

Antifungal potential of some medicinal plants on selected pathogenic fungi

Abstract

Introduction: Medicinal plants represent a rich source of antimicrobial agents. Plants are used medicinally in different countries and they are source of many potent and powerful drugs. A wide range of medicinal plant parts is used for extract as raw drugs.

Objective: The antifungal activity of different medicinal plant Methanol extracts for three different plant was selected for this studies. These plants were *Leptadenia hastata*, *Barringtonia racemosa* and *Barringtonia asiatica*. The aim of this test is to find out the efficacy of the plants against some selected pathogen. this may help in the discovery of new chemical classes of antifungal and also serve as selective probiotic agents, when administered in adequate amounts, confer a health benefit on the host.

Material and methods: *Leptadenia hastata*, *Barringtonia asiatica* and *Barringtonia racemosa* extracts were tested for potential antifungal properties. The roots were extracted with methanol solvent and then vaporized to give respective extracts. The antimicrobial activity against *Aspergillus niger*, *Aspergillus flavin*, *Candida tropicalis* and *Fusarium oxysporium* (fungi) was determined by agar disk method. The optical densities of the broth using UV mini spectrophotometer and zone of inhibition by the crude extracts were determined.

Results: the results showed the efficacy of the plants extracts at different concentration with varying data, higher rate of inhibition was observed in all the extract against all the fungi species. However, extract from *Leptadenia hastata* showed more significant inhibition with diameter rate with 3.38 ± 0.08 to 5.00 ± 0.09 days five for 25ppm to 250ppm, while *Barringtonia racemosa* and *Barringtonia asiatica* lesser compared to *Leptadenia hastata* as against the pathogen at the same concentration and same time frame.

Conclusion: It is showed that all plants extract was found maximally affected against the activity of *Aspergillus niger*, *Aspergillus Flavin*, *Candida tropicalis* and *Fusarium oxysporium* and aggressively inhibits the growth of species in various plant extracts. This may help in the discovery of new chemical classes of antifungal as well as antibiotic that could serve as selective agents for the maintenance of human health and may provide biochemical tools for the study of fungal infectious diseases.

Keywords: fungi, pathogenic, *leptadenia hastata*, *barringtonia racemosa*, *barringtonia asiatica*

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Introduction

Medicinal plants represent a rich source of antimicrobial agents. Plants are used medicinally in different countries and they are source of many potent and powerful drugs. A wide range of medicinal plant parts is used for extract as raw drugs. They possess varied medicinal properties, while some of these raw drugs are collected in small quantities by the local communities and traditional healers for local use.¹ The plants represent a rich source of antimicrobial agents.^{2,3} Many of the plant materials used in traditional medicine are readily available in rural areas at relatively cheaper than modern medicine.⁴⁻⁶ The activity of plant extracts on bacteria and fungi has been studied by a very large number of researchers in different parts of the world.⁷⁻⁹ The plant metabolites and plant based pesticides appear to be one of the better alternatives as they are known to have minimal environmental impact and danger to consumers in contrast to the synthetic drugs.^{10,11} *Leptadenia hastata* (Pers) Decne (Family-Asclepiadaceae), commonly known as hastata is edible non-domesticated vegetable and it is collected in wild throughout Africa. It is a voluble herb with creeping latex stems, glabrescent leaves, glomerulus and racemus

flowers as well as follicle fruits. It is typically grown in tropical dry lands in sandy soil. Wild foods like *Leptadenia hastata* provide food security during seasonal changes and are used medicinally in many areas. The breeders commonly used the leaf and stems for their parasitic activity and against placental retention.¹² This plant can be used as a probiotic supplements and foods to lower risk of infections, improved digestion and even a reduced risk for some chronic diseases as claimed by traditional healer.¹² However, *Barringtonia racemosa* are mangrove plants. This specie which is also known as putat, fish poison tree or powder puff tree is a type of highly valuable plant species due to its medicinal values. They are geographically found to be widely distributed from eastern Africa and Madagascar to Micronesian, Asia and Polynesian Island, the species was said to have been associated very well in various tribes around the world with diverse ethno-botanical uses. The therapeutic potential of this herb is as a result of the presence of diverse bioactive compounds such as Lupeol, Germanicol, Teraxanol Barringtogenic, Barringtogenol etc. However, most of the studies on *Barringtonia racemosa* are restricted to crude extracts, and many biologically active compounds are yet to be identified in order to claim the traditional uses of this

