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Extraction and Biological Activity of *Barringtonia asiatica* Stem-Bark Extracts on some Selected Fungi, Bacteria's, Cytotoxicity and Antioxidant Potentials

Isaac John Umaru[1, 2*], Fasihuddin Badruddin Ahmed [1], Kerenhappuch Isaac Umaru[3] and Ajoke Omorinoye Omolayo[4]

[1]Faculty of Resource Science and Technology, University of Malaysia Sarawak, Kota Samarahan Malaysia

[2]Department of Biochemistry Federal University Wukari Taraba State.

[3]Department of biochemistry, Modibo Adama University of Technology Yola Adamawa state. Nigeria

[4]Department of Geology and Mineral Sciences, Faculty of Physical Sciences, University of Ilorin, Nigeria

*Corresponding Author E-mail: umaruisaac@gmail.com

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ABSTRACT

Natural products from medicinal plants crude extracts provide fortuity for a new agent of medication because of the increasing failure of synthesised drug against resistance diseases and ailment as while as increasing demand for chemical diversity in screening programs, as well as seeking for therapeutic cure from natural product. Medicinal plants product contains various types of bioactive compounds. The aim of this paper was on crude extract and to evaluate the antioxidant, antibacterial, antifungal and cytotoxicity of the crude extract of the stem-bark of Barringtonia asiatica. The stem-bark of Barringtonia asiatica produces significant and high biological activity of the crude extract (cytotoxicity 34.059 µg/mL, Antioxidant 63.4±0.11 µg/mL, antibacterial 20.80 ± 0.10 µg/mL and antifungal 17.33 ± 0.04 µg/mL) which qualify the use as an agent for an ailment and disease control.

Keywords: *Extraction, Isolation, Characterization, Bioactive, Antioxidant Antibacterial, antifungal, Cytotoxicity of Barringtonia asiatica Stem-bark*

INTRODUCTION

Natural products from medicinal plants, either as pure compounds or as a crude extracts provide fortuity for a new agent of medication because of the increasing failure of synthesised drug against resistance diseases and ailment as while as increasing demand for chemical diversity in screening programs, seeking for therapeutic cure from natural product (Umaru et al., 2019).

Numerous efforts by researchers have been directed towards the provision of empirical proof to back the use of tropical plants in traditional medicinal practice (Maiti et al., 2009; Dahiru et al., 2010). Focus on medicinal plant research has increased worldwide and evidence abounds in the immense potentials of medicinal plants used in various traditional systems.

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Various medicinal plants have been studied using different scientific approaches and results from these studies have revealed the potentials of medicinal plants in pharmacology (Wakawa & Hauwa, 2013). These medicinal plants are of great importance to the health of the individuals and communities to larger extend, and nutritional benefits are derived from these plants since they are commonly used as vegetables.

According to the World Health Organization (WHO), a good number of the world population, more than 80% relies on traditional medicine for their primary healthcare. The use of herbal medicines represents a long history of human interactions with the environment. From time immemorial Plants are used for traditional medicine because of the wide range of substances that can be used to treat endemic immedicable diseases and ailment (Isaac et al., 2018; Umaru et al., 2018)

The consequences of adverse effects and microbial resistance to the synthesized drugs, researches have now turned to explore the thousands of phytochemicals from plants as safe and broadly effective alternatives with less adverse effect. Thus plants hold the key to the discovery and development of new pharmaceutical and biological resources that will champion the course of health and well-being of human. (Duraipandiyan et al., 2006).

Therefore, *Barringtonia asiatica* stem-bark was used in this study to isolate chemical compounds and to test the biological activity of the extracts in different solvents of different polarity as while as to demonstrate the effectiveness of a bioactive compound and to verify this traditional claim. Thus clinical trials directed towards understanding the efficacy and safety of the isolates and crude extracts were considered.

This paper provides details biological activity of stem-bark crude extract of *Barringtonia asiatica* with common biological assay, using Brine shrimp, Cytotoxicity, Disc diffusion and DPPH.

Plant Material

Sample Collection

Fresh plant material was collected from the Meranek river bank kota-samarahan Sarawak Malaysia, the plant Barringtonia asiatica was authenticated by a Botanist in the Universiti Malaysia Sarawak, the identified and certified plant materials were given a voucher number as 15010238/IJU/2016. The stem-bark was carefully peeled and washed under running tap water. It was then air dried to be free of water and then spread in the laboratory and allow to dry at room temperature until they are fully dried. They are cut into smaller pieces, spread in the laboratory and allowed to dry at room temperature until they are fully dried.

