

## THE PHYTOCHEMICAL COMPOSITION AND ANTIMICROBIAL ACTIVITY OF *DIALIUM GUINEENSE*, *VITEX DONIANA* AND *DENNETTIA TRIPETALA* LEAVES

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### ABSTRACT

*The phytochemical constituent and the antimicrobial activity of the leaves of three wild plant species Dialium guineense (Willd), Vitex doniana (Sweet) and Dennettia tripetala (Bak. F) were investigated to ascertain their pharmaceutical potential. The study revealed that the leaves contained alkaloid, flavonoids, saponin, phenols, tannin and steroids. The percentage phytochemical constituents of the leaves of the plants were as follows: Alkaloid (1.21- 2.42), flavonoids (0.18- 0.50), saponin (0.90- 1.25), phenols (0.075- 0.142) tannin (0.159- 0.577) and steroid (0.035- 0.048). The ethanolic extracts of the leaves of the plants were tested against Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa, and Salmonella typhi by the agar well diffusion method. The extracts inhibited the growth of these pathogens. The diameter of the inhibition zone ranged between 0.1 to 14 mm. The MIC (minimum inhibition concentration) values of the extracts ranged from 2.33 to 23.30 mg/ ml. The results obtained indicate that the leaves of the plants have pharmaceutical values and can be utilized in treatment of some ailments.*

**Keywords:** Phytochemical, antimicrobial activity, minimum inhibition concentration, *Dialium guineense*, *Vitex doniana*, *Dennettia tripetala*

### INTRODUCTION

The use of plants in the maintenance of good health is well reported (Burkill, 1995; Edeoga and Eriata, 2001; Moerman, 1996). It has also been reported that the bases of many modern pharmaceuticals used today for the treatment of various ailments are plants and plant based products (Kambba and Hassan, 2010). Plants have been generally utilized for the treatment of diseases worldwide. About 80% of the world populations depend on plants based medicine for their health care (WHO, 2001). WHO (1996), also observed that the majority of the population in the developing countries still rely on herbal medicines to meet their health need. The use of plants and plant based products to meet societal health need stems from the fact that indiscriminate use of commercial antimicrobial drugs commonly utilized in the treatment of infectious diseases has led to the development of multiple drug resistance (Gupta et al., 2008), the adverse effect on host, associated with the use of conventional antibiotics (Gupta et al., 2008), the safety and cost effectiveness of the use of plants in traditional as well as in modern medicine (Koche et al., 2011), and high cost, adulteration and increasing toxic side effects of these synthetic drugs (Shariff, 2001). Thus there has been the need to develop alternative antimicrobial drugs from medicinal plants for the treatment of infectious diseases; because antimicrobials of plant origin have been found to have enormous therapeutic potential (Werner et al., 1999). Furthermore, the studies by Perumalsamy and Ignacimuthu (2000) showed that antimicrobials from plant origin are effective in the treatment infectious diseases and on the other hand simultaneously mitigates many of the side effects that are linked with synthetic antimicrobials.

Many of these indigenous plants contain bioactive compounds that exhibit physiological activities against bacteria and other microorganisms and are also used as precursors for the synthesis of useful drugs. Thus the use fullness of these plant products in medicine is due to the presence of bioactive substances such as alkaloids, tannins, flavonoids, phenolic compounds, steroids, resins and other secondary metabolites which they contain and are capable of producing definite physiological action in the body (Bishnu et al, 2009; Edeoga et al., 2005).

