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Phytochemical And Anti-Fungal Potential of Aqueous Ethanolic Leaves of *Tripidium bengalense*

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ABSTRACT:
The current study focused on the phytochemical and antifungal properties of Tripidium bengalense's aqueous ethanolic leaves. The Tripidium benghalense leaves came from the UP-West area. They were recognized and verified by Dr. Alok Srivastava, a botanist (ref. number. RU/PS/PI/2-25-32). The leaves were cleaned to remove dust and allowed to dry at room temperature in the shade. The dried leaves were subsequently ground into coarse and fine powders. For fifteen days, the powder was weighed and slowly stirred while soaking in ethanol and distilled water separately. A rotating evaporator was used to dry the mixed slurry that was produced. Extraction of the leaves of *Tripidium benghalense* plant through cold maceration process using aqueous and ethanol solvent, separately. Preliminary phytochemical screening was performed. Evaluation of anti-fungal activity through well diffusion method against following microbial strains i.e., *Aspergillus niger, Penicillium notatum, Candida albicans, Rhizopus species.* ethanolic leaves of *Saccharum benghalense* exhibited much significant zone inhibition as 5.83 mm, 5.11 mm, 5.62 mm, and 6.79 mm, respectively. It can be concluded that the anti-fungal activity was much significant against *Rhizopus sp.* Its anti-fungal potential might be due to the destruction of cell wall and/ nucleic acid of fungal strains. In conclusion, aqueous and ethanolic leaves extract of *Tripidium benghalense* exhibited a significant anti-fungal potential through the destruction of *A. niger*, *P. notatum, C. albicans* and *Rhizopus species*. It could stop the growth and proliferation of microorganism which are deadly to us.

KEYWORDS: *Tripidium bengalense*, anti-fungal activity, phytochemical screening, zone inhibition.

INTRODUCTION
There are fungi and bacteria everywhere. They are essential to maintaining our living environment. Worldwide, just a small percentage of micro-organisms cause disease and infection [1]. There are many ways that fungal infections can spread. For an organism to spread, it must survive and find a host that is vulnerable to it. A lot of fungi have learned how to live in many places, such food, soil, and water [2]. Some resistance genes can move from one type of bacteria to another because they are carried on plasmids, which are circular pieces of DNA that are not part of a chromosome and can copy themselves [3]. The use of anti-fungal medicines for viral illnesses, infections that go away on their own without treatment and have little chance of serious side effects [4].

Plant Profile
Powders or solutions made from different parts of plants treat many diseases. Inorganic minerals were discovered in the stem, blossoms, and surrounding soils since Saccharum munja is a medicinal plant [5].
Tripidium bengalense, popularly known as munj grass, is synonymous with Saccharum benghalense. It grows along riverbanks and in desert areas. The tall grass's panicles are smooth and greenish brown in hue. The grass is overgrown and reaches a height of seven feet. These are longer than the typical internode, the straight, pale straw-colored leaf sheaths are villous at the apex. Sometimes the tallest sheath reaches beyond the base of the panicle [6]. It has aesthetic significance because of its white flowers.

Taxonomical classification

Kingdom - Plantae
Class - Liliopsida
Order - Poales
Family - Poaceae
Genus - *Tripidium*
Species - *benghalense*

Screening of phytoconstituents revealed Saccharum benghalense to be quite present with many moieties. Abundant were alkaloids, terpenoids, flavonoids, phenols, coumarins and betacyanin. While cardiac glycosides, tannins, steroids were acquired in moderate quantities. Glycosides, saponins, and anthocyanin were missing [7].

MATERIALS AND METHODS

Experimental requirements

Fresh leaves of *Tripidium benghalense*, Fungus strains i.e., *Aspergillus niger, Penicillium notatum, Candida albicans* & *Rhizopus species*, distilled water, rotatory evaporator, weighing machine and ethanol.

Collection, Identification, and extraction of plant

Tripidium benghalense leaves came from the UP-West area. They were recognized and verified by Dr. Alok Srivastava, a botanist (ref. number. RU/PS/PI/2-25-32). The leaves were cleaned to remove dust and allowed to dry at room temperature in the shade. The dried leaves were subsequently ground into coarse and fine powders. For fifteen days, the powder was weighed and slowly stirred while soaking in ethanol and distilled water separately. A rotating evaporator was used to dry the mixed slurry that was produced. The % yield of the extract of the *Tripidium benghalense* leaves was calculated [8] using below mentioned formula-

$$\text{percent yield} = \frac{\text{actual yield}}{\text{theoretical yield}} \times 100\%$$

Preliminary phytochemicals determination [9][10]

Estimation of alkaloids

Before being filtered, each extract was individually dissolved in diluted HCl.