Sample Preparation

Dried stem-bark was ground into fine powder form using laboratory pestle and mortar and electric grinder. The finely ground powdered samples was packed into clean, dry sample containers and labelled appropriately and kept for further use. Extraction was carried out by the conventional solvent extraction method described by Isaac et al. (2019). This was achieved by soaking the ground plant material in solvents in the order of increasing polarity. A total of 2 kg of the dried and ground powdered sample was extracted using cold soaking method with hexane. The sample were soaked in the hexane with the ratio of 1:3 (sample: hexane) in a 5 litres Erlenmeyer flasks at room temperature for 7 days. The resulting hexane solution was then filtered using Whatman filter paper No 4 and the residue was then re-extracted with fresh hexane for another 72 hrs and filtered. Both extracts were combined and evaporated to dryness with a rotary evaporator (Heidolph Laborota 4000 efficient) under reduced pressure below 50 ^{o}C to obtain the hexane crude extract. The residue was re-extracted using similar procedure with dichloromethane, followed by chloroform, ethyl acetate and methanol to obtain respective crude extracts. The dry weight and percentage yield of each crude extract were determined.

Brine shrimp (*Artemia salina*) Lethality Test

The LC_{50} of the plant extracts was determined using brine shrimp lethality test. The test was

conducted using larvae of *Artemia salina* based on method developed by McLaughlin et al. (1998) as reported by Wakawa et al. (2017). One spatula full of brine eggs was placed into a 250 mL beaker containing 150 mL of sea water placed under light environment. A source of oxygen supply was connected to the beaker using water pump at reduced pressure and allowed for 72 hrs to hatch. The brine shrimp (nauplii) were then used for the test. Exactly 4 mg of each extract was dissolved in 200 µL of DMSO (RCI Labscan limited) and a lower series of chosen concentration was prepared by serial dilution with DMSO, where thymol was treated as the control. The assay system was prepared with 5 mL of filtered seawater containing chosen concentration of extract and 1% yeast extract (for feeding) in a pre-marked 6-well microplate and 10 brine shrimps were carefully taken with micropipette and introduced into each microplate. This was done in triplicates making a total of 30 brine shrimps per concentration. Filtered seawater was added to DMSO and 10 brine shrimps in triplicates and this was used as the control groups. If the brine shrimp in these microplates shows a rapid mortality rate, then the test is considered invalid as the nauplii might have died due to some reasons other than the cytotoxicity of the extracts. The setup was allowed to remain for 24 hrs under constant illumination of fluorescent and number of survived nauplii were counted with a hand lens. Based on the data obtained, the average death of the brine shrimp at different concentrations of the extract and the LC_{50} of the extract was calculated using probit regression by statistical software SPSS 22 and the result was expressed as mean + SD at the 95% level of confidence ($p < 0.05$).

Antioxidant (DPPH (2, 2-diphenyl-1-picryl-hydrazyl) Free Radical Scavenging Assay)

The free radical scavenging assay of compound 2, 2-diphenyl-1-pycryl-hydrazyl (DPPH) was used to evaluate the antioxidant properties of the crude extract. The measurement was based on the method described by Wang et al. (2008). The sample was prepared by diluting 6 mg of crude extract into 6 mL of methanol, producing a concentration of 1000 µg/mL. The stock solution was sonicated to ensure the homogeneity of the sample. Three other concentrations were prepared at 10, 50 and 100 µg/mL, diluted from the 1000 µg/mL stock solution. Sample of 5000 µg/mL was prepared separately by diluting 25 mg of crude extract into 5 mL of methanol.

Approximately 3 mL of 0.1 mM solution of 2,2-diphenyl-1-pycrylhydrazyl (DPPH) in methanol was each added into five series of prepared concentrations (10, 50, 100, 1000 and 5000 µg/mL) of sample solutions (1 mL). Analysis was done in triplicate. The solution was mixed vigorously and left to stand at room temperature for 30 minutes in the dark after which its absorbance was measured spectrophotometrically at 517 nm using Jasco ultra violet spectrophotometer model V-630. Methanol was used as blank (only methanol) and negative control (1 mL methanol mixed with 3 mL DPPH), while ascorbic acid (vitamin C) as the standard. The concentration of the sample required to inhibit 50% of the DPPH free radical was calculated as IC_{50} and the value was determined using Log dose inhibition curve which performed by using PRISM version 3.02 software, based on the calculated values of the DPPH scavenging activity (%) of the sample (Tailor & Goyal, 2014).

