolic extracts of leaves of *Tridax procumbens* was studied. Fungal strains *Aspergillus niger* and *Aspergillus ocraceus* were selected for the study. Disc diffusion method was applied against the selected fungal strains and compared to the drug ciprofloxacin to observe the antifungal activity of the methanolic extracts of *Tridax procumbens* leaves. The present study demonstrated that the disks of methanolic extracts of *T. procumbens* showed effective inhibition against *A. niger* and *A. ocraceus* compared to ciprofloxacin. The results of the current investigation suggest that the methanolic extracts of *T. procumbens* leaves can be used for the treatment of the diseases caused by the tested organism and it has significant scope to develop a novel broad spectrum of antifungal herbal formulations.

Keywords: tridhara; extracts; medicinal plants; leaves

1. Introduction

Infectious diseases are one of the leading causes of death throughout the world that costs so many lives especially in the tropical countries, which are also becoming a serious problem in developed countries as resistance has grown within the infectious organisms against the antibiotics applied to eradicate them. In addition, antibiotics are sometime introduced with adverse effects including allergic reactions, immuno suppressant and hypersensitivity. Due to these reasons there is a constant need of new and effective therapeutic agents. Therefore, there is a need to develop alternative antimicrobial drugs for the treatment of infectious diseases from medicinal plants. Medicinal plants have been tested for hypoglycemic, antimicrobial and biological activities as safe natural resources. They also play important role in modern medicines (Hassawi *et al.*, 2006; Bhatt *et al.*, 2009). It is obvious that most synthetic drugs have their origin from plants (Ríos *et al.*, 2005).

T. procumbens (L) is a common medicinal herb used by ethno-medical practitioners belonging to family Asteraceae. The plant is a procumbent herb and is valued for its antifungal properties (Sahoo *et al.*, 1998). *T. procumbens* (L) is commonly known as “Tri-dhara” and in English popularly known as “coat buttons” because of the appearance of its flower. *T. procumbens* (L) is a common antifungal, antimicrobial and medicinal herb used by ethno-medical. Tridax plant is present throughout Bangladesh and is employed as indigenous medicine for verity of ailments. It has been found to possess significant medicinal properties against blood pressure, malaria, dysentery, diarrhea, headache, wound healing, it also prevent hair fall and check hemorrhage from cuts and bruises. Its flowers and leaves possess antifungal properties (Ali *et al.*, 2001; Pathak *et al.*, 1991).

The plant also shows various pharmacological activities like antifungal activity, antidiabetic, analgesic and market depressant action on respiration (Suresh *et al.*, 2004; Durgacharan *et al.*, 2010; Hemalatha *et al.*, 2008; Prakash *et al.*, 1989; Hitesh *et al.*, 2006). This study aims to investigate the antifungal activity of ethanolic

extract of leaves of *T. procumbens* (L) against *Aspergillus niger* and *Aspergillus ocraceous* comparing with ciprofloxacin.

2. Materials and Methods

2.1. Plant material

The leaves of *T. procumbens* plant was collected from local area of Jessore University of Science and Technology campus region of Jessore. It was then botanically identified. The cleaned plant parts are then allowed for complete shade drying and then made to fine powder with a mechanical grinder and stored in an airtight container.

2.2. Preparation of the extracts

The coarse powders from *T. procumbens* leaves were then subjected to successive extractions by methanol. The extraction was placed on a shaker at 40°C temperature for 24 hours for mild shaking. The crude extracts were then filtered by passing the extracts through filter paper and then concentrated under vacuum at 40°C by using a rotary evaporator. The residual extracts were stored in refrigerator at 4°C in small and sterile plastic bottles.

2.3. Fungal strain

Strains of the *A. niger* and *A. ocraceous* were obtained from Regional Agricultural Research Station. The fungus was grown at 27 °C on potato dextrose agar (PDA). Spores of the fungus were collected from cultures on agar plates after 3 days as described (Broekaert *et al.*, 2011).

2.4. Preparation of potato dextrose agar (PDA) media

Potato is boiled with water for 30 minutes then 50ml potato extract took in a conical flask with 1gm glucose and P^H was adjusted to 5.6 for growing fungus. Finally 1gm agar was added in the conical flask.

2.5. Preparation of test sample

100 mg of the leaf extract were accurately weighed by the electronic balance and taken in small volumetric flasks. Then 1600 µl of methanol was added and triturated in unidirectional manner using a vortex mixer. So, these samples were prepared by addition of solvent calculatively.

2.6. Application of discs

Three types of discs were used for antibacterial screening: 1. sample discs, 2. standard discs and 3. blank discs. Each potato dextrose agar (PDA) plates were divided into four portions i.e. two for samples one for standard (positive control) and one for blank (negative control).