Mayer's Test: The filtrates were treated with Mayer's reagent, also known as potassium mercuric iodide. When a yellow-colored precipitate appears, alkaloids are present.

Wagner's Test: The filtrates were treated with Wagner's reagent, which is potassium iodide mixed with iodine. When a brown or reddish precipitate appears, alkaloids are present.

Hager's Test: The filtrates underwent treatment with Hagers Reagent. Alkaloids may be present if yellow precipitation starts to develop.

Estimation of glycosides

Fehling's test: Fehling's solutions A & B were heated for one minute using distilled water dilution. This clear blue solution had eight drops of plant extract added to it. After that, it was mixed with one milliliter of Fehling's solution and cooked in a water bath for five minutes. The presence of glycosides is indicated by brick-red precipitation.

Estimation of saponins

Foam test: About 2g of the plant extract was mixed with 10ml of distilled water and agitated briskly to create a stable, long-lasting foam. The presence of froth is indicative of saponins.

Estimation of tannins

Ferric chloride test: A test tube containing 0.5 grams of the powdered material that had been dried was boiled in 20 milliliters of water and then filtered. It was determined if the coloration was blue-black or brownish green-black by adding a few drops of 0.1% FeCl3.

Lead acetate test: Two milliliters of plant extract and two milliliters of distilled water were combined. Shake well after adding 0.01g of lead acetate to the liquid. When precipitate and white turbidity appear, tannins are present.

Estimation of flavonoids

NaOH test: A small amount of extract was treated with aqueous NaOH and HCl, and this resulted in the development of a yellow-orange hue.

H_2SO_4 test: A fraction of the extract was treated with Conc.H2SO4, and the creation of orange color was observed.

Estimation of terpenoids

Five milliliters of the aqueous plant extract and two milliliters of chloroform were combined, allowed to evaporate on the water route, and then heated to a boil with three milliliters of concentrated H2SO4. As terpenoids took shape, a grey coloring appeared.

Estimation of steroids

Two milliliters of chloroform and five milliliters of aqueous plant crude extract were mixed with concentrated H2SO4. The lower chloroform layer began to become red, indicating the presence of steroids.

Estimation of carbohydrates

Molisch's test

To 2-3 ml of extract of each solvent, add a few drops of α-naphthol solution in alcohol, stir, and then add concentrate H2SO4 from the edges of the test tube. a ring of violet where two liquids meet.

Fehling's test

It is used to identify sugars that are declining. Solution A: Dissolve 34.66 grams of copper sulfate in 500 milliliters of purified water. To make up to 50 milliliters of distilled water, dissolve 50 grams of sodium hydroxide and 17.3 grams of potassium sodium tartrate (Solution B). Mix two solutions in the same volume before using. Boil Fehling's A and B solution in 1 mL of mixture for 1 minute. Add the test solution in equal portions. Put in a kettle of boiling water and heat for five to ten minutes. Brick red flashed, and was followed by yellow.

Evaluation of anti-fungal potential

- **Well diffusion method**

By using the well diffusion method, the anti-fungal activity of *Tripidium bengalense* extract against microbes was identified. Nutrient Agar broth was utilized to cultivate a 24-hour-old culture of microbes, which was then used to make a suspension of microorganisms. After sterilization at 121°C (1.05 kg/cm2 pressure) for 20 minutes, nutrient agar solution was added. A sterile spreader was used to cover the whole surface of the agar plates when we inoculated them with 500 l of each fungal suspension. With a sterile cork borer, 5 mm wells were made in the solidified media, and each well was filled with microbes. The diameter (mm) of the inhibitory zone surrounding the well was measured after 24 hours of incubation at 30°C. As a standard anti-microbial Gentamycin was utilized. Every anti-fungal activity was carried out in triplicate. Microbial strains i.e., *Aspergillus niger, Penicillium notatum, Candida albicans* and *Rhizopus species* were used for screening of anti-fungal potential [11].

RESULTS AND DISCUSSION

Percentage yield

Based on the practical yield, he aqueous and ethanolic leaves extract of *Tripidium benghalense* demonstrated the percentage yield of 43.68 % and 49.32 %, respectively.

