



(RESEARCH ARTICLE)



## Phytochemical analysis and antibacterial activity of *Bryophyllum pinnatum* (Africa Never Die) leaf extract on bacterial organisms isolated from urine of pregnant women attending a tertiary health facility in Enugu, Nigeria

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### Abstract

Leaves of *Bryophyllum pinnatum* is used in traditional medicine in various parts of the world as a remedy against numerous conditions to treat infections. This study was carried out to evaluate the antibacterial activities of the plant leaf extracts of *Bryophyllum pinnatum* on some bacteria isolated from urine of pregnant women. A total of 20 urine samples were collected from pregnant women attending antenatal care at University of Nigeria Teaching Hospital (UNTH) Enugu. The samples were cultured onto Nutrient agar, MacConkey agar and Mannitol salt agar. The isolates were characterized and identified on the basis of Gram staining reaction, biochemical tests and polymerase chain reaction (PCR). The leaves of *Bryophyllum pinnatum* were collected and subjected to maceration extraction processes using water, ethanol, chloroform and ethyl acetate to obtain aqueous, ethanol, chloroform and ethyl acetate extracts respectively. Isolates were subjected to quantitative and qualitative phytochemical screening by standard methods. Isolates were also screened for sensitivity to the extracts using the agar-well diffusion method.

The qualitative phytochemical analysis revealed the presence of various bioactive compounds which includes alkaloids, flavonoids, saponins, phenols, steroids, tannins and glycosides. Though they varied in the different extracts, ethanol extracted most of the active components than other solvents. Quantitatively, alkaloid was highest in ethanol concentration ( $3.80 \pm 0.57^a$ ). Prevalent bacterial organisms were *Proteus mirabilis* 6(30%), *Pseudomonas aeruginosa* 5(25%), and *Klebsiella aerogenes* 4(20%). The results indicated that the extracts showed varying inhibition rates. The organisms were most resistant to the aqueous and chloroform extract than the ethanolic and ethyl acetate extract. At 200mg/ml, ethanol extract showed high zones of inhibition at 22 mm on the isolates. The minimum inhibitory concentration (MIC) of the extracts showed that growth of the isolates was inhibited by all the extracts between the concentrations of 12.5 mg/ml and 50 mg/ml. The minimum bactericidal concentration (MBC) concentration at which no growth occurred revealed that the aqueous and chloroform extracts demonstrated lesser bactericidal activity at 200 mg/ml while that of ethanol and ethyl acetate showed higher inhibition rates at 100mg/ml each. The results of this finding showed that the extracts of *Bryophyllum pinnatum* has inhibitory effect on bacterial organisms isolated from urine of pregnant women and can be used to treat infections caused by these bacteria.

**Keywords:** Phytochemicals; Antibacterial; *Bryophyllum pinnatum*; Urine; Bacteria

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## 1. Introduction

Urinary tract infections (UTIs) are considered to be the most common bacterial infection, affecting more than 150 million people annually worldwide [1]. The prevalence of symptomatic and asymptomatic bacteriuria among women during pregnancy is very common and the previous history of the infection is a major risk factor. The effect of asymptomatic UTI can be subsided by employing suitable treatment which in turn prevents the adverse consequences of its progress. The predominant pathogen responsible for urinary tract infection (UTI) is *Escherichia coli*, followed by *Staphylococcus saprophyticus*. In addition to the above-mentioned bacterial species, *Klebsiella*, *Enterococcus*, *Proteus species*, *Pseudomonas* and *Enterobacter* are associated with UTI. The bacteria often enter the bladder through the urethra from the bowel [2]. Urinary tract infections are treatable and will usually not result in complications when diagnosis is made early and antibiotics are administered.

The emergence and spread of resistant strains of bacteria to routinely used antibiotics has made it imperative to continuously search for alternatives that can be used to cure infections, so as not to return to pre-antibiotic era [3].