2.7. Sample disc application

Sterile filter paper discs (5 mm in diameter) were taken as sample discs were placed on surface of the media at accurate positions. 10 µl of the test sample from 25 µg/µl and 50 µg/µl solutions were applied on the discs aseptically under the laminar air flow to get concentration of 250 µg and 500 µg per disc respectively for each sample.

2.8. Blank disc application

Sterile filter paper discs (5 mm in diameter) were taken as sample discs. 10 µl methanol was applied on the blank discs as negative control. They ensure that the residual solvent's activity and the filter paper were not active themselves.

2.9. Standard disc application

Ciprofloxacin (5 µg/ disc) antibiotic discs were used as positive control to ensure the activity of standard antibiotic against the test organisms as well as for comparison of the response produced by the known antimicrobial agent with that produced by test samples. Ciprofloxacin (5 µg/ disc) was placed on the labeled portion of the petri dish. Finally the plates were incubated upside down at 37°C for 16-18 hours.

3. Results

Methanolic extracts were found to be most active and significant than corresponding organic extracts. Methanolic extracts were found to be active against two tested fungi (*A. niger* and *A. ocraceous*). The fungal strains *A. niger* and *A. ocraceous* shown zone of inhibition 13 mm and 12 mm respectively (Figure 1). Negative control (disc containing only solvent) showed no zone against any fungus. The positive controls (ciprofloxacin) also produce zone of inhibition of 11 mm and 10 mm against *A. niger* and *A. ocraceous* respectively (Table 1). The results demonstrate that methanolic extracts of the selected plant leaves possess challenging antifungal activity against the studied fungal strains compared with the familiar antifungal ciprofloxacin.

4. Discussion

The therapeutic value of the medicinal plants lies in the chemical constituents present in it. The phytochemical constituents are the main agents for the bioactivity of plant extracts. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids and phenolic compounds (Hill, 1952). Antimicrobial activity also depends on variation of extraction and parts of the plant are being used. Microorganisms are the concealed enemies to the mankind. There are a lot of antifungal drugs of which some are discovered or established and over 250,000 undiscovered flowering plants with medicinal properties exist worldwide (Madureira *et al.*, 2008). Hence, the last decade witnessed an increase in the investigations on plants as a source of human disease management and more natural antifungals have driven scientists to investigate the effectiveness of inhibitory compounds such as extracts from plant (Nasar-Abbass *et al.*, 2004). There are several reports of antibiotics resistance of human pathogens to available antibiotics (Ganguly *et al.*, 2001; Martino *et al.*, 2002). The main objective of this work is to increase the utilization of biomass from herb in order to isolate new biologically active compounds. This study deals with two pathogenic fungal strains. In the present work, the antifungal potential of methanolic extracts of *T. procumbens* has been determined against microorganisms i.e., *A. niger* and *A. ocraceous*. In this study methanolic extracts is found to be very effective in inhibiting the growth of the tested fungal strains comparing with Ciprofloxacin the commercial antifungal drug. This investigation has opened up the possibility of the use of this plant in drug development for human and further studies are needed to isolate and characterize the bioactive principles to develop new antifungal drugs.

Table 1. Diameter of zone of inhibition (mm).

Sample fungi	Methanol (mm)	Ciprofloxacin (mm)
<i>A. niger</i>	13	11
<i>A. ocraceous</i>	12	10

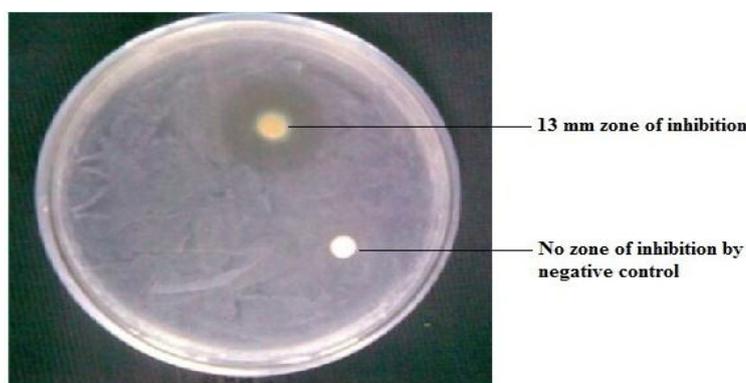


Figure 1. Methanol extract of *T. procumbens* produced highest zone of inhibition (13 mm) against *A. niger*.

5. Conclusions

The extracts of *T. procumbens* were found to be effective antifungal agents against human pathogens. This study paves the way for further attention and research to identify the active compounds responsible for the plant biological activity with the required minimum zone of inhibition. Further studies should be undertaken to elucidate the exact mechanism of action by which extracts exert their antifungal effect to identify the active

ingredients which can be used in drug development program for safe health care services. *T. procumbens* is a very commonly used medicinal plant for wound healing and other inflammations even in eyes and can be used for the development of new drug formulation for treating bacterial and fungal infections and also for the elimination of the concern of microbial resistance.

Conflict of interest

None to declare.

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