

Phytochemical and Antibacterial Activities of Some Selected African Medicinal Plant Extracts on Methicillin-Resistant and Methicillin-Sensitive Staphylococcus Aureus

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Abstract: In recent years, there has been a growing interest in researching and developing new antibacterial agents from various sources to combat microbial resistance. The ethanolic, ethyl acetate, N-hexane and warm water extracts of the leaves of *Piper guineense*, *Ocimum gratissimum* and the stem of *Cassythia filiformis* were examined for antibacterial activities and phytochemical constituents. The phytochemical screening of the samples was carried out according to the standard methods of Evans and Sofowora. The preliminary antibacterial activity of the plants was assayed using agar well diffusion. The minimum inhibitory concentration (MIC) assays were determined at concentrations of 200, 100, 50 and 25 mg/ml for all extracts against the test bacteria, Methicillin Resistant *Staphylococcus aureus* (MRSA), Methicillin Sensitive *Staphylococcus aureus* (MSSA). The presence of alkaloids, flavonoids, saponins, tannins phlobatannins, steroids, glycosides and quinones, was detected using qualitative and quantitative test methods. Alkaloid was detected only in *Cassythia filiformis* which was more extractable in ethanol than ethyl acetate and N-hexane extracts. The test pathogenic bacterial isolates at concentration of 1g of plant extracts in 1ml of dimethyl sulphoxide (DMSO), Showed appreciable inhibitory activities after 24 hours of incubation at 37^o C. The susceptibility test revealed that Vancomycin (VA₃₀) showed inhibition Zone 6.3±1.5 and 5.7±2.1 in MRSA. The ethanolic, ethyl acetate, N-hexane and warm water extract of *Piper guineense*, *Ocimum gratissimum* and *Cassythia filiformis* showed the high rate of inhibition zones of 7.5±1.5 on MRSA, 7.1±0.5 on MSSA,. The three plant extracts shows effective potency on the selected pathogenic bacterial isolates than highly rated antibiotic (reference drugs) in disease cure and prevention. This study was undertaken to screen bioactive constituents of *Piper guineense*, *Ocimum gratissimum*, *Cassythia filiformis* and to evaluate their antibacterial active against test organisms.

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I. Introduction

Plant materials remain an important resource to combat serious diseases in the world (Edeogal, 2005). The traditional medicinal methods, especially the use of medicinal plants, still play a vital role to cover the basic health needs in the developing countries. The medicinal value of these plants lies in some chemical active substances that produce a definite physiological action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannin, flavonoid, saponins, phlobatannins, steroid, glycosides and quinones compounds (Edeogal, 2005).

Within the recent years, infections have increased to a great extent and antibiotics resistance effects become an ever-increasing therapeutic problem (Mahesh, 2008). Natural products of higher plants may possess a new source of antimicrobial agents with possibly novel mechanisms of action (Ahmad, 2007). They are effective in the treatment of infectious diseases while simultaneously mitigating many of the side effects that are often associated with synthetic antimicrobials (Iwu, 1999). Therefore, it is of great interest to carry out a screening of these plants in order to validate their use in folk medicine and to reveal the active principle by isolation and characterization of their constituents. Systematic screening of them may result in the discovery of novel active compounds.

Traditional medicine in the African region of the World Health Organization (WHO) has become a part of the culture though this form of medicine is not as well organized as in other parts of the world, for example China and India. Many countries in Africa now have a division, department or task force on traditional medicine, usually attached to their ministries of health, and at least one research group investigating medicinal plants (Barbour, 2004).

II. MATERIAL AND METHODS

Collection and Identification of Plant Sample

The plants, *Piper guineense*, *Ocimum gratissimum* and *Cassytha filiformis* was collected on October 2012 from Ibesikpo Asutan, Local Government Area in Akwa Ibom State. The plant was identified by Dr. (Mrs.) M.E. Bassey, a plant taxonomist in the Department of Botany and Ecological Studies, University of Uyo. The herbarium number was giving to each plant. *Ocimum gratissimum* N0: 3078 Uyo, *Piper guineense* N0: 3079 Uyo and *Cassytha filiformis* linn N0: 3080 Uyo.