Phytochemicals are known to carry out important medicinal roles in the body. Alkaloids are known to have a powerful effect on animal physiology. They play some metabolic role and control development in living system (Edeoga and Eriata, 2001). They are also used as starting materials in the manufacture of steroidal drugs and carry out protective function in animals, thus are used as medicine especially steroidal alkaloids (Maxwell et al., 1995; Stevens et al., 1992). Isolated pure plant alkaloids and their synthetic derivatives are used as basic medicinal agent for their analgesic, antispasmodic and bacterial effect (Ogukwe, et al., 2004). Flavonoids are known to carry out antioxidant, protective effects and inhibit the initiation, promotion and progression of tumors (Kim et al., 1994; Okwu, 2004). Isoflavones, some kind of flavonoids are phytoestrogen which effectively modulate estrogen levels in human (Okwu and Omodamiro, 2005). A type of flavonoid anthocyanin helps in reducing the incidence of cardiovascular diseases, cancer, hyperlipidemias and other chronic diseases (de Pascual-Teresa and Sanchez-Ballesta, 2008). Phenolic compounds in plants are potentially toxic to the growth and development of pathogens (Singh and Sawhney, 1988). Research reports also show that phenolic compounds carry out potent antioxidant activity and wide range of pharmacologic activities which include anti- cancer, antioxidant and platelet aggregation inhibition activity (Rein et al., 2000; Rice – Evans et al., 1996;). Saponins play essential roles in medicine. These include serving as expectorant and emulsifying agent (Edeoga et al., 2009) and having antifungal properties (Osuagwu et al., 2007). Tannins are reported to inhibit pathogenic fungi (Burkill, 1995). They are also associated with many human physiological activities such as stimulation of phagocytic cells and host mediated tumor activity and a wide range of infective actions (Haslam, 1996). Steroid containing compounds are of importance in pharmacy due to their role in sex hormones (Okwu, 2001). Steroids such as equine estrogen are implicated in the reduction of risks of coronary heart and neurodegenerative diseases in healthy and young postmenopausal women (Perrella et al., 2003). At low concentration tannins show antimicrobial, cytotoxic and astringent properties (Zhu et al., 1997; Ijeh et al., 2004).

The phytochemical screening of some plants has been carried out and they are found to be rich in alkaloids, phenols, flavonoid, saponin and tannins (Osuagwu et al., 2007; Iniaghe et al., 2009; Ganjewala et al., 2009; Omoyeni and Aluko, 2010). The antimicrobial activities of plants have been reported (Arshad et al., 2010; Kamba and Hassan, 2010, Koche et al., 2011). They are therefore used in the treatment of many diseases such as rheumatism, diarrhea, malaria, elephantiasis, cold, obesity, dysentery, high blood pressure, malnutrition, gonorrhoea and others (Burkill, 1995; Edet et al., 2009; Akuodor et al, 2010).

*Dialium guineense* belong to the Caesalpiniaceae family. It is a tree up to 30m high, with a densely leaf crown but often shrubby with small black velvety fruit. The leaves of the plant are used in folk medicine for the treatment of ailments which include diabetes, fever and cough (Vadivu et al., 2008);

*Vitex doniana*, belongs to the Verbenaceae. It is a tree up to 25m with black edible fruit. The trees are evergreen in nature. The leaves are used in the treatment of swelling, oedema,

diabetes, ulcer and as diuretic in the treatment of high blood pressure (Burkill, 1997; Gill, 1992). Its decoction also is given during labour just before child birth to induce strong uterine contraction and shorten delivery time (Gill, 1992).

*Dennettia tripetala* belongs to the Annonaceae family. The tree grows up to 30m high and often shrubby. The fruit is edible and rich in vitamin C (Burkill, 1985). The leaves are used in folk medicine for the treatment of fever, cough, asthma catarrh, diarrhea and rheumatism (Burkill, 1985).

The objectives of this research is to ascertain the presence and quantity of some phytochemicals in the leaves of *D. guineense*, *V. doniana* and *D. tripetala* and to determine the antimicrobial activity of the leaves of these plants on the selected human pathogenic microorganism, in view of their use as alternative sources of antimicrobial drugs used in the treatment of diseases.

## MATERIALS AND METHODS

### Plant Samples

The leaves of *Dalium guineense*, *Vitex doniana* and *Dennettia tripetala* were collected from a wild farm in Amaokwe Item, Bende Local Government Area Abia State Nigeria. The plants were identified by Mr N. Ibe of the Forestry Department, College of Natural and Environmental management, Michael Okpara University of Agriculture Umudike Umuahia Abia State Nigeria. The leaves of *D. guineense*, *V. doniana* and *D. tripetala* were air dried for one week. The leaves were ground to powder using Wood land electric grinding machine. Powdered samples were stored in the Plant Science and Biotechnology laboratory to be used for analysis.