Antibacterial Assay

Bacterial strains are *Staphylococcus aureus*, (Gram +ve), *Exiguobacterium aquaticum* (Gram-ve) and *Escherichia coli* (Gram-ve), *Acinetobacter baumanni* (Gram-ve) were selected for the study. The bacterial strains were obtained from the Microbiology Laboratory, Faculty of Resource Science and Technology, Universiti Malaysia Sarawak, and were used for the antibacterial activities. The stock cultures were incubated at 37 °C for 24 hrs on nutrient agar (Oxoid Ltd, Wade Road, Basingstoke, Hants, RG2 8PW, UK.), and was stored at 4 °C. Plates containing Mueller-Hinton Agar (MHA) were used to grow the bacterial strains at 37 °C.

The antibacterial activity of the stem-bark of *Barringtonia asiatica* was determined using disk diffusion method as reported by various authors (Boyan et al., 2005; Prashanth et al., 2006). The extract was dissolved using methanol and sterilized by filtration and stored at 4 °C until use. Standard antibiotics (tetracycline) was used for comparison of the zone of inhibition of the pure strains of the bacteria. The extracts were then screened for their antibacterial activity against the bacterial strains. Set of four dilutions for antibacterial activity (50, 100, 250, 500 µg/mL) of the stem-bark *Barringtonia asiatica* and standard drug (tetracycline) was prepared in distilled water. Sterile plates containing Mueller-Hinton agar were seeded with indicator bacterial strains and control experiment using tetracycline as standard drug were kept for 3 hrs at 37 °C. They were then incubated for 24 hrs at 37 °C, and the zones of growth inhibition around the disks were measured in mm. The antibacterial activity of the test organisms on the plant extracts were determined by measuring the diameter of the inhibitory zones on the surface of the agar around the disk, and the values <9 mm were considered as not active against the microorganism for antibacterial activity (Prashanth et al., 2006). The experiment was carried out in triplicate and the mean values of the diameter of zones of inhibition was calculated using statistical software SPSS 22.

Antifungal Assay

The antifungal activities of the stem-bark extract and the pure compounds isolated of *Barringtonia asiatica* at varying concentration (25, 50, 100, 250 and 500 µg/mL) prepared in methanol were performed against a standard drug fluconazole (positive control) (500 µg/mL) using standard paper dilution method as described by Aboh et al. (2014) with slight modification. The fungi species (*Aspergillus niger, Aspergillus flavus, Fusarium oxysporium* and *Candida tropicallis*) were obtained from the Faculty of Resource Science and Technology, UNIMAS culture collection. The strains were maintained in culture medium of potato dextrose agar (PDA). Fresh cultures of the pathogens were grown in sterilized potato dextrose broth (PDB) and incubated at 25 °C for 24 hrs. The cultures were then diluted with PDB until use. The antifungal activity of the test organisms on the plant extracts were determined by measuring the diameter of the inhibitory zones on the surface of the agar around the disk, and the values <16 mm were considered as active against the microorganism for antifungal activity (Aboh et al., 2014). The experiment was carried out in triplicate and the mean values of the diameter of zones of inhibition was calculated using statistical software SPSS 22.

RESULTS AND DISCUSSION

Cytotoxicity of crude stem-bark extract of *Barringtonia asiatica*

Brine shrimp (Artemia salina) was used to test for cytotoxicity of *Barringtonia asiatica* stem-bark crude extract From the result obtained shown in Table 1, methanol fraction of the stem-bark extract was observed to exhibit the highest cytotoxic activity with LC_{50} value of 34.059 µg/mL, and the dichloromethane fraction extract exhibited the lowest brine shrimp cytotoxic activity with LC_{50} value of 47.722 µg/mL.