Phytochemical screening

Tripidium benghalense exhibited a high level of presence of several moieties during the initial screening of phytoconstituents. There were high amounts of alkaloids, terpenoids, flavonoids, and cardiac glycosides. In contrast, substantial amounts of coumarins and phenol were found. Anthocyanins, glycosides, and saponins were not present. *Tripidium benghalense* showed the following phytoconstituents as below-

Table 1. Phytochemicals in *Tripidium benghalense* leaves extract

Phytoconstituents	**Leaves of *Tripidium benghalense***	
	Aqueous extract	**Ethanolic extract**
Alkaloids	++	+++
Tannins	+	+
Cardiac glycosides	++	+++
Glycosides	–	–
Saponins	–	–
Terpenoids	+++	+++
Steroids	+	+
Flavonoids	++	+++
Phenols	+	++
Coumarins	++	++
Anthocyanin	–	–
Betacyanin	+	++

Absent (-), Moderate (++), Abundance (+++)

Fig 1. Phytochemicals in *Tripidium benghalense* leaves extract

Evaluation of anti-fungal potential

- **Well diffusion method**

The anti-fungal activity was evaluated using different 4 strains of fungus i.e., *A. niger*, *P. notatum*, *C. albicans and Rhizopus species*. Zone inhibition (anti-fungal activity) was recorded as 5.12 mm, 4.26 mm, 5.19 mm, and 6.24 mm in *A. niger*, *P. notatum*, *C. albicans* and *Rhizopus species*, respectively. Moreover, ethanolic leaves of *Saccharum benghalense* exhibited much significant zone inhibition as 5.83 mm, 5.11 mm, 5.62 mm, and 6.79 mm, respectively. It can be concluded that the anti-fungal activity was much significant against *Rhizopus sp.* Its anti-fungal potential might be due to the destruction of cell wall and/ nucleic acid of fungal strains.
Gentamycin (std. anti-fungal) exhibited highest inhibition zone ranging from 15-27 mm. Against *Rhizopus sp.* it exhibited highest zone inhibition as 27.0.

Table 2. Anti-fungal activity of *Saccharum benghalense*

Treatment	Zone inhibition (mm) of *Saccharum benghalense*			
	A. niger	*P. notatum*	*C. albicans*	*Rhizopus sp.*
Aqueous leaves of *Saccharum benghalense*	5.12	4.26	5.19	6.24
Ethanolic leaves of *Saccharum benghalense*	5.83	5.11	5.62	6.79
Gentamycin	15.0	12.0	21.0	27.0