Preparation of Plant Extracts

The fresh leaves of *Piper guineense*, *Ocimum gratissimum* and *Cassytha filiformis* were dried for 23 days at the normal room temperature and milled with the help of grinding machine or miller. Then 300g of each of milled plants were weight, while 400ml of solvents were measured into each conical flask according to their labeling, for soaking. After 24 hours of soaking all milled plants with N-hexane, Ethanol, Ethyl acetate and warm water. 100ml for each of the plant extract were filtered into breakers for air drying. After 16 hours to 24 hours all breaker containing N-hexane were dry to complete dryness, and after 28 hours to 36 hours all the breaker containing Ethyl acetate were dry to complete dryness, and after 48 hours to 72 hours all the breaker containing ethanol were dry to complete dryness, meanwhile after 336 hours and above all the breaker containing warm water were dry to complete dryness. The dry extract was placed in a beaker and covered with aluminum foil to prevent contamination and stored in a refrigerator at 4⁰C until used for the experiment reported in this research. The extraction method adopted was maceration otherwise called cold extraction. This method was preferentially chosen to prevent the thermolabile constituents of plants.

Phytochemical Screening

The phytochemical screening of the sample was carried out according to the Standard methods of Treasure and Evans (2002), Sofowora (1999) and Harborne (1998).

Test for Alkaloids

Mayer's Test: About 0.5g of the extract was dissolved in 10ml of dilute sulphuric acid, boiled and filtered hot. A freshly Mayer's reagent was then added by a few drops to the filtrate formation of milky colour indicates a positive test (Harborne, 1998 and Evans, 2002).

Test for Saponins

Frothing test: The plant extract (0.5g) was dissolved in distilled water and shaken vigorously. The presence of frothing indicates of positive test (Trease and Evans, 2002).

Test for Tannins

Bromine water test: About 5 drops of the plant extract was added to 100ml of water and stirred with bromine water. Decolourisation of bromine water indicates a positive test (Trease and Evans, 2002).

Test for Anthraquinones

Borntrager's test was used for the detection of anthraquinones. About 0.5g of each plant extract was shaken with 10ml benzene, filtered and 5ml of 10percent ammonia solution added to the filtrate. The mixture was shaken and the presence of a pink, red, or violet colour in the ammoniacal (lower phase) indicates a positive test (Evans, 2002).

Test for phlobatannins

Formaldehyde Test: - Three drops of 10% formaldehyde and six drops of dilute hydrochloric acid was added to 5ml of the test solution. The precipitate which was formed was washed with hot water first, then with warm alcohol (ethanol), and finally 5% potassium hydroxide solution. A bulky precipitate which leaves a coloured residue after this washing treatment indicates a positive test of phlobatannins (Evans, 2002).

Test for Flavonoids

Shinoda's Test: - Few pieces of magnesium metal strip were added to 5mls of the plant extract and then 2mls of concentrated hydrochloric acid was added. The formation of orange, red, crimson or magenta colouration indicates a positive test (Evans, 2002).

Test for Cardiac Glycosides

Keller Killiani Test (Deoxy-sugar):- The plant extract (0.5g) was dissolved in 2mls glacial acetic acid containing one drop of ferric chloride solution. This was then underlayered with 1ml of concentrated sulphuric acid. A brown ring obtained at the interface indicated the presence of a desoxy sugar characteristic of cardenolides. A violet ring may appear below the brown ring while, in the acetic acid layer a greenish ring may form just above the brown ring and gradually spread throughout this layer (Sofowora, 1999).