### Determination of the Phytochemical Content of the Plant Samples

Both qualitative and quantitative tests were carried out on the samples to determine the presence and the amount of the phytochemicals in the powdered samples.

### Qualitative Analysis of the Plant Samples

#### *Test for Presence of Alkaloids.*

The presence of alkaloids in each sample was investigated using the method described by Harborne (1973).

An alcoholic extract was used and obtained by dispersing 2g of the powdered sample in 10 ml of ethanol. The mixture was through shaken before filtering using Whatman No (40) filter paper. 2 ml of the filtrate was added into a test tube and 3 drops of pirovic acid was mixed with it. The formation of light green colouration indicates presence of alkaloid.

#### *Test for the Presence of Flavonoid*

The determination of presence of flavonoid in the sample was carried out using the acid alkaline test described by Harborne (1973).

2ml of the aqueous extract was added into a test tube and a few drops of Bench Concentrated ammonia (NH<sub>4</sub>) were also added. The formation of a yellow colouration shows presence of flavonoid. Confirmatory test was carried out by adding few drops of concentrated hydrochloric (HCL) into the yellow solution which turned colourless.

#### *Test for the Presence of Phenols.*

The presence of phenols in the sample was carried out using the Harborne (1973) methods.

The fat free sample was boiled with 50ml of ether for 15 minutes. 5ml of the extract was pipette into a 50ml flask and 10ml of distilled water added into it. 2ml of ammonia hydroxide solution and 5ml of concentrated amyl alcohol were also added. The mixture was allowed to react for 30 minutes for colour development.

#### ***Test for the Presence of Saponin***

The presence of saponins in the samples was determined using Harborne (1973) method.

Two tests were involved in the investigation, the froth test and emulsion test.

In the froth test, 2 ml of the aqueous extract was mixed with 5 ml of distilled water in a test tube. The mixture was shaken vigorously. A stable froth on standing indicates the presence of saponins.

In the emulsion test, 3 drops of groundnut oil, was added to the aqueous extract mixed with 5 ml of distilled water and shaken well. Formation of emulsion indicates the presence of saponins.

#### ***Test for the Presence of Tannin***

The presence of tannins in the samples was determined using the method described by Harborne (1973).

2 ml of the aqueous extract filtrate and 3 ml distilled water was put into a test tube. A few drops of 0.1% ferric chloride was added to the mixture. The formation of a very dark precipitate indicated presence of tannin.

### **Quantitative Determination of the Phytochemical Constituents of the Plant Samples**

#### ***Alkaloid Determination***

The determination of the concentration of alkaloid in the leaves of the plants was carried out using the alkaline precipitation gravimetric method described by Harborne (1973).

5 g of the powdered sample was soaked in 20 ml of 10% ethanolic acetic acid. The mixture was stood for four (4) hours at room temperature. Thereafter, the mixture was filtered through Whatman filter paper (No 42). The filtrate was concentrated by evaporation over a steam bath to  $\frac{1}{4}$  of its original volume. To precipitate the alkaloid, concentrated ammonia solution was added in drops to the extract until it was in excess. The resulting alkaloid precipitate was recovered by filtration using previously weighed filter paper. After filtration, the precipitate was washed with 9% ammonia solution and dried in the oven at 60°C for 30 minutes, cooled in a dessicator and reweighed. The process was repeated two more times and the average was taken. The weight of alkaloid was determined by the differences and expressed as a percentage of weight of sample analyzed as shown below.

$$\% \text{ Alkaloid} = \frac{W_2 - W_1}{\text{Weight of sample}} \times \frac{100}{1}$$

Where:-

$W_1$  = weight of filter paper

$W_2$  = weight of filter paper + alkaloid precipitate

#### ***Flavonoid Determination***

The flavonoid content of the leaves of the plant was determined by the gravimetric method as was described by Harborne (1973).

