Qualitative and Quantitative Phytochemical Screening, and Antibacterial Activity of *Phyllanthusamarus* Plant Extracts,

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Abstract: Phytochemical screening and antimicrobial activities of water, ethanol and methanol extracts of parts of *Phyllanthusamarus* plant were investigated. Qualitative results from the different extracts, showed the presence of alkaloids, tannins phenols, saponins and flavonoids. However, phenol was absent in ethanol seed extract, but present in other crude extracts obtained with water and ethanol. The percentage of alkaloids in the aqueous extracts of leaf, seed, stem and root showed that the quantity in seed (2.40 ±0.23) and leaf (2.23 ±0.15) were higher than those of stem (0.40±0.00) and root (1.68±0.00). The phytochemical constituents of ethanol extracts obtained from different parts of the plant varied. However, the quantity in root for the different phytochemical compounds considered were more than that present in the other parts of the plant, alkaloid(5-00±0.11), flavonoid (3.00±0.12); tannin(3.82±0.11) ; saponins (4.00±12) and phenols(2.40±0.12). The estimated quantity in crude methanol extracts of *P. amarus* parts showed that alkaloids present in root (5.00±0.11) was higher compared with the other parts; leaf (3.00±0.60), seeds (2.00±0.00); and stem(2.40±0.00). Except for stem where saponin as 1.40±0.23. The saponin content in other part were higher compared with-other phytochemicals. Water was the best extractant of saponin from root, while ethanol and methanol were better extractant of alkaloids from root. The crude extracts of *Phyllanthusamarus* plant parts were potent against *Escherichia coli* and *Staphylococcus aureus* strains. The aqueous extract of leaf produced the highest zone of inhibition against 25.00 ±1.67mm for *E. coli* strain 7.

Keywords: *Phyllanthusamarus*, plant parts antibacterial, quantitative phytochemical, three solvents

Introduction

The number of angiosperms and gymnosperms on this planet is estimated at 250,000 (Ayensu and DeFilipps,1978, Borris,1996) having 500,000 at upper level (Tipo and Stern, 1977)and 215,000 at lower level (Cronquist, 1988). Out of these, records show that only 6% have been screened for biological activity and 15% evaluated phytochemically (Verpoorte , 2000), thus the need for more search for bioactive principle among plants to discover their potential medicinal value for use by man. Medicinal plants contain compounds which are harmful to microorganisms, thereby having inhibiting effect on the microbes or eradicating the microorganism responsible for infectious diseases. Substances present in medicinal plants include the basic metabolites, phenolic compounds, terpenes, steroids, alkaloids, glycosides, and many secondary metabolites (Bargah, 2015). In many cases, these substances serve as plant defense mechanism against predation by microorganisms, insects and herbivores. *Phyllanthusamarus*is a tropical herbal plant useful ethnobotanically for treatment of microbial and other diseases.. The aim of study was to determine qualitatively and quantitatively the phytochemicals and antibacterial activity of extracts of *Phyllanthusamarus* plant parts.

Experimental

Preparation of extracts

Phyllanthusamarus was collected from Abraka and Obiarukuarea, Delta State, Nigeria and was identified at Botany Department, Delta State University, Abraka. The extraction of leaves, stem, seeds and roots was carried out using water, ethanol and methanol. All crude extracts were obtained using cold maceration method except for seeds where soxhlet extraction was used. One hundred grams of leaves was soaked in distilled water, ethanol and methanol separately for three days, filtered with Whatmann No. 1 Filter paper and concentrated invacuo with rotary evaporator (Model-Buch- Labortechnik type R 11, Switzerland) . Further drying was done using oven at 40°C. The extract from water was concerntrated using water bath regulated to 70-80°C. The extracts obtained were then stored and used for qualitative and quantitative phytochemical analysis and antimicrobial screening.

Qualitative Phytochemical Screening

Test for alkaloids:

Dragendroff's Test: Five hundred milligrams each of dried ethanol extracts were weighed and re-extracted with (5 ml) of five percent Hydrocholoric acid (HCl). The HCl extracts were then filtered with Whatman No. 1 filter paper so as to have a clear solution and also to prevent false results. To 2.5 ml of the filtrate, few drops of Dragendorff's reagent (potassium bismuth iodide) were added. The presence of alkaloids was indicated by an orange coloured precipitation (Harborne, 1998; Trease and Evans, 1989).