The most exploring feature of this study is that the crude extract exhibited mortality rate of the brine shrimp with concentration dependent increment. However, this result obtained is not an affirmation of the toxicity of either the crude extract of the isolated pure compounds, but rather an indication of a proof the bioactive components of the plants products to have an antimicrobial activity (Abhilasha et al., 2013; Adelowotan et al., 2008; Prashith et al., 2010)

Brine shrimp bioassay has been established as safe, practical and economical method for the determination of bioactivities of isolated compounds. This established a significant correlation with *in-vitro* growth inhibition of human solid tumour cell lines as demonstrated by the National Cancer Institute (NCI, USA) where it shows the value of this bioassay as a pre-screening tool for antitumor drug research (Anderson et al., 1991).

Table 1: Average death of brine shrimp (*Artemia salina*) at different concentrations of the stem-bark extract

Plant part	Solvent system	Average death of *Artemia salina* Concentration (µg/mL)				LC_{50} (µg/mL)
		5	**10**	**50**	**100**	
Stem-bark	-ve Control	0	0	0	0	0
	+ve Thymol	5 ± 0.55	7 ± 0.55	10 ± 0.55	10 ± 0.55	1.16
	Hexane	3 ± 0.57	4 ± 0.55	5 ± 0.55	7 ± 0.55	35.462
	Dichloromethane	3 ± 0.57	3 ± 0.57	4 ± 0.57	5 ± 0.00	**47.722**
	Chloroform	3 ± 0.56	3 ± 0.56	4 ± 0.56	5 ± 0.10	46.924
	Ethyl acetate	2 ± 0.55	3 ± 0.55	4 ± 0.55	4 ± 0.00	62.220
	Methanol	3 ± 0.54	4 ± 0.54	4 ± 0.54	7 ± 0.54	**34.059**

The result is Mean ± SD. N = 30

There was an observed concentration dependent increment in mortality rate of the brine shrimp.

Antioxidant of *Barringtonia asiatica* crude stem-bark extract

Many medicinal plants as well as the pure bioactive isolates have demonstrated tremendous beneficial therapeutic potentials, and many herbs were reported to contain antioxidant properties, and most of these activities are largely attributed to their phytochemicals (Aqil et al., 2006). Antioxidants are substances that possess free radical chain reaction breaking properties (Prakash et al., 2009); their antioxidant activity is a fundamental property important for life (Velioglu et al., 1998) and has been shown to reduce oxidative stress-induced tissue injury (Pourmorad et al., 2006).

Several authors have in recent times reported on the antioxidant activity of medicinal plants all over the world (Chayarop et al., 2011; Sridharamurthy et al., 2011; Bharti et al., 2012; Patel Chirag et al., 2013). Plants that are reported to possess ethnopharmacological properties and antibacterial activities, that are used for the treatment of diseases in traditional medicine have been proven to contain antioxidant activities and useful in treatment of cancerous lesions.

The antioxidant effect of the stem-bark and isolated pure compounds of *Barringtonia sciatica* are shown in Table 2. The results of the study showed that chloroform fractions of the stem-bark crude extract exhibit strong antioxidant activity with IC_{50} values of 34.46±0.32 µg/mL, while weaker antioxidant activity was observed with hexane extract of IC50 values less than 100 µg/mL of 63.4±0.11 µg/mL when compared to the test control Ascorbic acid of 17.27.4±0.16 µg/mL.

The plot of DPPH free radical scavenging activity of ascorbic acid and *Barringtonia asiatica* crude extract against concentrations of the extract in different solvents at absorbance of 517 nm is shown in Figure 1.

Table 2: IC_{50} values of stem-bark crude extract of *Barringtonia asiatica*

Plant	Crude extract	IC_{50} (µg/mL)	R^2
Stem-bark	Standard Ascorbic acid	17.27.4±0.16	0.9937
	Hexane	**63.4±0.11**	0.9824
	Dichloromethane	51.91±0.921	0.9362
	Chloroform	**34.46±0.32**	0.9886
	Ethyl acetate	44.26±0.26	0.9949
	Methanol	35.33±0.210	0.9838