In recent times however, the continuous emergence and spread of resistant strains of bacteria to routinely used antibiotics, the shy attitude of most people in discussing infections that affect genitourinary tract and the high cost of health care in developing countries has necessitated the use of other alternatives such as herbs. Traditional medicine as defined by the World Health Organization is the total of the knowledge, skills, and practices based on the theories, beliefs, and experiences indigenous to different cultures, whether explicable or not, used in the maintenance of health as well as in the prevention, diagnosis, improvement, treatment of physical and mental illness [4]. The use of local plants as primary health remedies, due to their pharmacological properties, is quite common in Asia, India, Latin America, USA, China, Japan and Africa [5]. Globally, there are evidence-based studies to verify the efficacy of medicinal plants, and some of these evidences have provided insights into the synthesis of plant-based compounds with therapeutics application.

The plant kingdom consists of a variety of plants which are of immense importance to humans and of valuable use in the treatment of various illnesses [3,6]. The medicinal value of these plants lies on their chemical and phytochemical substances they contain. Different parts (bark, root, twig, fruit and leaves) of different plants have been studied and found to be sources of antimicrobial agents. Many of these indigenous medicinal plants are used as spices and food plants that are sometimes added to foods for pregnant and nursing mothers for medicinal purposes [6]. Medicinal plants contain biologically active chemical substances such as saponins, tannins, essential oil flavonoids, alkaloids and other chemicals which have curative properties. These complex chemical substances of different composition are found as secondary plant metabolite in one or more of these plants [7].

*Bryophyllum pinnatum* belongs to the Crassulaceae family and is commonly found in tropical Africa, America, China, India and Australia [8]. In Nigeria, it is referred to as 'odaopue' by the Igbos 'shuka halinka' by the Hausas, and 'abamoda' by the Yorubas. The plant is widely used in traditional medicine for the treatment of variety of ailments. *Bryophyllum pinnatum* is used in ethnomedicine. The leaves and leaf juice have been used traditionally as antiinflammatory, antipyretic, antimicrobial, anti-oxidant, antitumour, antidiabetic, antiulcer, antiseptic, hypocholesterolemic, and cough suppressant [9,10,11]. The leaves or the whole plant are used as analgesic and generally for the treatment of ear ache, cough, asthma, diarrhoea, dysentery, jaundice, abscesses, ulcers, insect bites, heart troubles, epilepsy, arthritis, dysmenorrhoea, whitlow and other ailments [10]. This wide range of traditional uses justifies its being called "life plant", "resurrection plant", "goodluck", love plant, cathedral bells or miracle leaf and is distinctive for the profusion of miniature plantlets that form on the margins of its Phylloclades, a trait it has in common with some other members of its genus [10]. Thus, this study is aimed at the determination of the phytochemical constituents and antibacterial activity of *Bryophyllum pinnatum* leaf extract on bacterial isolates from cultured urine of pregnant women.

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## 2. Materials and methods

### 2.1. Sample Collection

The fresh leaves of *Bryophyllum pinnatum* (Africa never die or oada opue in Igbo) were bought from Ogbete market in Enugu State. The leaves were placed in clean and sterile Ziplock bags and transported immediately to the Applied Microbiology and Brewing laboratory Enugu State University of Science and Technology (ESUT), Enugu State. It was identified and authenticated by Prof. C. C Eze at the Department of Biology Enugu State University of science and Technology (ESUT), Enugu.