5g of the powdered sample was placed into a conical flask and 50ml of water and 2ml HCl solution was added. The solution was allowed to boil for 30 minutes. The boiled mixture was allowed to cool before it was filtered through Whatman filter paper (No 42). 10ml of ethyl acetate extract which contained flavonoid was recovered, while the aqueous layer was discarded. A pre weighed Whatman filter paper was used to filter second (ethyl-acetate layer), the residue was then placed in an oven to dry at 60°C. It was cooled in a dessicator and weighed. The quantity of flavonoid was determined using the formular.

$$\% \text{ Flavonoid} = \frac{W_2 - W_1}{\text{Weight of Sample}} \times \frac{100}{1}$$

Where:-

$W_1$  = Weight of empty filter paper

$W_2$  = Weight of paper + Flavonoid extract

### ***Determination of Phenols***

The concentration of phenols in the leaves of the plants was determined using the folin- cio Caltean colorimetric method described by Pearson (1976).

0.2 g of the powdered sample was added into a test tube and 10ml of methanol was added to it and shaken thoroughly the mixture was left and to stand for 15 minutes before being filtered using Whatman (No42) filter paper. I ml of the extract was placed in a text-tube and I ml folin-cio Caltean reagent in 5ml of distilled water was added and color was allowed to develop for about 1 to 2 hours at room temperature. The absorbance of the developed colour was measured at 760 nm wave. The process was repeated two more times and an averaged taken. The phenol content was calculated thus.

$$\% \text{ Phenol} = 100 / w \times AU / AS \times C / 100 \times VF / VA \times D$$

Where,

W= weight of sample analyzed

AU= Absorbance of test sample

AS= Absorbance of standard solution

C= concentration of standard in mg/ml

UF= total filtrate volume

VA= Volume of filtrate analyzed

D= Dilution factor were applicable

### ***Determination of Saponins***

The saponin content of the sample was determined by double extraction gravimetric method (Harborne, 1973).

5 g of the powered sample was mixed with 50 ml of 20% aqueous ethanol solution in a flask. The mixture was heated with periodic agitation in water bath for 90 minutes at 55°C; it was then filtered through what man filter paper (No42). The residue was extracted with 50 ml of 20% ethanol and both extracts were poured together and the combined extract was reduced to about 40 ml at 90°C and transferred to a separating funnel where 40 ml of diethyl ether was added and shaken vigorously. Separation was by partition during which the ether layer was discarded and the aqueous layer reserved. Re extraction by partitioning was done repeatedly until the aqueous layer become clear in color. The saponins were extracted, with 60 ml of normal butanol. The combined extracts were washed with 5% aqueous sodium chloride

(NaCl) solution and evaporated to dryness in a pre-weighed evaporation dish. It was dried at 60° C in the oven and reweighed after cooling in a dessicator. The process was repeated two more times to get an average. Saponin content was determined by difference and calculated as a percentage of the original sample thus

$$\% \text{ Saponin} = \frac{W_2 - W_1}{\text{Wt of sample}} \times \frac{100}{1}$$

Where

$W_1$  = weight of evaporating dish

$W_2$  = weight of dish + sample

### ***Steroid Determination***

The steroid content of the leaves of the plants was determined using the method described by Harborne (1973).

5g of the powdered sample was hydrolysed by boiling in 50 ml hydrochloric acid solution for about 30 minutes. It was filtered using Whatman filter paper (N042) the filtrate was transferred to a separating funnel. Equal volume of ethyl acetate was added to it, mixed well and allowed separate into two layers. The ethyl acetate layer (extract) recovered, while the aqueous layer was discarded. The extract was dried at 100°C for 5 minutes in a steam bath. It was then heated with concentrated amyl alcohol to extract the steroid. The mixture becomes turbid and a reweighed Whatman filter paper (N042) was used to filter the mixture properly. The dry extract was then cooled in a dessicator and reweighed. The process was repeated two more times and an average was obtained.