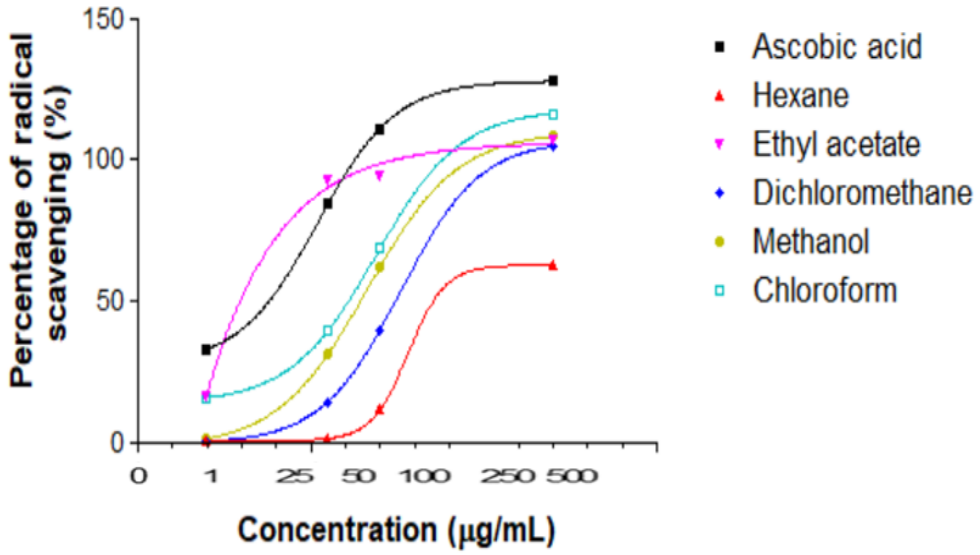

Fig. 1: Radical scavenging activities of stem-bark extract in different solvents at absorbance of 517 nm.

Antibacterial activity of *Barringtonia asiatica* crude stem-bark extract

For centuries, plant extract has been known and the therapeutic efficacy of this plants in the treatment of several disorders and infectious diseases has been described by various practitioners of traditional medicine (Wakawa et al., 2017; Umaru et al., 2018)). Harmful microorganisms are being control with synthetic drugs, and continuous treatment results in emergence of multiple drug resistance bacteria which creates an alarming clinical situation. Therefore, explore researchers in search of natural product to cutile this menace associated to synthetic drug resistance pathogens (Siddiq et al., 2009).

Table 3 shows the mean value of zone of antibacterial activity of *Barringtonia asiatica* crude extract. Significant activity was observed in all the fraction at 500 µg/mL, higher activity was observed with ethyl acetate extract and methanol extract of 20.80 ± 0.10 mm and 20.80 ± 0.10 mm, respectively on *Exiguobacterium aquaticum*. While, lower activity was observed with chloroform extract of 7.57 ± 0.06 on *Exiguobacterium aquaticum* at 25 µg/mL. This shows that polar solvent extracts exhibit significant antibacterial activity against Gram –ve and Gram +ve bacterial.

Antifungal activity of *Barringtonia asiatica* crude stem-bark extract

Antifungal activity of the extracts Table 4 was performed against four fungal strain *Aspergillus niger*, *Aspergillus flavus*, *Fusarium oxysporium and Candida tropicallis* at 500 µg/mL. All the fractions of the stem-bark extract of *Barringtonia asiatica* exhibited considerable antifungal activity against the selected fungi with zone of growth inhibition between 10.07± 0.08 mm to 17.33 ± 0.04 mm as compared to fluconazole with diameter of 24.67 ± 0.11 mm, except in hexane fraction which did not show any considerable activity when compared to the report of Aboh et al., (2014) that < 16 mm is considered as active against the microorganism of antifungal activity.

The activity of the crude extract increases with increase in the concentration gradient, this shows stem-bark extract of *Barringtonia asiatica* to contain phytochemicals which are responsible in the potential sources of antimicrobial efficacy. Thus, the agents could be used to develop effective drugs against the human pathogens.

Table 3: Mean values of the zone of growth inhibition of extract of stem-bark extract of *Barringtonia asiatica* of different concentration on selected bacteria