Test for saponins

Frothing Test: Five hundred miligrams of powered plant material was shaken with water in a test-tube. Persistent frothing was taken as preliminary evidence of saponin (Odebiyi and Sofowora 1978).

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Test for cardiac glycosides

Keller- Killiani test: One hundred milligrams of extract was dissolved in 1 ml of glacial acetic acid containing one drop of ferric chloride solution. This was then underlayered with 1 ml of concentrated sulphuric acid. Observation of reddish brown colouration at the junction of two layers and the bluish green colour in the upper layer shows the presence of cardiac glycosides (Harborne, 1998)

Test for tannins

Five hundred milligrams each of the extract was boiled with 10 ml of distilled water in a test tube and filtered. Ferric chloride reagent was added to the filtrate. Appearance of brownish green or blue-black colouration showed the presence of tannins (Trease and Evans, 1989).

Test for flavonoids

Five millilitres of extract was treated with few drops of sodium hydroxide solution. Development of yellow colouration within three minutes indicated the presence of flavonoids (Evans, 2006).

Quantitative phytochemical determination

Alkaloid

Alkaloid was determined by the method of Harborne (Harborne, 1973). About 5g of sample was 250 ml beaker and 200 ml of 10% acetic acid in ethanol was added, then covered and allowed to stand for 4h. Filtrate obtained was reduced to a quarter of the original volume by heating on a water-bath. Concentrated ammonium hydroxide was added drop wise until precipitation was observed. This was allowed to stand and the precipitate was collected and washed with dilute ammonium hydroxide and then filtered. The residue (alkaloid) derived was dried and weighed.

Saponin

The method of Obadoni and Ochuko, (2001)was adopted. Twenty grams of each ground plant material was put in a conical flask and 100ml of 20% aqueous ethanol was added. The sample was heated on a water bath for 4h with continuous stirring at 55°C. The mixture was filtered and the residue re-extracted with another 200ml 20% ethanol. The combined extracts was reduced to 40ml on a water bath at 90°C. The concentrate was transferred in to a separating funnel and 20 ml of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. 60ml of n-butanol was added. The combined extract was washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was evaporated over water bath. The sample was dried in the oven to a constant weight .The percentage of the saponin was calculated

Phenols

The phenolic component was determined by boiling fat free sample in 50ml of ether for 15min. To 5ml of extract was added 10ml of distilled water in a 50ml flask. 2ml ammonium hydroxide solution and 5ml of concentrated amyl alcohol were added. The sample was made up to mark and left to stand for half an hour for development of colour Absorbance was measured at 505nm using a spectrophotometer.

Tannin

Determined using method of Van-Burden and Robinson (1981). 0.5g of sample was put into 50ml plastic bottle, 50ml of distilled water was added and shaken for 1hr in a mechanical shaker. This was filtered in to 50ml volumetric flask and made up to mark. To 5ml filtrate pipetted into a testtube,2ml 0.1 M FeCl3 in 0.1 N HCl and 0.008M Potassium ferrocyanide. Absorbance was measured at 120mn within 10 minutes.

Flavonoid

The method of Bohm and Kocipai-Abyazan, (1974). 10g of plant material was extracted repeatedly with 100ml of 80% aqueous methanol at room temperature. The whole solution was filtered with through Whatman filter paper no 42 (125mm). The filtrate was later transferred into a crucible and evaporated into dryness over a water bath and weighed to a constant weight.

Antimicrobial Sensitivity Testing

The bacterial cultures previously identified were stored as agar slants in McCartney bottles at 4°C in the refrigerator until required. The bacterial was subcultured from agar slant onto nutrient agar when required. Sub-culturing of strains were repeated every fourteen to twenty-one days throughout the experimental period.

The agar well diffusion method as was employed in the determination of the antibacterial activity of plant extracts. Standardized (0.5 McFarland) 24hr old culture suspension (0.2 ml) was inoculated on sterile Muller Hinton agar plates and allowed to set. With the aid of a sterile cork borer, wells of diameter 5 mm were bored in agar plates. Each respective plant extract (0.5 ml), was dispensed into the well and then allowed to set for 1 hour. Plates were then incubated aerobically for 24hours at 37 °C. Diameters of the zones of growth inhibition formed on the agar surface in triplicates were measured using a millimeter rule and the mean diameter of the zones of inhibition calculated and recorded.