- **Preparation of Crude Extract from the Plant Materials:** The fresh leaves of *Bryophyllum pinnatum* were washed severally to remove dirt and contaminants. They were dried under shade at room temperature for 21 days and then pulverized using electric blender. They were stored in an airtight bag for further use [12]. Maceration extraction method was used for the extraction. A total of 100g of air-dried *Bryophyllum pinnatum* leaf powder was weighed into a conical flask with 300 ml of distilled water. The mixture was left at room temperature for 3 days for maceration and occasionally shaken to enable proper diffusion of the active ingredients. After that, the solution was filtered using Whatman's filter paper (No 1) filter paper. The filtrates were evaporated to dryness in a water bath at 50 °C and it was then stored at 4 °C for further use. The same method of preparation was carried out for ethanol, ethyl acetate, and chloroform to obtain ethanol extract, ethyl acetate extract, and chloroform extract respectively. The percentage yield of each extract was calculated.
- **Phytochemical Screening of Plant Extract:** The extract and fractions were subjected to both quantitative and qualitative phytochemical screening using standard phytochemical methods as outlined by Uthayarasa [13]. Test for alkaloids, saponins, steroids, phenol, flavonoids, tannins and glycosides were carried out.
- **Media Preparation:** All media used for the microbiological analysis were prepared according to manufacturer's instructions.
- **Isolation of Organism:** A total of 20 urine samples were collected from pregnant women attending antenatal care at University of Nigeria Teaching Hospital Enugu, Enugu state. The samples were cultured onto Nutrient agar, MacConkey agar and Mannitol salt agar for identification. All isolates were sub cultured onto nutrient agar plates and further sub cultured into nutrient agar slants for further use.
- **Standardization of Bacterial Inoculum:** A 100µl of each of the pure isolates were transferred into sterile 5ml nutrient broth in a test tube and incubated at 37 °C for 24hr. 1ml of the organism from the nutrient broth was dispensed into 5ml of sterile saline they were vortexed thoroughly. Adjustment was made with extra inoculum or diluents until 0.5 MacFarland turbidity standard were obtained.
- **Characterization and Identification of Bacteria Isolates:** The isolates were characterized and identified on their basis of Gram staining reaction and biochemical tests (catalase, citrate, coagulase, oxidase, urease, Indole, methyl red and sugar fermentation test as described by Naveena [14]. They were further characterized by polymerase chain reaction (PCR).
- **Antimicrobial Sensitivity Test:** Agar-well diffusion method was used for antimicrobial sensitivity test by method described by Ofokansi *et al.* [15]. Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal concentration (MBC) was also determined.

### 3. Result

#### 3.1. Phytochemical Analysis of *Bryophyllum pinnatum* Leaf Extract

The plant showed varying results in the various extracts (Table 1 and 2).

#### 3.2. Identification Scheme of the Bacterial Isolates

From the study, organisms isolated were *E. coli*, *Pseudomonas aeruginosa*, *Staphylococcus sp.* *Proteus mirabilis*, *Klebsiella sp.*, and *Streptococcus sp.* (Table 3).

#### 3.3. Occurrence of Bacterial Isolates from Urine Samples

Out of 20 samples analyzed, *Proteus mirabilis* occurred highest at 30%, followed by *Pseudomonas aeruginosa* 25%, *Klebsiella aerogenes* 20%, *E. Coli* 15%, *Staphylococcus sp.* 5% and *Streptococcus sp.* 5% (Table 4).

#### 3.4. Molecular Characterization of Isolates

From this study, *Pseudomonas aeruginosa*, *Proteus mirabilis* and *Klebsiella aerogenes* have 93.78, 95.51 and 90.66 percentage similarity respectively after being identified according to the percentage of similarity to reference sequences of strains (Table 5).

#### 3.5. Antibacterial Activity of *Bryophyllum pinnatum* Leaf Extract

The *Bryophyllum pinnatum* leaf from various extracts showed an appreciable inhibitory activity in different concentrations for the test organisms (Table 6).

### 3.6. Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC)

The least inhibitory concentration of the extract inhibition was found at 12.5mg/ml and the least concentration at which no growth occurred were recorded in aqueous and chloroform extract at 200mg/ml each (Table 7 and 8).

**Table 1** Qualitative Phytochemical Analysis of the *Bryophyllum pinnatum* Leaf Extract

Constituents	Experimental method	Ethanol extract	Aqueous extract	Ethyl acetate extract	Chloroform extract
Saponins	Foam Test	+++	+	+	-
Tannins	Ferric Chloride Test	++	+	+	-
	Lead Acetate Test	++	-	-	-
Flavonoids	Alkaline Test	+	+	+	+
	20% NaOH Test	++	+	-	+
Alkaloids	Wagner's Test	+	+	+	+
	Mayer's Test	++	+	+	+
	Dragendorff Test	+	+	+	+
Cardiac Glycosides	Keller-Kilani Test	+	+	+	+
Phenol	5%FeCl <sub>3</sub> Test	++	+	+	-
Steroids	Salkowski's Test	++	-	-	-
Terpenoids		+	-	+	-