		Crude extract Diameter of the zone of inhibition in mm					
Concn (µg/mL)	**Control and Organism**	**Tetracycline**	**Hexane**	**DCM**	**Ethyl acetate**	**Chloroform**	**Methanol**
25	*Staphylococcus aureus*	20.77 ±0.03	8.63 ± 0.06	8.55 ± 0.07	7.87 ± 0.06	8.67 ± 0.15	7.87 ± 0.12b
	Exiguobacterium aquaticum	19.79 ±0.06	8.43 ± 0.06	8.47 ± 0.06	7.57 ± 0.06b	**7.47 ± 0.15**	8.57 ± 0.06b
	Escherichia coli	21.16 ±0.11	11.73 ±0.06	11.60 ± 0.00a	11.67 ± 0.06	9.67 ± 0.06a	9.73 ± 0.23
	Acinetobacter baumanni	20.76 ±0.18	11.60 ±0.00	12.57 ± 0.06	10.0 ± 0.00b	8.63 ± 0.06	10.63 ± 0.12
50	*Staphylococcus aureus*	20.77 ±0.03	9.70 ± 0.00	10.63 ± 0.15	10.87 ± 0.06a	8.67 ± 0.06	11.00 ± 0.20
	Exiguobacterium aquaticum	19.79 ±0.06	18.60 ± 0.00	17.50 ± 0.00	17.70 ± 0.10	12.73 ± 0.06b	13.70 ± 0.00
	Escherichia coli	21.16 ±0.11	13.93 ± 0.15	14.90 ± 0.10	13.77 ± 0.06	12.80 ± 0.10	13.60 ± 0.00
	Acinetobacter baumanni	20.76 ±0.18	11.73 ± 0.06	11.70 ± 0.00	12.83 ± 0.06	10.70 ± 0.17	14.80 ± 0.10
100	*Staphylococcus aureus*	20.77 ±0.03	11.73 ± 0.06	10.67 ± 0.15	11.83 ± 0.12a	10.73 ± 0.21	14.93 ± 0.06
	Exiguobacterium aquaticum	19.79 ±0.06	19.73 ± 0.06	19.70 ± 0.10	**20.80 ± 0.10**	11.83 ± 0.06b	**20.80 ± 0.10**
	Escherichia coli	21.16 ±0.11	16.97 ± 0.06	15.00 ± 0.10	14.87 ± 0.06	13.90 ± 0.20	15.83 ± 0.06
	Acinetobacter baumanni	20.76 ±0.18	12.80 ± 0.10a	12.77 ± 0.06	13.90 ± 0.00	10.83 ± 0.06a	15.77 ± 0.21b
250	*Staphylococcus aureus*	20.77 ±0.03	14.83 ± 0.12	15.93 ± 0.15b	15.80 ± 0.17b	12.77 ± 0.66	16.70 ± 0.20
	Exiguobacterium aquaticum	19.79 ±0.06	16.73 ± 0.06*	17.93 ± 0.06*	18.83 ± 0.12*	14.00 ± 0.10	23.03 ± 0.06b
	Escherichia coli	21.16 ±0.11	11.03 ± 0.06	16.10 ± 0.10	15.97 ± 0.06	15.03 ± 0.12a	17.03 ± 0.06
	Acinetobacter baumanni	20.76 ±0.18	12.87 ± 0.06	11.97 ± 0.06	14.00 ± 0.00	11.03 ± 0.12	15.97 ± 0.06b
500	*Staphylococcus aureus*	20.77 ±0.03	12.87 ± 0.23	17.77 ± 0.12a	17.90 ±0.10	13.87 ± 0.15	17.97 ± 0.06b
	Exiguobacterium aquaticum	19.79 ±0.06	14.83 ± 0.06	16.03 ± 0.06	16.20 ± 0.10	15.07 ± 0.06*	27.30 ± 0.10b*
	Escherichia coli	21.16 ±0.11	18.10 ± 0.10a	18.23 ± 0.06*	18.03 ± 0.06	15.03 ± 0.06	18.06 ± 0.06
	Acinetobacter baumanni	20.76 ±0.18	13.97 ± 0.06	14.00 ± 0.10	16.10 ± 0.10	14.13±0.06ab	14.07 ± 0.06

Values are Mean ± SD for three determinations

*Significantly ($p < 0.05$) higher compared to different plant part at the same concentration in each column

Concentration of standard is 30 µg/mL of tetracycline

Table 4: Mean values of the zone of growth inhibition of *Barringtonia asiatica* stem-bark extract of different concentration on selected fungi