The concentration of steroid was determined and expressed as a percentage thus

$$\% \text{ Steroid} = \frac{W_2 - W_1}{\text{Wt of sample}} \times \frac{100}{1}$$

Where,

$W_1$  = weight of filter paper.

$W_2$  = weight of filter paper + steroid

### ***Tannin Determination***

The tannin content of the leaves of the plants was determined using the Folin Dennis spectrophotometric method described by Pearson (1976).

2 g of the powdered sample was mixed with 50 ml of distilled water and shaken for 30 minutes in the shaker. The mixture was filtered and the filtrate used for the experiment. 5 ml of the filtrate was measured into 50 ml volume flask and diluted with 3 ml of distilled water. Similarly 5 ml of standard tannic acid solution and 5 ml of distilled water was added separately. 1 ml of Folin- Dennis reagent was added to each of the flask followed by 2.5 ml of saturated sodium carbonate solution. The content of each flask was made up to mark and incubated for 90 minutes at room temperature. The absorbance of the developed colour was measured at 760 nm wave length with the reagent blank at zero. The process was repeated two more times to get an average. The tannin content was calculated as shown below

$$\% \text{ tannin} = 100/W \times AY / AS \times C/100 \times VF/VA \times D$$

Where,

W = weight of sample analysed



AY=Absorbance of the standard solution

C= Concentration of standard in mg /ml.

VA= volume of filtrate analysed

D= Dilution factor where applicable

## **Determination of Antimicrobial Activity**

### ***Preparation of Plant Extracts***

The ethanolic extracts of the leaves of *Dalium guineense*, *Vitex doniana* and *Dennittia tripetala* were prepared using the method of Ijeh et al., (2005).

Fifty grams of the powdered sample were soaked in 200ml of absolute ethanol and allowed to stand for 24 hours. They were filtered using Whatman No1 Filter Paper. The filtrates were evaporated to dryness with rotary evaporator at 40°C to thick residues. The residues were dissolved in deionised water to obtain the desired plant extracts for the antimicrobial tests.

### ***Preparation of Innocular***

The human pathogens; *Escherichia coli*, *Staphylococcus aureus*; *Salmonella typhi* and *Pseudomonas aeruginosa* used in the research were obtained from the stock culture of the Microbiology Laboratory, Federal Medical Centre, Umuahia, Abia State, Nigeria. Viability test of each isolate was carried out by resuscitating the organism in buffered peptone broth and thereafter sub-cultured into nutrient agar medium and incubated at 37°C for 24 hours.

### ***Antimicrobial Activity Test***

The sensitivity of the test organism to the ethanolic extracts of the leaves of *D. guineense*, *V. doniana* and *D. tripetala* was carried out using the diffusion method described by Ebi and Ofoefule (1997).

20ml of the molten nutrient agar was seeded with 0.2ml of broth culture of the test organisms in sterile Petri dishes. The Petri dishes were rotated slowly to ensure a uniform distribution of the organisms. They were left to solidify and dish cups of 8.0mm diameter were made in the agar using a sterile Pasteur pipette. The Petri-dishes were allowed to stand for about 30 minutes at room temperature to allow for the proper diffusion of the extracts to take place. The plates were then incubated at 37°C for 24 hours. The zones of inhibition in millimetres were measured and recorded.

The test was carried out in the Laboratory of the Department of Plant Science and Biotechnology, Michael Okpara University of Agriculture, Umudike, Umuahia, Abia State, Nigeria.

### **Minimum Inhibitory Concentration (MIC) Test**

The agar dilution method described by Baron and Finegold (1990) was used to determine the minimum inhibitory concentration.

Six grams of nutrient agar were dissolved in 250ml of distilled water in a conical flask. After sterilization, the nutrient agar was poured into sterilized Petri dishes to solidify. The microorganisms were introduced into the wells using swap sticks. Extracts of 5mg/ml, 15mg/ml, 20mg/ml and 25mg/ml were made from the original test samples. The Petri dishes were then placed in the incubator at 37°C for 24 hours. The inhibition zones in millimetres were measured and recorded.

## Statistical Analysis

The tests were carried out in triplicate; data obtained were analysed using mean and standard deviation.