plant as acclaimed by the herbal and traditional practitioners.¹³ While *Barringtonia asiatica* is one among the species belonging to the family *Barringtonia*, it is a native to mangrove habitats on the tropical, it is a common plant in the Malaysian Mangroves and wetlands such as the Kuching wetlands Sarawak and Bako National Park, it is also found in tropical Africa, Nigeria and Madagascar.¹⁴

Material and method

Plant preparation

Freshly harvested leaves of *Leptadenia hastata*, *Barringtonia racemosa* and *Barringtonia asiatica* were used for the preparation of the crude extract. *Leptadenia hastata* was collected from an uncultivated farm in Nigeria, while the other two species were collected in Sarawak Malaysia. This three important medicinal plants were selected based on the traditional claim of their efficacy against some antimicrobial and antifungal activity. The plants were collected and washed with distilled water. These plants leaves were dried and their different parts were grinded to form powder extract. Screening bioassay preparation of plant extract was conducted in lab condition. For this purpose, 150mg of the powdered leaves were weighed into a beaker and mixed with distilled water three times the quantity of the sample and allow to stand for two days with continues shaking at time interval for 12hrs. The mixture was then filtered using Whatman filter paper No.4 and the solvent was then evaporated using a rotary evaporator (Heldolph Laborato 400). It was then stored under frozen condition for further use.

Reconstitution of the extracts

2g of each of the plants the extract was weighed and dissolved in 20ml of 50% Dimethyl sulfoxide (DMSO) to make a stock concentration of 100mg/ml from which the various concentrations used were calculated.

Microorganisms used

Isolates of *Aspergillus flavin*, *Aspergillus niger*, *Candida tropicalis*, and *Fasarium oxysporium* from four different Concentration were used. They were collected from the virology Laboratory Faculty of Resource science, Universiti Malaysia Sarawak.

Drug used

Fluconazole common name Diflucan was used as reference standard for antifungal studies obtained from Medical Resource Sarawak. The disc of 5mm diameter of different fungi 6days old culture were prepared and smeared of the Petri plates. Three replicates were kept for each treatment and incubated at 28±2°C. The radial growth of mycelium to ascertain the effect of the plants were measured every day for five to six days and compared with the results of control.

Result and discussion

Result

Result obtained from the studies is presented in Tables 1–3 this revealed that all the fungal species were affected by the administration of *Leptadenia hastata*, *Barringtonia racemosa* and *Barringtonia asiatica* in a dose dependent manner. The inhibition of the microorganism significantly increases at ($P < 0.05$) at 50ppm 100ppm, 500ppm and in some instances when compared with control.