		Concentration (µg/mL)					
Plant parts	Organism	Control	25	50	100	250	500
Hexane	*Aspergillus niger*	24.67±0.11	10.48 ± 0.17	13.75 ± 0.55ad	14.12± .21ad	16.62±0.02ad	16.03±0.19ad
	Aspergillus flavin	23.4 ± 0.05	10.25 ± 0.27	12.00 ± 0.26a	14.02 ± 0.46a	15.41 ± 0.05a	15.45 ± 0.23a
	Candida tropicalis	23.1 ± 0.08	11.97 ± 0.05	13.28 ± 0.14a	12.82 ±0.16a	14.12 ± 0.04a	15.56 ± 0.26a
	Fusarium oxysporium	23.2 ± 0.10	**10.07 ± 0.08**	12.15 ± 0.19a	12.05 ± 0.10a	13.93 ± 0.06a	14.13 ± 0.20a
Dichloromethane	***Aspergillus niger***	24.67±0.11	10.15 ± 0.48	13.45 ± 0.05ad	14.55±0.05ab	15.92 ± 0.04a	15.34 ± 0.10a
	Aspergillus flavin	23.4 ± 0.05	11.10 ± 0.06	12.22 ± 0.08a	14.33± .12ab	15.10±0.03ad	16.23±0.09ad
	Candida tropicalis	23.1 ± 0.08	12.98 ± 0.08	14.00 ± 0.06a	13.07 ± 0.05a	14.93 ± 0.18	16.45 ± 0.08a
	Fusarium oxysporium	23.2 ± 0.10	11.85 ± 0.14	12.90 ± 0.11a	12.97 ± 0.10a	14.64 ± 0.03	15.53 ± 0.16
Chloroform	*Aspergillus niger*	24.67±0.11	11.70 ± 0.06	13.75 ± 0.05	14.58 ± 0.08	15.15 ± 0.02a	15.22 ± 0.27
	Aspergillus flavin	23.4 ± 0.05	12.45 ± 0.05	14.62 ± 0.04ad	16.13 ± 0.08d	16.87 ± 0.02d	16.23 ±0.04a
	Candida tropicalis	23.1 ± 0.08	11.27 ± 0.05	14.03 ± 0.05	12.90 ± 0.09	14.54 ± 0.06	14.13 ± 0.06
	Fusarium oxysporium	23.2 ± 0.10	11.68 ± 0.12	14.47 ± 0.08	14.02 ± 0.04	14.32 ± 0.08	14.34 ± 0.04
Ethyl acetate	*Aspergillus niger*	24.67±0.11	11.28 ± 0.08	14.58 ± 0.08ab	15.15 ± 0.05a	15.44 ± 0.03ad	15.35 ± 0.18a
	Aspergillus flavin	23.4 ± 0.05	11.48 ± 0.29	13.08±0.12abd	15.45 ±0 .26d	16.72 ± 0.20a	**17.33 ± 0.04a**
	Candida tropicalis	23.1 ± 0.08	10.72 ± 0.08	15.13 ± 0.08b	13.82 ± 0.10	15.34 ± 0.19	15.46 ± 0.05
	Fusarium oxysporium	23.2 ± 0.10	11.20 ± 0.11	14.40 ± 0.13	15.62 ± 0.12b	15.56 ± 0.01ad	15.56 ± 0.06
Methanol	*Aspergillus niger*	24.67±0.11	10.07 ± 0.05	14.20 ± 0.06a	15.30 ± 0.06a	14.45 ± 0.04a	15.76 ± 0.05a
	Aspergillus flavin	23.4 ± 0.05	11.87± 0.05	16.10 ± 0.09ad	15.93 ± .05ad	15.12±0.03d	16.22 ±.05ad
	Candida tropicalis	23.1 ± 0.08	10.65 ± 0.05	13.52 ± 0.08	13.20 ±0.06 a	14.46 ± 0.04a	14.24 ± 0.08a
	Fusarium oxysporium	23.2 ± 0.10	11.12 ± 0.08	12.83 ± 0.05a	14.57 ± 0.05a	16.71 ± 0.01a	16.56 ± 0.05a

Values are Mean ± SD for three determinations

*Significantly ($p < 0.05$) higher compared to different plant part at the same concentration in each column

Concentration of standard is 30 µg/mL of tetracycline.

CONCLUSION

Crude extracts of the stem-bark of *Barringtonia asiatica* was obtained after successive extraction using five solvents, namely hexane, dichloromethane, chloroform, ethyl acetate and methanol. The crude extract was screened for biological activity, which include cytotoxicity, antioxidant, antifungal and antibacterial activity. The results obtained indicated that the plant could be used as an agent for ailment and diseases control.

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Conflict of Interest

The authors declare no competing of interest.

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Availability of data and materials

The data sets generated and analysed during the current study are available from the corresponding author on reasonable request.

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