Determination of Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC)

Minimum inhibitory concentrations of the extracts were determined by two- fold serial dilution method. The initial stock concentration of the plant extract (500 mg/ml) was diluted using double dilution method by transferring 5 ml of the sterile plant extract (stock solution) into 5 mlof sterile Mueller-Hinton broth to obtain 250mg/ml concentration. This was further diluted to obtain other concentrations 125, 62.5, 31.25, 15.625, 7.8125 and 3.91 (mg/ml).

Isolates were standardized to 0.5 McFarland's standard. The minimum inhibitory concentration (MIC) was determined by adding 2 ml of a specific concentration of plant extract and 0.1 ml of standardized test organism. After incubation for 24 hours at 37 °C the least concentration of the plant extracts with no visible growth was taken as the MIC. The Minimum Bactericidal

Concentration (MBC) was determined bystreaking out from the last broth that showed visible growth (turbid) and all cultures in which there was no growth (no turbidity) on to nutrient agar. The lowest concentration that prevented bacterial growth after incubation was recorded as the minimum bactericidal concentration.

Results and Discussion

Table1 : Qualitative phytochemical extracts of *Phyllanthusamarus* with aqueous, ethanol and methanol

Qualitative Test	Aqueous				Ethanol				Methanol			
	Leaf	Seed	Stem	Root	Leaf	Seed	Stem	Root	Leaf	Seed	Stem	Root
Alkaloid	+	+	+	++	++	+	++	+++	++	++	++	+++
Flavonoid	+	++	+	++	+	++	+	+	++	+	+	+
Tannins	++	+	+	++	+++	+	+	++	++	+	+	++
Saponins	+++	+	+	+++	++	+	++	++	++	++	+	+
Phenol	+	+	+	+	+	-	++	++	+	+	++	++

key
+++ = very highly present
++= Highly present
+= Fairly present
- = Absent

Table 2 : Quantitative phytochemical extracts o. *Phyllanthusamarus* with aqueous, ethanol and methanol

Tests Plant parts	Alkaloid(%)	Flavonoid(%)	Tannin (%)	Saponin (%)	Phenol (%)
AQUEOUS					
Leaf	2.13±0.15	1.80±0.60	3.30±0.00	5.57±0.32	1.00±0.00
Seed	2.40±0.23	3.30±0.17	1.20±0.12	5.0±0.12	2.87±0.70
Stem	0.40±0.00	0.30±0.00	1.00±0.00	0.50±0.00	1.60±0.10
Root	1.68±0.00	1.73±0.17	3.40±0.23	7.00±0.00	0.50±0.00
ETHANOL					
Leaf	3.10±0.60	1.50±0.00	3.20±0.00	2.50±0.12	1.00±0.00
Seed	1.00±0.00	1.50±0.00	0.40±0.00	1.20±0.12	1.00±0.00
Stem	2.40±0.23	1.00±0.00	1.50±0.12	2.30±0.00	2.27±0.00
Root	5.00±0.11	3.00±0.12	3.82±0.01	4.00±0.12	2.40±0.12
METHANOL					
Leaf	3.00±0.60	1.87±0.24	1.50±0.12	3.50±0.00	1.80±0.12
Seed	2.00±0.00	1.60±0.12	0.80±0.00	4.23±0.13	0.60±0.00
Stem	2.40±0.00	0.80±0.12	3.20±0.00	1.40±0.23	2.80±o.12
Root	5.00±0.11	1.20±0.12	2.40±0.21	3.33±0.24	3.00±0.12

Table 3 :Mean diameter of inhibition zones of methanol extracts of various parts of *Phyllanthusamarus*

Bacteria	Plant part	concentration of extracts mg/ml				
		31.25	62.50	125	250	500
Escherichia coli 1	Leaf	7.00±0.33	9.33±0.88	16.17±2.33	17.00±0.33	18.33±1.67
Escherichia coli 2	Leaf	7.00±0.33	9.00±0.57	10.67±1.20	14.00±0.33	17.33±1.67
Escherichia coli 4	Leaf	8.00±0.58	10.67±0.67	11.33±1.20	13.00±0.33	16.33±1.67
Escherichia coli 5	Leaf	7.67±0.33	9.00±0.33	11.00±0.67	14.00±0.33	19.33±1.67
Escherichia coli 6	Leaf	7.00±0.58	11.33±0.33	12.00±0.67	11.00±0.58	17.33±1.15
Escherichia coli 7	Leaf	9.67±1.20	13.00±0.67	17.17±2.33	16.00±0.58	20.00±1.33
Escherichia coli 9	Stem	10.00±0.33	12.67±0.88	12.67±0.67	15.00±0.58	16.00±1.00
Staphylococcus aureus 2	Leaf	9.67±1.20	11.33±0.67	12.00±1.15	14.67±0.67	18.00±1.15
Staphylococcus aureus 3	Leaf	8.67±0.67	11.00±1.00	12.00±0.58	13.00±0.58	18.00±1.00
Staphylococcus aureus 5	Leaf	8.00±0.67	10.0±0.33	12.00±0.33	14.67±1.00	19.00±1.33
Staphylococcus aureus 7	Leaf	8.00±0.33	10.00±0.58	11.00±0.88	13.00±0.58	19.00±1.67