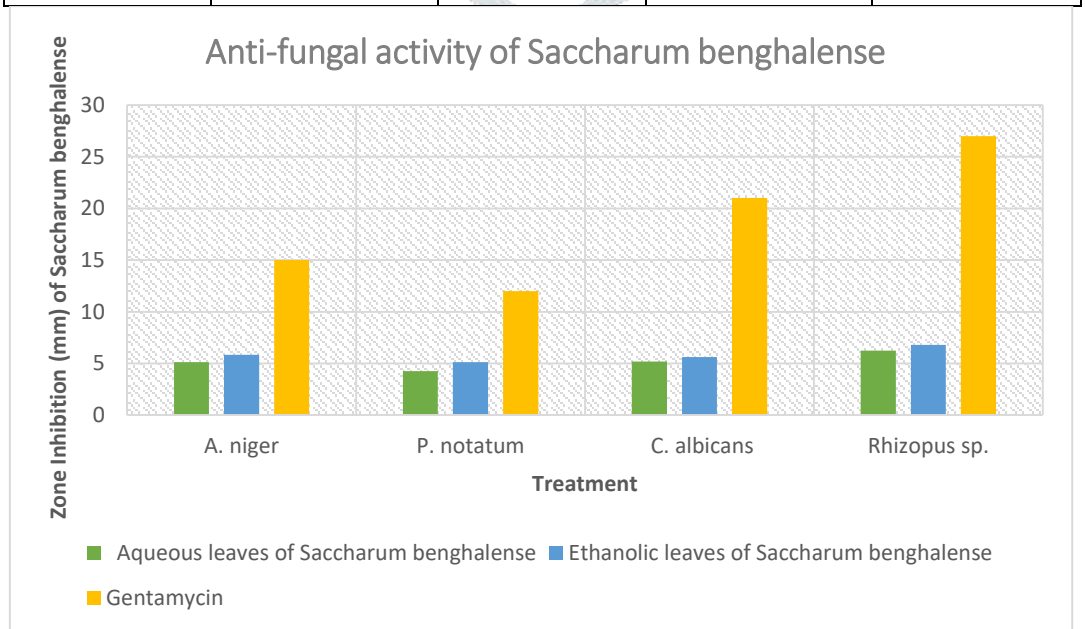

Fig 2. Graphical data of anti-fungal activity of *Saccharum benghalense*

Potential possibilities for the search for biologically significant substances include plants, and the findings of our investigation demonstrate the pharmacological characteristics of L. cylindrica. L. cylindrica leaf extracts in ethanol and ethyl acetate shown antibacterial, anti-inflammatory, and antioxidant properties [12]. Two flavonoid chemicals, luteolin and apigenin, are present and are essential parts of the L. cylindrica leaf extract. Both substances are known to have antibacterial, anti-inflammatory, and antioxidant properties [13] and may have made a significant contribution to the pharmacological effects observed in the plant extracts. Throughout the plant kingdom, flavonoids are extensively distributed and frequently found in fruits, vegetables, and some drinks. The plant flavones luteolin (3',4',5,7-tetrahydroxyflavone) and apigenin (4',5,7-trihydroxyflavone) are found in large quantities in a variety of fruits, vegetables, and medicinal plants. It has been discovered that these physiologically necessary flavonoids can be effective as medications [14].

For thousands of years, medicinal plants have been a significant source of pharmaceuticals. Many of the medications utilized in 21st-century medicine were created from vegetable sources. Plants have long been utilized for their antibacterial, anti-inflammatory, and antioxidant properties. The process of finding, evaluating, and incorporating novel compounds into existing treatment regimens is exceedingly difficult, costly, and time-consuming. The study of plant extracts is frequently the focus of interest since it is widely known that, in certain circumstances, the synergism of molecular structures causes the phytocomplex's activity to surpass that of pure phytochemicals [15].

The antioxidant qualities of the essential oil extracted from Cinnamommum malabatrum, along with additional pharmacological effects like enzyme inhibition and antibacterial activity against various Gram-positive and Gram-negative bacteria, were assessed by Aswathi Moothakoottil Kuttithodi and his associates [16].

Razvan Lucian Coseriu and his team looked into the effects of 16 common essential oils on clinical isolates of multidrug-resistant (MDR) Pseudomonas aeruginosa and the impact of the most powerful one, cinnamon essential oil, on the expression of the mex efflux pump gene. Even at very low concentrations, they discovered that cinnamon essential oil exhibited the strongest antibacterial effect. In contrast, the antibacterial activity of essential oils of thyme, turmeric, peppermint, basil, clove, and lavender showed varying findings. According to the team's findings, cinnamon essential oil may be employed as an adjuvant in the antibacterial treatment of infections caused by Pseudomonas aeruginosa strains that are resistant to multiple drugs [17][18].

Development of aqueous and ethanolic leaves extract of *Tripidium benghalense* exhibited a statistically significant anti-fungal potential by killing the different fungal strains. It can counter the skin problems specially dermatitis, pruritic and other epidermal ailments. The *Tripidium benghalense* would have a significant impact against microbial infections without affecting the patient health.

CONCLUSION

In conclusion, aqueous and ethanolic leaves extract of *Tripidium benghalense* exhibited a significant anti-fungal potential through the destruction of *A. niger*, *P. notatum*, *C. albicans* and *Rhizopus species*. It could stop the growth and proliferation of microorganism which are deadly to us. As they are obtained from the plant source mainly, so it will become an easiest way to avail widely with minimum risk of adverse effects.

To identify the chemical makeup of the bioactive substances causing the observed antifungal action, more research is required.

Fungicides made from natural plants may contain novel active substances with antifungal properties.