Discussion

The result obtained as presented in Tables 1–3 shows that the

methanol root extract of the plant *Leptadenia hastata*, *Barringtonia racemosa* and *Barringtonia asiatica* were effective in significantly reducing the growth rate of some of the selected pathogen as compared to the control. However, all the roots extract showed antifungal activity against *Aspergillus niger*, *Aspergillus flavus*, *Candida tropicalis* and *Fasarium oxysporium* with maximum inhibition (mm) recorded in *Candida tropicalis*, *Fusarium oxysporium* and *Aspergillus flavus* (1.95±0.05mm, 1.80±0.06mm and 5.00±0.09mm) higher compared to the control in all concentration on each fungi on the same days. The antifungal activities of the methanol extract of *Leptadenia hastata* were observed in various concentrations with zones of inhibition diameters ranging from 1.23±0.15 mm to 3.75±0.05 mm. Among the six concentrations, 100ppm was the most active with 3.75±0.05mm when compared to the control rate activity on the pathogen. The extracts inhibited the growth of *Aspergillus niger* at all concentration except at 500ppm and 1000ppm which indicated less active as when compared with the control as well as the other concentration showed in Table 1. However, in Table 2 the methanol root extract of *Barringtonia racemosa* decreased the mycelial growth of all the pathogen with increasing of dose concentration. The highest result impact was observed in *Fusarium oxysporium*. the concentration of the extract at 1000ppm completely if not all inhibited the growth of *Fusarium oxysporium*. Concentration of the extract at 25 to 100ppm has the least effect on the mycelial growth of the fungi in general. This shows that the pathogen has been adversely affected by the increase in concentration of the methanol root extract. This difference in inhibition rate might be as a result diversity of the pathogens and also selectivity properties of chemical substance as well as dose concentration and mechanism of action of the component with time duration. Table 3 Indicated the antifungal activity of the extracts of *Barringtonia asiatica* from six different concentration of the methanol extracts. The result of the MIC studies revealed the anti-fungal activity of the extracts against the tested strain of the microorganism between concentration ranges of 25ppm, 50ppm, 100ppm, 250ppm 500ppm to 1000pp. The result of zone of inhibition study revealed that the extracts possess anti-fungal activity in concentration dependent manner against the test organism and was comparable with the standard drug.

Mean±SD yield of various extracts concentration confirm that 100ppm of the extracts has minimum yield with 0.85±0.05 at day one and maximum yield at 3.17±0.05 day five for the pathogen especially *Aspergillus niger*, were as *Aspergillus flavin* has maximum inhibition at 3.78±0.04. however, *Barringtonia racemosa* extract was found to be less active against *Aspergillus niger* but showed significant inhibition on *Candida tropicalis* and *Fusarium oxysporium* when compared with the *Leptadenia hastata* as well as the sister species *Barringtonia asiatica* at the same time and as well as concentration as shown in tables above. Though the extract exhibits its potency against all the pathogen. The *Barringtonia asiatica* methanol has higher effect on broad spectrum compared to *Barringtonia racemosa* because of its significant activity on all the pathogen at concentration 100, 250, and 500ppm thus the extracts impart significant antifungal activity against all test strains. This suggests that the plant parts of *Leptadenia hastata* and *Barringtonia a asiatica* have a broad spectrum of activity compared to *Barringtonia racemosa*, although the degree of susceptibility could differ between different organisms. The broad spectrum of antifungal activity found in this present study may be attributed to the presence of secondary metabolites of various chemical types present in the plant species. Our results indicate the potential usefulness of *Leptadenia hastata*, *Barringtonia asiatica* and *Barringtonia racemosa* in the treatment of various pathogenic diseases as it may help in the discovery of new chemical classes of antifungal as well as antibiotic that could serve as selective agents

for the maintenance of human health and may provide biochemical tools for the study of fungal infectious diseases. This current results demonstrate that selected medicinal plant extracts effective suppressed

the radial mycelia growth of all species at various concentration and these findings confirm the result.¹⁵⁻¹⁷