## RESULTS AND DISCUSSIONS

The result of the phytochemical screening and the antimicrobial activity of the leaves of *Dialium guineense*, *Vitex doniana*, and *Dennettia tripetala* are summarized in tables 1- 4.

The phytochemical screening of the leaves of *D. guineense*, *V. doniana* and *D. tripetala* revealed that they contain alkaloids, flavonoids, phenols, saponin, steroids and tannin (table 1) Presence of these phytochemicals had also observed in other plants ( Osuagwu et al, 2007; Iniaghe et al., 2009; Omoyeni and Aluko, 2010). The presence of these phytochemicals in the leaves of these plants confer on them their medicinal value (Gill, 1992; Arbonnier, 2004; NNMDA, 2008; Vadivu et al., 2008).Pharmaceutical and therapeutic values of plants and their products lie on the presence of these phytochemicals in them (Edeoga *et al.*, 2005; Bishnu *et al.*,2009). The percentage concentration of alkaloids, flavonoids, phenols, saponin, steroids and tannins in the leaves of these plants are summarized in table 2. The phytochemical content of the leaves of the plants are as follows: alkaloid (1.21-2.42%), flavonoids (0.18-0.50%), phenols (0.08-0.14%), saponin (0.90-1.23%), steroid (0.035-0.14%) and tannins (0.169-0.50%).The results indicate that the leaves of these plants have appreciable amount of these phytochemicals, hence their medicinal value. *D. tripetala* had more alkaloid content (2.42%) when compared with those of *V. doniana* (1.41%) and *D. guineense* (1.21%). The flavonoid content of the leaves plants are relatively low; *D. guineense* (0.31%), *V. doniana* (0.50%) and *D. tripetala* (0.18%). The least amount of phytochemicals in these three plants was steroids (0.035-0.145%).On the other hand, the highest occurring phytochemical was alkaloid (1.21-2.42%). Variations in the concentration of phytochemicals in plants were also observed by other researchers (Ganjewala *et al.*, 2009; Ayoola and Adeyeye, 2010).

The results of the antimicrobial activity of the ethanolic extracts of the leaves of *D. guineense*, *V. doniana* and *D. tripetala* on *Escherichia coli*, *Staphylococcus aureus*, *Salmonella typhi* and *Pseudomonas aeruginosa* are summarized in Tables 3 and 4.

The ethanolic extracts of the leaves of *D. guineense*, *V. doniana* and *D. tripetala* had antimicrobial activity on all the human pathogens used in the study (Table 3). The inhibition zone ranged from 0.1 mm to 14 .00 mm. Leaf extracts of *V. doniana* had the highest inhibitory effect on *E. coli* (14.00mm) when compared to those of *D. tripetala* (10.0mm) and *D. guineense* (0.10mm). *S. aureus*, *S. typhi* and *P. aeruginosa* were more sensitive to the leaf extracts of *D. tripetala* (11.60mm) when compared to the leaf extracts of *D. guineense* and *V. doniana*. The medicinal value of the leaves of these plants stems from their ability to inhibit these human pathogens. The use of the leaves of these plants for treatment of diseases has been reported (Gill, 1992; Arbonnier, 2004; NNMDA, 2008). The antimicrobial activity of the leaves of other plants has been reported (Arshad *et al.*, 2010; Kamba and Hassan 2010 Koche *et al.*, 2011). The ability of the extracts to inhibit the growth of the test microorganisms might be as a result of the presence of bioactive substances (alkaloids, flavonoids, phenols saponins, steroids and tannins in their leaves (Bishnu *et al.*, 2009; Iniaghe *et al.*, 2009; Omoyeni and Aluko, 2010). There is an observed relationship between the concentration of the extracts and the rate of inhibition of the growth of the pathogens. There was corresponding increase in the rate of inhibition of the pathogens as the concentration of the extracts increased. This trend was also reported by other researchers (Valarmathy *et al.*,



2010; Subban *et al.*, 2011). The minimum inhibitory concentration of the ethanolic extracts of the leaves of the three plants ranged from 2.00 to 19.33 mg/ml (table 4).