REFERENCES

1. Bannister B.A., Begg N.T., Gillespie S.H., editors. Structure and classification of pathogens. 2nd edn. Blackwell Science Ltd; Oxford, UK: 1996. pp. 23–34. (Infectious Disease). ch. 2.
2. Engelkirk P.G., Burton G.R., editors. Epidemiology and public health. 8th edn. Lippincott Williams and Wilkins; Baltimore: 2006. (Burton's Microbiology for the Health Sciences). ch. 11.
3. Crawford F, Hollis S. Topical treatments for fungal infections of the skin and nails of the foot. Cochrane Database Syst Rev. 2007; 3: 1-118.
4. Rotta I, Ziegelmann PK, Otuki MF, Riveros BS, Bernardo NL, Correr CJ. Efficacy of topical antifungals in the treatment of dermatophytosis: a mixed-treatment comparison meta-analysis involving 14 treatments. JAMA Dermatol. 2013; 149(3): 341–349.
5. Tirmizi S. A., F. H. Wattoo, M. H. S. Wattoo, S. Kanwal and J. Iqbal. Inorganic nutrients of Saccharum bengalense, Jour. Chem. Soc.Pak.Vol.27, No.2, 186-189; 2005.
6. Rahar Sandeep, Nagpal Navneet, Swami Gaurav, Arora Manisha, Bansal Suraj, Goyal Sandeep, Singla Shwali, Singh Preeti, Kapoor Reni. Medicinal Aspects of Saccharum munja. Research J. Pharm. and Tech. 3 (3): July-Sept. 2010; Page 636-639.
7. Kabata A. Pendias and H. Pendias, Trace Elements in Soils and Plants. CRC Press, Inc. Florida (1986).
8. Khan Mohammad Asif, S B Tiwari, H Gupta, Huma Noor. Evaluation of anxiolytic and antidepressant potential of hydro-alcoholic leaves extract of Azadirachta indica in albino rats. PharmacologyOnline, 2020; 3: 207-213.
9. Khandelwal K R. (2002). Practical Pharnacognosy, Nirali Prakashan, Pune, 9th Edition, 157-158.
10. Bhatt J. S. and S. Dhyani, (2012). Preliminary Phytochemical Screening of Ailanthus Excelsa Roxb., Int. J. Curr. Pharmaceut. Res., 4(1), 87-89.
11. Mounyr Balouiri, Moulay Sadiki, Saad Koraichi Ibnsouda. Methods for in vitro evaluating antimicrobial activity: A review. J Pharm Anal.. 2015 Dec 2;6(2):71–79.
12. Kaushik K, Sharma RB, Sharma A, Agarwal S. Evaluation of antifungal activity of crude methanolic extract of leaves of Ipomoea carnea and its formulation as a gel. J Res Pharm. 2020; 24(3): 368-379.
13. Lopez-Lazaro M. Distribution and biological activities of the flavonoid lute-olin. Mini Rev Med Chem 2009;9(1):31-59.
14. Liu R, Zhang H, Yuan M, Zhou J, Tu Q, Liu JJ, et al. Synthesis and biolog-ical evaluation of apigenin derivatives as antibacterial and antiproliferative agents. Molecules 2013;18:11496-511.
15. Ramesh M, Rao YN, Rao AV, Prabhakar MC, Rao CS, Muralidhar N, et al.Antinociceptive and anti-inflammatory activity of a flavonoid isolated from Caralluma attenuata. J Ethnopharmacol 1998;62(1):63-6.
16. Bora, L.; Avram, S.; Pavel, I.Z.; Muntean, D.; Liga, S.; Buda, V.; Gurgus, D.; Danciu, C. An Up-To-Date Review Regarding Cutaneous Benefits of Origanum vulgare L. Essential Oil. Antibiotics 2022, 11, 549.

17. Coșeriu, R.L.; Vintilă, C.; Pribac, M.; Mare, A.D.; Ciurea, C.N.; Togănel, R.O.; Cighir, A.; Simion, A.; Man, A. Antibacterial Effect of 16 Essential Oils and Modulation of mex Efflux Pumps Gene Expression on Multidrug-Resistant Pseudomonas aeruginosa Clinical Isolates: Is Cinnamon a Good Fighter? Antibiotics 2023, 12, 163.
18. Awadelkareem, A.M.; Al-Shammari, E.; Elkhalifa, A.O.; Adnan, M.; Siddiqui, A.J.; Patel, M.; Khan, M.I.; Mehmood, K.; Ashfaq, F.; Badraoui, R.; et al. Biosynthesized Silver Nanoparticles from Eruca sativa Miller Leaf Extract Exhibits Antibacterial, Antioxidant, Anti-Quorum-Sensing, Antibiofilm, and Anti-Metastatic Activities. Antibiotics 2022, 11, 853.