+ Present in trace concentration; Present in moderate high concentration ++; +++ present in high concentration; - not detected

**Table 2** Quantitative Phytochemical Analysis of different solvent extract of *Bryophyllum pinnatum* Leaf Extract

Extracts/Constituents	Alkaloids %	Saponins %	Flavonoids %	Phenols %	Tannins %	Glycosides %
Aqueous extract	2.00±0.05 <sup>a</sup>	3.00±0.42 <sup>a</sup>	0.86±0.02 <sup>a</sup>	0.43±0.01 <sup>a</sup>	0.05±0.94 <sup>a</sup>	0.29±0.00 <sup>a</sup>
Ethanol extract	3.80±0.57 <sup>a</sup>	5.50±0.35 <sup>b</sup>	0.40±0.00 <sup>c</sup>	0.46±0.00 <sup>d</sup>	6.75±1.89 <sup>c</sup>	1.43±0.00 <sup>c</sup>
Chloroform extract	0.20±0.70 <sup>a</sup>	0.03±0.53 <sup>ab</sup>	0.15±0.01 <sup>ab</sup>	0.01±0.02 <sup>b</sup>	0.06±1.15 <sup>b</sup>	0.45±0.00 <sup>b</sup>
Ethyl acetate extract	0.02±0.98 <sup>a</sup>	1.3±2.8 <sup>b</sup>	0.01±0.00 <sup>bc</sup>	0.24±0.01 <sup>c</sup>	4.50±0.09 <sup>c</sup>	1.4±0.00 <sup>b</sup>

Mean values with different letters of the alphabet down the column are significantly different ( $p \leq 0.05$ ) while mean values with same letters of the alphabet down the column are not significantly different ( $p \geq 0.05$ )

**Table 3** Cultural Identification, Morphological and Microscopic Characteristics of bacterial Isolates

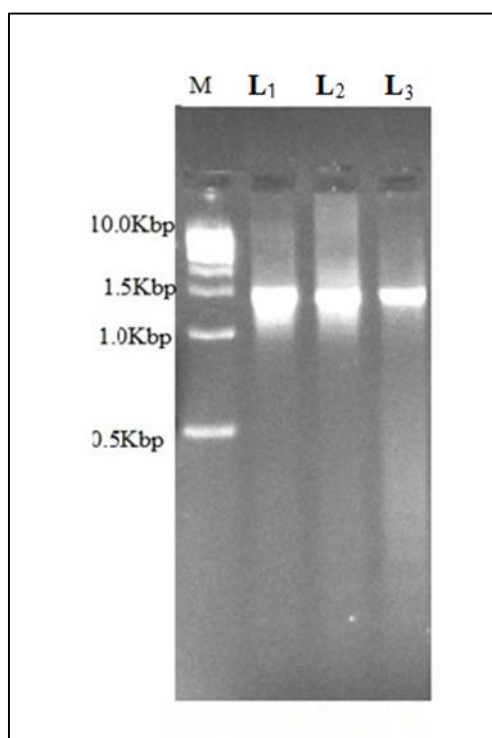
Sample	Growth Appearance on Media	Gram reaction	Biochemical Test					Sugar Fermentation Test						Suspected Organisms
			Catalase test	Oxidase test	Citrate test Test	Coagulase test	Methyl red test	Indole test	Glucose	Fructose	Maltose	Mannitol test	Lactose	
1	Bright, pink convex round colony on MacConkey agar.	-ve short rod in pairs	+ve	-ve	-ve	-ve	+ve	+ve	AG	AG	AG	AG	AG	<i>E. coli</i>
2	Round, flat, and colorless colonies on MacConkey agar.	-ve short rod in pairs	+ve	+ve	+ve	-ve	+ve	+ve	-ve	A	A	A	-ve	<i>Pseudomonas aeruginosa</i>
3	Large yellow colonies on mannitol salt agar.	+ve cocci in clusters	+ve	-ve	+ve	-ve	+ve	-ve	A	A	A	A	A	<i>Staphylococcus sp.</i>
4	Large raise mucoid and whitish colony on nutrient agar.	-ve short rod in pairs	+ve	-ve	+ve	-ve	+ve	-ve	A	A	A	-ve	-ve	<i>Proteus mirabilis</i>
5	Medium pinkish raised mucoid colonies on MacConkey agar.	-ve short rod in pairs	+ve	-ve	+ve	-ve	-ve	-ve	A	A	A	A	A	<i>Klebsiella sp.</i>
6	small greyish-white, moist colonies on nutrient agar.	+ve cocci in clusters	-ve	-ve	+ve	-ve	+ve	-ve	A	A	A	A	A	<i>Streptococcus sp.</i>