Table 4:Mean diameter of inhibition zones of aqueous extracts of various parts of *Phyllanthusamarus*

Bacteria	part of plants	concentrations (mg/ml) 31.25	62.50	125	250	500
Escherichia coli 1	Leaf	12.50±0.58	13.67 ±0.67	16.00 ±0.58	18.0 ±0.880	24.00± 1.33
Escherichia coli 2	Leaf	8.00± 0.33	10.00 ±0.58	12.64±0.66	14.10 1.16±	22.00± 1.79
Escherichia coli 3	Leaf	14.00± 0.58	17.00 ±0.58	18.00± 0.33	20.67 ± 2.18	24.00±01.30
Escherichia coli 5	Leaf	12.00± 0.33	13.00±0.60	17.00± 0.88	19.00± 0.67	23.00± 1.67
Escherichia coli 6	Leaf	12.00±1.00	62.50	125	250	500
Escherichia coli 7	Stem	9.0±0 0.58	13.67 ±0.67	16.00 ±0.58	18.0 ±0.880	24.00± 1.33
	Leaf	12.00± 0.58	10.00 ±0.58	12.64±0.66	14.10 1.16±	25.00± 1.67
Escherichia coli 9	Leaf	8.00± 0.20	15.00 ±1.54	16.00± 1.42	19.00 ±0.88	23.2±0 1.58
Staphylococcus aureus 2	Leaf	8.00 ±0.30	11.001.00	12.00± 1.55	13.00± 1.00	17.00 ±1.33
Staphylococcus aureus 3	Stem	12.67± 0.67	15.00± 0 .58	18.00± 0.00	20.33± 0.33	22. 00 ±1.79
	Leaf	8.67± 0.33	11.00±1.20	13.60± 0.55	16.22± 0.88	24.0±0 1.00
Staphylococcus aureus 4	Leaf	12.00± 0.58	12.00 ±0.67	14.00±1.15	16.32± 0.58	20.00± 1.00
Staphylococcus aureus 5	Leaf	9.00± 1.56	12.00±1.15	13.67± 0.33	14.00± 0.58	18.00±1.58
Staphylococcus aureus 7	Stem	9.00±0.58	12.00± 1.00	13.67± 0.33	18.0±0 0.00	22.00 ±1.00
Staphylococcus aureus	Leaf	8.00 ±0.67	14.00± 0.58	15.00 ±0.58	16.00± 0.58	20.00 ±1.58

Table 5 :Mean diameter of inhibition zones of ethanol extracts of various parts of *Phyllanthusamarus*