This study showed that the leaves of *D. guineense*, *V. doniana* and *D. tripetala* used for the investigation have high phytochemical content and have antimicrobial activity on the human pathogens used in this research. This shows that they are of high medicinal value. Thus could be exploited to be used in the formation of cheap alternative antimicrobial drugs which will be used to cure and control human infectious diseases.

Key: + = presence

- = absence

**Table 1. The qualitative analysis of the phytochemicals in the leaves of *Dialium guineense*, *Vitex doniana* and *Dennettia tripetala***

Plant species	Alkaloids	Flavonoids	Phenols	Saponins	Steroids	Tannins
<i>Dialium Guineense</i>	+	+	+	+	+	+
<i>Vitex Doniana</i>	+	+	+	+	+	+
<i>Dennettia Tripetala</i>	+	+	+	+	+	+

**Table 2. The percentage alkaloids, flavonoids, phenols, saponins, steroids and tannins content of the leaves of *D. guineense*, *V. doniana* and *D. tripetala*.**

Plant Species	Alkaloids	Flavonoids	Phenols	Saponin	Steroids	Tannins
<i>D. Guineense</i>	1.12 ± 0.01	0.31 ± 0.01	0.142 ± 0.01	1.25 ± 0.01	0.145 ± 0.01	0.577 ± 0.01
<i>V. Doniana</i>	1.41 ± 0.01	0.50 ± 0.01	0.08 ± 0.001	0.90 ± 0.02	0.035 ± 0.01	0.159 ± 0.03
<i>D. Tripetala</i>	2.42 ± 0.05	0.18 ± 0.03	0.095 ± 0.005	0.90 ± 0.01	0.048 ± 0.01	1.97 ± 0.039

**Table 3. The antimicrobial activity of the ethanolic extracts of the leaves of *D. guineense*, *V. doniana* and *D. tripetala* on *Escherichia coli*, *Staphylococcus aureus*, *Salmonella typhi* and *Pseudomonas aeruginosa*.**

Pathogenic	<i>D. guineense</i>	<i>V. doniana</i>	<i>D. tripetala</i>
Organism	Zone	Of	Inhibition (mm)
<i>E. Coli</i>	0.10 ± 0.00	14.00 ± 1.00	10.00 ± 1.00
<i>S. Aureus</i>	0.16 ± 0.03	11.00 ± 0.05	11.60 ± 1.15
<i>S. Typhi</i>	0.33 ± 0.11	11.00 ± 0.01	11.60 ± 1.00
<i>P. Aeruginosa</i>	7.60 ± 1.20	5.70 ± 0.60	10.30 ± 1.53

**Table 4. The minimum inhibitory concentration (mg/ml) of the ethanolic extracts of the leaves of *D. guineense*, *V. doniana* and *D. tripetala* on *E. coli*, *S. aureus*, *S. typhi* and *P. aeruginosa*.**

Pathogenic organisms	<i>D. Guineense</i>					<i>V. Doniana</i>					<i>D. Tripetala</i>				
	12.25	25	50	100	200	12.25	25	50	100	200	12.25	25	50	100	200
<i>MIC</i>	12.25	25	50	100	200	12.25	25	50	100	200	12.25	25	50	100	200
<i>E. coli</i>	2.3	5.3	11.3	12.7	14.3	9.0	11.3	14.1	18.7	17.2	3.0	12.3	14.0	18.3	19.3
<i>S. aureus</i>	6.1	8.3	9.3	9.3	11.4	9.0	10.7	15.0	17.0	19.2	9.0	10.0	13.0	16.0	17.0
<i>S. typhi</i>	11.0	10.0	14.0	12.6	14.7	3.0	9.0	12.3	12.5	15.0	5.0	10.0	7.0	11.0	12.1
<i>P. aeruginosa</i>	10.0	10.7	18.7	15.3	16.7	2.0	5.3	7.0	11.3	18.0	6.0	10.6	12.3	14.0	15.0

MIC = Minimum Inhibitory Concentration

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