KEY: A=Acidic, AG=Acid and Gas, G=Gas, -ve (Negative); +ve (Positive)

**Table 4** Percentage (%) occurrence of Bacterial Isolates from Urine Samples

Isolates	Number of isolates (n=20)	Percentages (%)
<i>E. Coli</i>	3	15
<i>Pseudomonas aeruginosa</i>	5	25
<i>Staphylococcus sp.</i>	1	5
<i>Proteus mirabilis</i>	6	30
<i>Klebsiella sp.</i>	4	20
<i>Streptococcus sp.</i>	1	5

### 3.7. Molecular Identification of Isolates



**Figure 1** Agarose gel electrophoresis of PCR amplified products of *Pseudomonas aeruginosa*, *Proteus mirabilis* and *Klebsiella aerogenes* Isolates

**Table 5** Molecular Characterization of Isolates

Bacteria Isolates	Strain	% Identity	Accession Number
<i>Pseudomonas aeruginosa</i>	PHB3	93.78	JQ327806
<i>Proteus mirabilis</i>	K1WTRM5	95.51	MH985194
<i>Klebsiella aerogenes</i>	CX-70	90.66	MH368390

**Table 6** Antibacterial Activity of Leaf Extracts of *Bryophyllum pinnatum* on Isolates Concentrations (mg/ml) of the zones of Inhibition (mm)

Test organisms	Ethanol					Ethyl acetate					Chloroform					Aqueous					Control (Ciprofloxacin)
	200	100	50	25	12.5	200	100	50	25	12.5	200	100	50	25	12.5	200	100	50	25	12.5	
<i>Klebsiella aerogenes</i>	20.5	17	13	10	0	10	8	4	0	0	6	4	0	0	0	8	5	0	0	0	22
<i>Proteus mirabilis</i>	17	14	10	7	0	15	13	8	0	0	4	2	0	0	0	7	3	0	0	0	19
<i>Pseudomonas aeruginosa</i>	22	19	17	12	0	17	15	11	0	0	8	5	0	0	0	9	8	0	0	0	25

**Table 7** Minimum inhibitory concentration of *Bryophyllum pinnatum* plant extract against test bacteria

Test Organism	Extract/Conc. (mg/ml)	MIC (mg/ml)
<i>Klebsiella aerogenes</i>	Ethanol	12.5
	Aqueous	50
	Ethyl Acetate	25
	Chloroform	50
<i>Proteus mirabilis</i>	Ethanol	12.5
	Aqueous	50
	Ethyl Acetate	25
	Chloroform	50
<i>Pseudomonas aeruginosa</i>	Ethanol	25
	Aqueous	50
	Ethyl Acetate	25
	Chloroform	50

**Table 8** Minimum bactericidal concentration of *Bryophyllum pinnatum* plant extract against test bacteria

Test Organism	Extract/Conc. (mg/ml)	MBC (mg/ml)
<i>Klebsiella aerogenes</i>	Ethanol	100
	Aqueous	200
	Ethyl Acetate	100
	Chloroform	200
<i>Proteus mirabilis</i>	Ethanol	100
	Aqueous	200
	Ethyl Acetate	100
	Chloroform	200

