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Available online at: www.jpardonline.com**Evaluation of antifungal activity of *Gmelina arborea* Roxb. fruits****Bhabani Shankar Nayak^{*1}, P. Ellaiah², Subas Chandra Dinda¹**¹Department of Pharmaceutics, Institute of Pharmacy & Technology, Salipur, Cuttack – 754202, Odisha, India.²Department of Pharmaceutical Technology, Jeypore College of Pharmacy, Rondapalli, Jeypore-764002, Koraput, Odisha, India.³Department of Pharmacy, School of Pharmacy, Mekelle University, Ethiopia.

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ABSTRACT: Background: *Gmelina arborea* Roxb. is found in the tribal areas of Koraput and Ganjam district. It is extensively used traditionally by the tribal people as anthelmintic, antimicrobial, antidiabetic, hepatoprotective and antiepileptic. **Aim:** The present study is an attempt for the investigation of antifungal activity of different fruit extracts of plant *G. arborea*. **Method:** The fruits of *G. arborea* were extracted by Soxhlation using ethanol, ethyl acetate, n-butanol and petroleum ether as solvents. The antifungal activity of above extracts was evaluated by disc diffusion method using *Candida albicans* and *Aspergillus niger* as test organisms. **Results:** The data obtained from pictorial evidence revealed that no such zone of inhibition was found out from extracts of *G. arborea*. **Conclusion:** It could be concluded that the fruits of *G. arborea* does not possess antifungal activity.

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Key words: *Gmelina arborea*, antifungal, disc diffusion, *C. albicans*, *A. niger*.

INTRODUCTIONS:

The need for effective antifungal drugs has been realized more accurately with the emergence of acquired immunodeficiency syndrome (AIDS) and AIDS related complex (ARC) diseases, which are often associated with opportunistic infections. The incidence of opportunistic fungal infections is increasing at an alarming rate in the case of the patients undergoing treatment with immunosuppressive drugs, intensive chemotherapy, suffering AIDS and neonates ^[1]. These mycoses are very difficult to eradicate constituting

enormous challenges for healthcare providers. Although it appears that there is an array of drugs for the treatment of systemic and superficial mycoses, none of them is ideal in terms of efficacy, safety or antifungal spectrum [162]. Many of the drugs have undesirable side effects or are very toxic (Amphotericin B), produce recurrence, show drug – drug interactions (azoles) or lead to the development of resistance (Fluconazole, 5 – flucytosine). Numerous useful drugs from higher plants have been discovered by following up ethnomedical uses [2,3]. The diversity of plants growing in India, along with their known ethnopharmacological uses, offer an enormous possibility of findings novel structures with antifungal properties.

Gmelina arborea Roxb (Family *Verbenaceae*) fruits are oval in shape, ¾ inches in length and are yellow in color. The fruits are sweet in taste and sometimes astringent [4,5]. The plant, *G. arborea* was reported to have several medicinal properties such as aphrodisiac, astringent, analgesic, antipyretic, antidiabetic, diuretic, anti-inflammatory and tonic characteristics [6]. The literature survey reveals that fruits of *G. arborea* contain cardiac glycosides, flavonoids and steroids. The ethanol extract contains alkaloids, carbohydrates, anthraquinone glycosides, gums, mucilages, tannins, phenolic compounds and flavonoids [7]. The study prompted us to investigate the antiepileptic activity of *G. arborea* fruit extracts.

MATERIALS AND METHODS:

Drugs and Chemicals:

Clotrimazole was received as gift sample from Glenmark Pharmaceutical Ltd., Baddi, India. Dimethyl sulfoxide, ethanol AR and ethyl acetate AR 60-80°C (Emsure® ACS) were procured from Merck Pvt. Ltd., Navi Mumbai, India. n-butanol GR 80°C and petroleum ether AR 40-60°C were procured from Loba Chemie Pvt. Ltd., Mumbai, India. All other chemicals and reagents were procured from authorized dealer.

Collection of plant materials, identification and size reduction:

The fruits of *G. arborea* were collected from local area of Koraput district (India) in the month of April and May 2016. The plant was identified and authenticated by the Biju Patnaik Medicinal Plants Garden and Research Centre, Dr. M.S. Swami Nathan Research Foundation, Jeypore, Koraput (District), Orissa (Letter no. MJ/ DBT

(16)/ 1067, dated 11.09.2016). The fruits were shade dried under normal environmental condition. The dried fruits were pulverized to form coarse powder by using electrical grinder and stored in a closed air tight container for further use.



Antifungal activity of ethanol extract of *G. arborea* against *A. niger*.



Antifungal activity of ethyl acetate extract against *A. niger*.



Antifungal activity of n-butanol extract of *G. arborea* against *A. niger*.

Fig 1. Antifungal activity of *G. arborea* extracts against *A. niger*.

Preparation of extract:

The coarse powder form of dried fruits was extracted by Soxhlation method by using ethanol, ethyl acetate, n-butanol and petroleum ether as solvents. In this extraction process, a total amount of 1500 g coarse powdered fruits were extracted with 1200 ml of each solvent. For each solvent, 10 cycles were run to obtained thick slurry. The thick slurry was then concentrated under reduced pressure to obtained crude extract. All crude extracts were kept in closed air tight container under cool and dark place for further study.

EXPERIMENTAL:**Testing Organisms:**

The test organisms include one yeast and one fungus viz; *Candida albicans* ATCC3417 and *Aspergillus niger* ATCC6275. These two strains were obtained from National Chemical Laboratory (NCL), Pune, India.

Antifungal Activity:

The antifungal activity of the extracts was determined by disc diffusion method [8-10] using the test samples, reference and blank discs. The reference standard (Clotrimazole) and the extracts (1000 µg/ml) were dissolved in DMSO (Di-methyl sulfoxide). Clotrimazole (25 µg/ml) was used as a reference standard drug. Solvent control (only DMSO) was also maintained throughout the experiment. The selected micro-organisms are *Aspergillus niger* and *Candida albicans*.



Antifungal activity of ethanol extract against *C. albicans*.



Antifungal activity of ethyl acetate extract against *C. albicans*.



Antifungal activity of n-butanol extract against *C. albicans*.

Fig 2. Antifungal activity of *G. arborea* extracts against *C. albicans*.

Methodology:

Each extract was re-dissolved in DMSO to make a 1000 µg/ml solution and then filtered. From this solution, 80 µl aliquots were transferred onto blank paper disks (6 mm diameter) and dried. The dried disks were placed onto Mueller Hinton agar medium (Merck) previously inoculated with a fungal suspension and incubated at 28±1 °C for 48 h. Plates were then examined for the presence of growth inhibition zones, and their diameters were measured. Clotrimazole (25 µg/ml) was used as positive control. A disk loaded with 80 µl DMSO served as the negative control.

RESULT AND DISCUSSIONS:

The antifungal activity of *G. arborea* fruits extracts is shown in Fig 1 and 2. Effect of four different extracts at 100 mg/ml was tested against *A. niger* and *C. albicans* but no activity was observed. Hence, it is concluded that the plant *G. arborea* fruits extracts did not possess any antifungal activity.

CONCLUSION:

It could be concluded from the above study that the fruits extract of *G. arborea* Roxb. does not possess any antifungal activity.

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