Bacteria	plant parts	concentration of extracts mg/ml 31.25	62.50	125	250	500
Escherichia coli 1	Leaf	8.00 ± 1.52	10.00± 0.33	12.67 ± 0.88	14.67± 0.20	16.67± 1.30
Escherichia coli 2	Stem	12.33± 0.33	12.00 1.15±	15.33 ± 1.45	18.33± 1.67	21.00 ± 1.70
	Seed	12.6± 70.67	13.00 ± 1.15	14.67± 1.33	16.00± 2.33	16.00± 1.76
Escherichia coli 4	Stem	12.00± 0.58	13.00 ± 0.58	12.00± 0.00	16.67 ± 0.88	21.00± 1.62
	Seed	10.00± 0.58	10.67± 0.33	12.6± 7 0.33	15.33 ± 0.33	20.00± 1.33
Escherichia coli 5	Stem	9.67± 0.33	13.33 ± 0.67	15.33± 0.67	16.00 ± 0.67	20.00± 1.33
	Seed	7.00± 0.33	9.00 ± 1.15	10.33± ± 0.33	12.33 ± 0.33	16.00± 1.20
	Leaf	7.00± 0.57	8.00± 0.33	10.00± 0.33	15.00 ± 0.58	16.00± 1.18
Escherichia coli 6	Seed	12.33± 0.33	13.00± 1.15	12.00± 0.57	11.00 ± 0.67	17.00± 1.33
	Leaf	7.00± 1.00	9.00± 0.58	12.00± 1.45	15.00 ± 0.88	18.00± 1.58
Escherichia coli 7	Stem	11.00± 0.67	9.00± 0.58	9.67± 0.33	11.00± 0.67	18.00 ± 1.57
	Seed	7.00± 1.00	9.00 ± 1.00	12.00± 0.88	15.00± 1.15	19.00± 2.90
	Root	6.0± 0 1.00	7.67 ± 0.33	13.00± 0.88	15.00± 0.88	19.00± 1.33
Escherichia coli 9	Stem	8.67± 0.88	9.33± 0.58	12.67± 0.33	14.00± 0.33	15.00± 1.62
	Seed	10.00± 0.58	12.00 ± 1.00	12.33± 0.28	15.33 ± 1.15	18.00± 1.88
	Root	8.00± 0.00	9.67 ± 0.33	11.33± 0.88	12 33± 0.88	18.00± 1.00
Staphylococcus aureus 2	Seed	6.00 ± 1.00	8.33± 067	13.00± 0.58	17.00±. 0.00	19.00± 1.00
	Root	8.67± 1.33	10.67 ± 0.58	14.00± 0.58	16.00 0.88±	20.00± 1.00
Staphylococcus aureus 3	Stem	9.00± 0.58	9.00± 0.00	14.67± 1.20	16.67 ± 1.33	19.00 ± 1.00
	Seed	7.67± 0.88	8.33 ± 0.88	12.33± 0.33	15.00± 0.58	20.0± 0 1.58
	Leaf	11.00± 0.58	12.00 ± 1.00	10.00± 0.58	13.33± 0.67	16.00± 1.33
Staphylococcus aureus 4	Stem	8.00 ± 0.58	11.60 ± 0.58	12.00± 0.88	13.00± 1.00	16.60 ± 1.61
	Root	7.00± 0.58	9.00± 0.67	10.00± 0.58	12.00 ± 1.20	14.00± 1.17
Staphylococcus aureus 5	Leaf	8.32 ± 0.88	10.00± 0.88	12.00± 0.67	14.00± 1.54	18.00± 1.67
Staphylococcus aureus 7	Stem	9.00 ± 0.58	10.00 ± 1.15	12.00± 1.10	12.00 ± 0.57	18.00± 1.88

Seed	11.00± 0.58	11.00 ± 1.45	11.33± 0.67	14.00 ± 0.67	20.00± 1.67
Leaf	8.00 ± 0.67	10.00 ± 0.58	11.33± 0.67	14.00± 1.15	15.0± 0 1.00

Table 6: Inhibitory and Bactericidal Concentration of *Phyllanthusamarus* plant parts.

Plant/Extract

		E. coli strains							*S. aureus* strains				
		1	2	4	5	6	7	9	2	3	4	5	7
***P. amarus*	MIC	250	250	-	125	2505	-	250	-	250	-	125	250
	MBC	500	500	-	250	00	-	500	-	500	-	250	500
**P amaus*	MIC	125	125	125	125	250	125	-	125	125	125	125	125
	MBC	250	250	250	250	500	250	-	250	250	250	250	250
**P. amarus*	MIC	125	125	125	125	125	125	125	125	250	-	125	125
	MBC	250	250	250	250	250	250	250	250	500	-	250	250
****P amarus stem*	MIC	-	125	125	125	250	250	-	-	125	250	250	-
	MBC	-	250	250	250	500	500	-	-	250	500	500	-
**P. amaruscstem*	MIC	-	-	-	-	250	-	-	-	125	-	-	250
	MBC	-	-	-	-	500	-	-	-	250	-		500
****P amarus seed*	MIC	-	125	125	125	125	125	125	125	125	-	-	125
	MBC	-	250	250	250	250	250	250	250	250	-	-	250
***P. amarus root	MIC	-	-	-	-	250	-	-	-	125	-	-	125
	MBC	-	-	-	-	500	-	-	-	250	-	-	125