<i>Pseudomonas aeruginosa</i>	Ethanol	100
	Aqueous	200
	Ethyl Acetate	100
	Chloroform	200

#### 4. Discussion

This study evaluated the phytochemical analysis and antibacterial activity of *Bryophyllum pinnatum* leaf extracts on bacterial isolates from urine samples of pregnant women attending a tertiary health facility in Enugu. Microbial resistance to several antibiotics is becoming a source of challenge and concern in pregnancy and to public health. In view of the increasing rate of antimicrobial drug resistance ravaging not only the African continent but the world at large, alternative, effective and affordable substitutes are essential if bacterial infections are to be properly controlled. Phytochemicals are secondary metabolites of plants known to exhibit diverse pharmacological and biochemical effects on living organisms. The phytochemical analysis of the leaf extracts of *Bryophyllum pinnatum*, as presented in (Table 1), gives insights into the composition of these extracts which are known to exhibit diverse pharmacological and biochemical effects on living organisms. The result outlines the presence and concentration of various constituents in different solvent extracts, including ethanol, ethyl acetate, aqueous and chloroform extracts. The ethanolic and aqueous extracts from the plant leaf in this study contained alkaloids, flavonoids, phenol, saponins and glycosides, with the additional presence of steroids and tannins in the ethanolic extract. Tannins are known to exhibit antibacterial activities. The antimicrobial activity could be attributed to the presence of phenolic compounds which have been detected in this plant extract to include: saponin, tannins, alkaloids and other secondary metabolites which are antimicrobial [16]. The presence of these phytochemicals (steroids, tannins, flavonoids, alkaloids, saponins and glycosides) in different solvent extract of *B. pinnatum* used in this study, supports their use as medicinal plants. The outcome of this study conforms to the detectable phytochemicals present in the leaves of *Bryophyllum pinnatum* such as alkaloids, saponins, flavonoids, tannins, steroid and glycosides as reviewed by several authors [17,18,19,20]. The variations in some phytochemicals could be due to the solvent used. Higher polarity possessed by the ethanol gives it higher penetration property and therefore the extract removes more bioactive compounds from the powdered leaf than the aqueous, ethyl acetate and chloroform solvent. The presence of these phytochemicals in different solvent extracts of *Bryophyllum pinnatum* used in this study supports their use as medicinal plants. These chemical constituents could be responsible for their antibacterial activity [4]. The quantitative phytochemical analysis of *Bryophyllum pinnatum* leaf extracts reveals significant variations in the composition of different constituents across various solvents (ethanol, aqueous, ethyl acetate and chloroform). Ethanol extract demonstrated higher concentrations of alkaloids ( $3.80 \pm 0.57$ ), saponins ( $5.50 \pm 0.35$ ) and tannins ( $6.75 \pm 1.89$ ) compared to aqueous, chloroform and ethyl acetate extracts. Tannins were found to be highest in concentration compared to other phytochemicals while phenols were found to be lowest in concentration. These evaluations proved that the plant leaf extracts are beneficial and have potential antimicrobial effects and equally can be employed in pharmacological examinations (Table 2). From the study, various bacterial organisms isolated were *E. coli*, *Pseudomonas aeruginosa*, *Staphylococcus sp.*, *Proteus mirabilis*, *Klebsiella sp.*, and *Streptococcus sp.* (Table 3). Out of 20 samples analyzed, *Proteus mirabilis* occurred highest at 30%, followed by *Pseudomonas aeruginosa* 25%, *Klebsiella aerogenes* 20%, *E. Coli* 15%, *Staphylococcus sp.* 5% and *Streptococcus sp.* 5% (Table 4). For further identification and characterization, the isolates were subjected to molecular identification and polymerase chain reaction (PCR) was used to detect *Pseudomonas aeruginosa strain*, *Proteus mirabilis strain* and *Klebsiella aerogenes strain* for 16S rRNA genes with amplification sizes of 1.5kbp (Figure 1 and Table 5). The antimicrobial result showed that the ethanol, chloroform, ethyl acetate and aqueous extracts of *Bryophyllum pinnatum* exhibited variable inhibition rates against the test isolates investigated. The extent of sensitivity of the test isolates to the plant extracts were shown by the clear zones of inhibition produced by the extracts after the period of incubation. The sensitivity of the test isolates to ethanolic extracts of *Bryophyllum pinnatum* is in agreement with the findings of Obi and Onuoha (2000) [21], who described ethanol as the best solvent for the extraction of bioactive substances from plants. All the extracts were active at high concentrations and less or inactive at lower concentrations with activity increasing with increase in concentration. Thus, this study suggests that the inhibition of the test organisms is concentration dependent. The result of this study is therefore in line with the report of Joshi *et al.*, (2016) [22] who investigated the in vitro antibacterial activity of *Bryophyllum pinnatum* leaf extract. At the highest concentration of 200 mg/ml, ethanol extracts inhibited most of the test organisms; *Pseudomonas aeruginosa* was inhibited at 22mm, *Proteus mirabilis* at 17mm and *Klebsiella aerogenes* at 20.5mm (Table 6). This study also revealed that the ethanol extract of *Bryophyllum pinnatum* was most effective against the test organisms than ethyl acetate, aqueous and chloroform solvents. This is in-line with the assertion of Akinnibosun and Edionwe, (2015) [23] who postulated that ethanol gives higher antimicrobial effect than other extracting solvent such as aqueous, chloroform and ethyl acetate. The more