Table 1 Effect of methanol root extract of *Leptadenia hastata* on fungi

Organisms	Day	Concentration (ppm)						
		Control	25	50	100	250	500	1000
Aspergillus niger	1	0.80±0.00	0.68±0.04	0.78±0.04	0.85±0.05b	0.58±0.04	0.45±0.05	0.25±0.05
	2	1.05±0.07	0.90±0.06	1.02±0.12	1.18±0.07b	0.87±0.05	0.47±0.05	0.37±0.05
	3	2.45±0.07	2.45±0.05	2.28±0.08	2.07±0.05	1.65±0.05	1.13±0.10	0.65±0.05
	4	3.20±0.28	3.00±0.09	2.95±0.05	2.73±0.08	2.05±0.05	1.73±0.08	1.02±0.08
	5	3.65±0.07	3.38±0.08a	3.27±0.42a	3.75±0.05ab	2.62±0.08a	2.15±0.08a	1.23±0.15a
Aspergillus flavin	1	0.95±0.07	0.80±0.06	0.80±0.06	0.95±0.05	0.80±0.00	0.67±0.52	0.47±0.52
	2	1.45±0.07	1.30±0.06	1.43±0.05	1.55±0.05b	1.18±0.10	0.95±0.05	0.73±0.05
	3	3.25±0.07	3.17±0.05	3.05±0.05	2.80±0.00	2.68±0.12	1.51±0.05d	0.83±0.12
	4	3.55±0.14	3.60±0.28a	3.78±0.04ab	3.75±0.08	2.37±0.21	2.08±0.08d	1.15±0.12d
	5	3.45±0.07	3.58±0.08	3.47±0.05	3.97±0.08ab	3.19±0.04a	2.87±0.05ad	1.43±0.05a
Candida tropicalis	1	1.80±0.00	1.68±0.04	1.83±0.05b	1.57±0.05	1.35±0.05	1.12±0.08	0.88±0.08
	2	2.35±0.07	2.05±0.05	2.30±0.09	2.47±0.05b	1.98±0.08	1.85±0.05	0.98±0.08d
	3	2.25±0.00	2.42±0.08	2.48±0.04	2.67±0.05	2.85±0.05abd	1.27±0.15	1.02±0.08
	4	2.80±0.00	2.63±0.05	2.78±0.04	2.93±0.05b	1.93±0.08	1.62±0.04	0.95±0.05
	5	3.10±0.14	2.98±0.08a	3.18±0.04a	3.33±0.05ab	2.68±0.08	1.75±0.08a	1.20±0.06a
Fusarium oxysporium	1	2.60±0.00	2.37±0.05d	2.50±0.89d	2.67±0.05bd	1.93±0.05d	1.50±0.06d	1.02±0.08d
	2	2.65±0.07	2.62±0.04d	2.77±0.05d	3.00±0.06bd	2.40±0.06d	1.92±0.08d	0.95±0.08
	3	3.50±0.14	3.18±0.12d	3.40±0.00d	3.60±0.06bd	2.22±0.21	1.30±0.11	1.17±0.08d
	4	4.00±0.00	3.73±0.05d	3.93±0.08d	4.13±0.08bd	2.67±0.05d	1.88±0.08	0.97±0.05
	5	4.70±0.14	3.90±0.06ad	4.22±0.08ad	4.75±0.05ad	5.00±0.09abd	2.78±0.39a	1.57±0.05ad

Values are Mean±SD for six determinations

^aSignificantly (p<0.05) higher compared to different treatment days at the same concentration on each fungus

^bSignificantly (p<0.05) higher compared to the control on each fungus in each row

^cSignificantly (p<0.05) higher compared to other fungi on the same day on each concentration