= No activity, MIC Minimum Inhibitory Concentration; (mg/ml) MBC Minimum Bactericidal concentration(mg/ml)

*Aqueous ** Methanol *** Ethanol

The qualitative phytochemical composition of *Phyllanthusamarus* plant parts obtained from three different solvents is shown in Table 1. Alkaloids, flavonoids, tannin phenols and saponin were present in the different extracts, obtained from different parts of *P. amarus*. However, phenol was absent in ethanol seed extract, but present in other crude extracts obtained from water and ethanol. Table 2 shows the percentage quantities of crude phytochemical constituents in *P. amarus* plants parts obtained with aqueous, ethanol and methanol. The percentage quantity of alkaloids in the aqueous extracts of leaf, seed stem and root showed that the quantity in seed (2.40 ±0.23) and leaf (2.23 ±0.15) were higher than that of stem (0.40±0.00) and root (1.68±0.00). Similar trend was observed in the aqueous extracts for flavonoid. The quantity in seeds (3.30±0.17) was more than that obtained from other parts. Contrastingly, the quantity of tannins present in leaf (3.30±0.17) and root (3.40±0.23) were higher than those in seed (1.20) and stem (1.00±0.00). The quantity of saponin in all parts were considerably high (leaf, 5.57±0.32; seeds, 5.00±1.12; root, 7.00±0.00), except stem (0.5±0.00) which was low. Mean while, phenol constituent was low in all parts except in seeds (2.87±0.70).

The phytochemical constituents of extracts of ethanol obtained from different parts of the same plant varied. However, the quantity in root for the different phytochemical compounds considered were more than that present in the other parts of the plant, alkaloid(5-00±0.11), flavonoid (3.00±0.12); tannin(3.82±0.11), saponins (4.00±12) and phenols(2.40±0.12).

Considering the quantity of crude extracts of methanol of *P. amarus*, alkaloid present in root (5.00±0.11) was higher compared with the other parts (leaf:3.00±0.60);(seeds:2.00±0.00); and (stem: 2.40±0.00). Tannin was higher in stem (3.20±0.00) than in other parts. Contrastingly, for saponins, the constituents was lowe (1.40±0.23) in stem than that obtained from the other parts. Flavonoid was more in leaf (1.87±0.24)than in the other parts of plant. Phenolic constituent was higher in root (3.00±0.12) compared with the others plant parts.

The results of the antimicrobial activity of the extracts from the various parts of the plant is presented in Tables 3-5. Methanolic extract was potent against *Escherichia coli* strains 1,2,4,5,6, 7 and 9. Also the extract was potent against *Staphylococcus aureus* strains 2,3,5 and 7.*E. coli*7was the most sensitive organism to the extract showing inhibition zone of 20.00±1.33mm (Table3). Table 4 shows the mean diameter of inhibition zones of aqueous extract of the parts of *P. amarus*. Similar trend was observed in that *E. coli* strains were susceptible to the leaf only except *E. coli* strain 9 which was sensitive to the stem. *Staphylococcus aureus* strains were susceptible to leaf and stem. The highest zone of inhibition was 25.00 ±1.67mm for *E. coli* strain 7 and the lowest zone of inhibition was 14.0±1.58mm for *S. aureus* strain 3. Ethanol extract was also potent against strains *E. coli* and *S. aureus* however, *E. coli* strain 5 and *S.aureus*strain3 were susceptible to stem, seed and leaf. *Escherichia coli*

strain 7 was sensitive to stem, seed and root. These organisms (*E.coli* 5,7*S.aureus* 3) showed more susceptibility to these crude extracts.

Table 6 shows the minimum inhibitory concentration and minimum bactericidal concentration of *Phyllanthusamarus* plant parts. The minimum inhibitory concentration of ethanolic leaf extract was 125mg/ml for *E. coli* strain 5, *S.aureus* strain 5, while the minimum bactericidal concentration for same extract was 500mg/ml for the various organisms except for *Escherichia coli* strain 5, *Staphylococcus aureus* strain 5, where the MBC was 250mg/ml. The MIC for *P. amarus* methanolic leaf extract was 125mg/ml for all organisms except for *E. coli* strain 6 where it was 250mg/ml. The MBC for same extract was 250mg/ml for the various organisms except *E. coli* strain 6 where the MBC was 500mg/ml. Similarly, MIC for *P. amarus* aqueous leaf extract was 125mg/ml for all organisms except *S. aureus* strain 3, where it was 250mg/ml. The MBC was within 125mg/ml to 500mg/ml.