significant inhibition was observed with a higher extract concentration which could be due to the stronger extraction capacity of ethanol. This is in agreement with the observations of Ammara *et al.*, (2009) [24], who concluded that the stronger extraction capacity of ethanol could have been responsible for the higher antimicrobial activity. The stronger extraction capacity of ethanol for *Bryophyllum pinnatum* could have been responsible for the higher antibacterial and antifungal activities as the biologically active components in the plant could have been enhanced in the presence of ethanol [24]. The minimum inhibitory concentration (MIC) of the ethanol, ethyl acetate, chloroform and aqueous extracts was read as the lowest extract concentration that showed growth. The results showed that growth of the isolates was inhibited by all the solvent extracts between the concentrations of 12.5 mg/ml and 50 mg/ml (Table 7). The minimum bactericidal concentration (MBC) of ethanol and ethyl acetate extracts of *Bryophyllum pinnatum* were 100 mg/ml each while aqueous and chloroform were 200 mg/ml each for all the isolates (Table 8).

Based on the findings of this study, the ethanol, ethyl acetate, aqueous, and chloroform plant extracts of *Bryophyllum pinnatum* were active against all the test isolates but its ethanol extract demonstrated greater inhibitory activity. The ability of the ethanolic extract of *Bryophyllum pinnatum* to be more effective than that of the ethyl acetate, aqueous, and chloroform extracts could be linked to the fact that, the active antimicrobial agent in the leaves is more soluble in ethanol and as such, it is able to extract the antimicrobial constituents from the leaf [25].

This study suggests that the inhibitory effects of *Bryophyllum pinnatum* plant leaf extracts can be an alternative to the use of most antibiotics for the treatment of pathogenic strains of *Pseudomonas aeruginosa*, *Klebsiella aerogenes* and *Proteus mirabilis*. The effect of the plant extract on the test isolates is in agreement with the findings of Igwe *et al.*, [26] who suggests the efficacy of the traditional use of *Bryophyllum pinnatum* and its adoption in medicinal use.

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## 5. Conclusion

The findings from this study shows the antibacterial activities of ethanol, aqueous, ethyl acetate and chloroform extracts of *Bryophyllum pinnatum* against urine bacterial pathogens; *Klebsiella aerogenes*, *Proteus mirabilis* and *Pseudomonas aeruginosa*. The ethanol extracts showed high inhibition rates against the test isolates and the antibacterial activities were observed to be dependent on the solvents used for extraction and concentration of the extract used.

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## Compliance with ethical standards

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### Disclosure of conflict of interest

There was no conflict of interest declared

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