Table 2 Effect of methanol root extract of *Barringtonia racemosa* on fungi

Organisms	Day	Concentration (ppm)						
		Control	25	50	100	250	500	1000
Aspergillus niger	1	0.47±0.05	0.42±0.08	0.57±0.10b	0.63±0.14b	0.73±0.08b	0.93±0.08b	1.03±0.08b
	2	0.63±0.08	0.55±0.05	0.67±0.05b	0.82±0.10b	0.93±0.08b	1.02±0.08b	1.17±0.05b
	3	0.87±0.05	0.68±0.04	0.87±0.05	0.95±0.05b	1.02±0.08b	1.20±0.06b	1.28±0.04b
	4	0.93±0.05	0.72±0.08	0.83±0.05	1.02±0.04b	1.13±0.05b	1.27±0.05b	1.35±0.05b
	5	0.93±0.05	0.77±0.05	0.88±0.04	1.10±0.06b	1.18±0.04b	1.33±0.05b	1.45±0.05ab
	6	1.00±0.09a	0.92±0.08a	1.00±0.06a	1.15±0.08ab	1.22±0.08ab	1.35±0.05ab	1.45±0.05ab
Aspergillus flavin	1	0.63±0.08	0.50±0.06	0.55±0.05	0.60±0.06	0.73±0.08b	0.87 ± 0.05b	0.98±0.08b
	2	0.77±0.08	0.63±0.08	0.82±0.08b	0.92±0.08b	1.07±0.05b	1.15±0.05b	1.25±0.05b
	3	1.02±0.08	0.73±0.08	0.88±0.08	1.07±0.05b	1.17±0.05b	1.27±0.05b	1.35±0.05b
	4	1.07±0.05	0.78±0.08	0.87±0.05	1.12±0.10b	1.20±0.13b	1.28±0.12b	1.38±0.12b
	5	1.02±0.08	0.88±0.04	1.05±0.05b	1.20±0.06b	1.33±0.08b	1.40±0.06b	1.52±0.08ab
	6	1.27±0.05a	1.02±0.08a	1.20±0.06a	1.28±0.08a	1.35±0.05ab	1.42±0.10ab	1.52±0.10ab
Candida tropicalis	1	1.33±0.08ac	0.80±0.06c	0.93±0.05c	1.00±0.09c	1.13±0.08c	1.25±0.05c	1.32±0.08c
	2	1.20±0.06c	0.83±0.05c	0.93±0.05c	1.03±0.05	1.17±0.05c	1.28±0.08bc	1.37±0.05bc
	3	1.02±0.08	0.97±0.05c	1.05±0.05bc	1.12±0.13b	1.25±0.05bc	1.35±0.05b	1.45±0.05b
	4	0.87±0.05	0.98±0.04b	1.10±0.06b	1.23±0.05b	1.27±0.05b	1.37±0.05b	1.45±0.05b
	5	0.58±0.08	1.05±0.05b	1.17±0.05b	1.33±0.08b	1.42±0.08b	1.47±0.05b	1.55±0.05b
	6	0.70±0.05	1.07±0.05ab	1.23±0.05ab	1.38±0.08ab	1.47±0.05ab	1.55±0.05ab	1.68±0.04ab
Fusarium oxysporium	1	0.85±0.05	0.63±0.05	0.75±0.05	0.82±0.08	1.03±0.08b	1.10±0.09b	1.17±0.05b
	2	0.98±0.04	0.83±0.08c	0.93±0.05c	1.05±0.05bc	1.13±0.08b	1.20±0.06b	1.25±0.05b
	3	1.30±0.09c	0.87±0.05	1.03±0.08	1.22±0.08c	1.30±0.06	1.37±0.05bc	1.47±0.05bc
	4	1.35±0.08c	1.07±0.05c	1.27±0.05c	1.33±0.05c	1.42±0.04bc	1.47±0.05bc	1.53±0.05bc
	5	1.47±0.05c	1.20±0.06c	1.32±0.08c	1.42±0.08c	1.55±0.05bc	1.65±0.05bc	1.72±0.05bc
	6	1.60±0.09ac	1.25±0.05ac	1.37±0.05ac	1.45±0.05ac	1.58±0.08ac	1.68±0.04abc	1.80±0.06abc

Values are Mean±SD for six determinations

^aSignificantly (p<0.05) higher compared to different treatment days at the same concentration on each fungi

^bSignificantly (p<0.05) higher compared to the control on each fungus in each row

^cSignificantly (p<0.05) higher compared to other fungi on the same day on each concentration