The qualitative, quantitative and antimicrobial activities of *Phyllanthusamarus* plant parts was investigated using water, ethanol and methanol. Fresh leaf paste has wound healing property and used to cure white spots on skin and jaundice. The crude extracts of *Phyllanthusamarus* plant parts were active against the bacteria tested. Both *E. coli* and *S. aureus* previously isolated from clinical specimens of urethritis were susceptible to the extracts in this study. Earlier studies have shown the effect of *P. amarus* on *Escherichia coli* from urinary tract infection (Gbodamosi, 2015). Other researchers have also reported the effect of *P. amarus* water and alcohol extract on microorganisms (Okoli *et al.*,2009). Thus buttressing the reason for use of plant locally.

The phytochemical constituents present in this plant may be responsible the therapeutic properties. Alkaloids are the largest group of plant metabolites comprising of organic heterocyclic nitrogen compounds that are basic forming water-soluble salts. They are usually derived from amino acid (Carson and Hammer, 2010 ;Omajaite *et al.*, 2014). Pharmacological effects produced by alkaloids include antibacterial, local anaesthetic, and hypertensive agent and antimalarial (Awasthi *et al.*, 2015). Alkaloid was present in all the parts of plant except in stem extracted with water thus corroborating the finding of Awashti *et al* (2015) and also Awomakwu *et al.* (2015) who found minimal amount of alkaloids in stem of *P. amarus* screened.

Flavonoids are abundant in photosynthesizing cells. They are large polyphenolic components made by plants (Havteen, 1980; Cushnie and Lamb, 2005).They are potent water-soluble antioxidant and free radical scavengers which prevent oxidative cell damage (Okwu, 2004). Flavonoid have antibacterial and antimalarial functions (Dakoro, 1995) also possess antifungal properties because they inhibit spore germination of plant pathogens and UV –B radiation (Harborne and Williams, 2000). Appreciable flavonoids were present in aqueous seeds and ethanol root extracts. Awasthi et al. (2015)findings contrasted this result , flavonoid was absent in root but present in other parts with the highest amount in stem.

Tannins are polymeric phenolic substances found in nearly all parts of plants. Their molecular action is to complex with proteins. Tannins have stringent feature, fasten the healing of injuries and inflamed mucous membranes. Plants possessing tannins are used to heal hemorrhoids, burn and varicose ulcers (Harborne and Williams, 2000) thus the ethnobotanical use of *P. amarus* for hemorrhoids. The mode of antimicrobial action of tannins may be related to their ability to inactivate microbial enzymes, cell envelopes, adhesions and transport protein (Verma et al., 2014).

Saponin produce foam upon shaken and known as natural detergent, upon hydrolysis, a glycine called sapgenin is produced (Jimoh and Oladji, 2005; Omajiate*et al.*, 2014). Saponins protect plants from parasitic fungi as such, serve as natural antibiotics that help the body to fight against infections and microbial invasion(Haedi, 1964; Okwu, 2005). Other functions of saponin include making bronchi secretion more light, reduce the congestion of bronchi and ease coughing (Okwu, 2005). Appreciable quantity of Saponin was found in all parts of plant using the different solvents, which agrees with the findings of Gbadamosi(2015)which showed that saponin content was more in both *P. amarus* and *P. niruri* than other phytochemicals studied.

Phenols have antimicrobial and antifungal effect (Huang and Ferraro,1992). They have the ability to block specific enzymes that cause inflammation and to prevent disease (Huang and Ferraro,1992). Phenolic compound are well known phytotoxic (Manar, *et al.*, 2006), and exist in free form as ester or as glycoside when combined with sugars, such compounds contribute to the bitter taste, flavour and colour foods (Omaye ,2004). Water was the best extractant of saponin from root, while ethanol and methanol were better extractants of alkaloids from root than water which agrees with Cowan (1999) and Tiwari *et al*(2011).

Conclusion

The qualitative, quantitative and antibacterial activities of *Phyllanthusamarus* plant parts using water, ethanol and methanol was investigated. Results showed the presence of alkaloids, flavonoids, tannins, saponins and phenols. Saponin content of crude extracts obtained from the various plant parts was higher compared with other phytochemicals. Plant parts were potent against *Escherichia coli* and *Staphylococcus aureus* strains. Aqueous extract of leaf produced the highest zone of inhibition on bacteria.

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