Table 3 Effect of methanol root extracts of *Barringtonia asiatica* on fungi

Organisms	Day	Concentration (ppm)						
		Control	25	50	100	250	500	1000
<i>Aspergillus niger</i>	1	0.93±0.04	0.90±0.06d	0.97±0.10b	0.65±0.16	0.82±0.12d	0.52±0.12	0.45±0.19
	2	1.03±0.12	0.63±0.12	0.95±0.10d	0.58±0.10	1.05±0.08	0.90±0.06d	0.60±0.06d
	3	1.86±0.11	1.38±0.23d	1.42±0.31d	1.12±0.18d	1.82±0.15	1.43±0.14	0.92±0.08
	4	1.71±0.14	1.05±0.10d	1.20±0.14d	1.18±0.17d	1.32±0.50	1.40±0.24d	1.47±0.14d
	5	1.67±0.12	1.48±0.17ad	1.75±0.55ad	1.12±0.21ad	1.65±0.20ad	1.32±0.19ad	1.90±0.06ad
<i>Aspergillus flavin</i>	1	0.87±0.05	0.75±0.10	0.85±0.08	0.67±0.10	0.72±0.08	0.60±0.09	0.55±0.10
	2	0.83±0.08	0.75±0.08	0.85±0.08	0.82±0.08	1.08±0.08bd	0.83±0.05	0.60±0.13d
	3	1.75±0.05	1.12±0.45	1.30±0.13	1.95±0.10	1.85±0.10d	1.40±0.19	0.95±0.05
	4	1.88±0.08	1.02±0.13	1.73±0.37	1.88±0.08	1.45±0.24d	1.60±0.25	1.28±0.15
	5	1.46±0.05	1.25±0.27a	1.00±0.26a	1.02±0.46a	1.48±0.15a	1.97±0.23a	1.53±0.12a
<i>Candida tropicalis</i>	1	0.82±0.08	0.83±0.12	1.03±0.08bd	0.87±0.06d	0.72±0.12	0.52±0.13	0.53±0.15
	2	1.00±0.6	0.87±0.05d	0.90±0.06	0.85±0.08d	1.00±0.06	0.82±0.07	0.60±0.06d
	3	1.10±0.09	1.055±0.10	1.07±0.14	1.98±0.08	1.60±0.09	1.53±0.12	1.15±0.16d
	4	1.39±0.08	1.00±0.09	1.15±0.24	1.07±0.33	1.90±0.09	1.48±0.21	1.05±0.12
	5	1.80±0.08	1.97±0.05a	1.80±0.14a	1.28±0.16a	1.17±0.24a	1.82±0.26a	1.35±0.10a
<i>Fusarium oxysporium</i>	1	0.82±0.08	0.58±0.08	0.77±0.08	0.70±0.09	0.68±0.08	0.93±0.10bd	0.88±0.17bd
	2	0.85±0.05	0.62±0.10	0.63±0.05	0.70±0.06	0.98±0.08b	0.80±0.09	0.53±0.12
	3	1.97±0.08	1.68±0.08	1.97±0.08	2.00±0.06	1.62±0.08	1.57±0.12d	1.07±0.10
	4	1.99±0.08	1.57±0.05	1.08±0.15	1.92±0.08	1.77±0.12	1.18±0.23	0.93±0.14
	5	1.96±0.10	1.07±0.08a	1.15±0.19a	1.05±0.10a	1.97±0.16a	1.93±0.20a	1.10±0.17a

Values are Mean±SD for six determinations

^aSignificantly ($p < 0.05$) higher compared to different treatment days at the same concentration on each fungus

^bSignificantly ($p < 0.05$) higher compared to the control on each fungus in each row

^cSignificantly ($p < 0.05$) higher compared to other fungi on the same day on each concentration

Conclusion

This research work on the three medicinal plant extract as an antifungal show that the efficacy of plants as an antifungal cannot be underestimated, it is showed that all plants extract was found maximally affected against the activity of *Aspergillus niger*, *Aspergillus Flavin*, *Candida tropicalis* and *Fusarium oxysporium* and aggressively inhibited the growth of species in various plant extracts (Tables 1–3). Thus justify their use in folk medicine for the treatment of skin and other related infections, the plants can as well be used as a probiotic supplements and foods to lower risk of infections, improved digestion and even a reduced risk for some chronic diseases since the leaves can be consumed as vegetable because of its antifungal activities.

Acknowledgments

None.

Conflict of interest

Authors declare that there is no conflict of interest.

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