Test for Steroids

Salkowski test (Steroidal ring):- The plant extract (0.5g) was dissolved in 2mls of chloroform and then 1ml of concentrated sulphuric acid was carefully added to form a lower layer. A reddish brown colour at the interphase indicates a positive test (Presence of a steroidal ring) (Hostettmann, 1991).

Source of Test Micro-organisms

The test micro-organisms used for the antibacterial activity screening namely: Methicillin-resistant *Staphylococcus aureus* and Methicillin-sensitive *Staphylococcus aureus*, were obtained from the Department of Pharmaceutical Microbiology and Biotechnology, Faculty of Pharmaceutical Science, Nnamdi Azikiwe University Agulu Campus, Anambra State, Nigeria. The test micro-organisms were collected and stored in the bijoux-bottle for use.

Preliminary Antibacterial Screening

The antibacterial activities of the crude plant extracts were evaluated by agar well (Halo) diffusion (Nair and Chando, 2005). Nutrient agars were used for the sub-culturing the bacterial isolates from bijoux bottle. Mueller-Hinton Agar (Hi-media) supplemented with 4g of NaCl was used for the sensitivity screening. Plant extracts (1g) in different solvent (Ethanol, Ethyl acetate, N-hexane, warm water) were prepared in 1ml of aqueous dimethyl sulphoxide (DMSO). The MHA plates were prepared by pouring 15 ml of molten media into sterile petri-plates. The plates were allowed to solidify for 5 minutes, and each of the organism then streaks on the medium. With a sterile cork-borer halo of equal depth ($\Delta=7$ mm diameter) were dug with a previously sterilized cork-borer. The wells were aseptically filled up with the extracts avoiding splash and overfilling. The plates were incubated at 37⁰C for 24 hours. The sensitivity of the test organisms to each of the extract were indicated by clear halo around the wells. The halo diameters were taken as an index of the degree. Aqueous DMSO was used as negative control, while Vancomycin (VA₃₀), Cefoxitin (FOX₃₀), Oxacillin (OX₁) and Cloxacillin (OB₅) were used as the positive control. At the end of incubation, inhibitory zones formed around were measured with transparent ruler in millimeter and compared with control disc. All experiments were carried out in triplicates.

Minimum Inhibitory Concentration (MIC)

The MIC method was applied on all the extracts against microorganisms by the Agar Diffusion Assay. The different concentrations of extracts (200, 100, 50, and 25) were prepared using 500mg/ml as a stock (2g of extract in 4ml of DMSO). The Mueller-Hinton Agar (MHA) plates were prepared by pouring 15ml of molten media into sterile Petri plates. The plates were allowed to solidify for 5 minutes, and each test organisms were then streak on the medium. They plates were halo with 7mm diameter cork-borer. Then each plate contains four halos, one for each concentration. The plates were incubated aerobically at 37⁰C and zones of inhibition were measured and recorded. Each test was assayed in triplicate.

Statistical Analysis

The inhibitory zones of all crude extract were expressed as the mean \pm standard Deviation.

III. Result

Table 1. Quantitative Phytochemical screening of Plant Extract of *Piper guineense*, *Ocimum gratissimum* and *Cassytha filiformis*

Test	Piper guineense(mg/100g)				Ocimum gratissimum(mg/100g)				Cassytha fillformis (mg/100g)			
	E.E	E.A.E	N.H.E	W.W.E	E.E	E.A.E	N.H.E	W.W.E	E.E	E.A.E	N.H.E	W.W.E
Alkaloids	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	2.63	0.0	0.61	1.7
Flavonoids	0.36	4.5	0.0	2.33	6.1	1.2	0.67	5.77	3.18	3.44	0.38	0.29
Saponins	2.22	0.41	0.29	4.97	3.15	0.37	0.41	2.56	4.0	0.35	0.44	6.79
Tannins	4.5	3.99	0.43	7.01	2.4	1.99	2.55	4.88	1.77	4.1	0.78	5.66
Steroids	6.88	2.77	1.09	0.99	3.4	2.9	0.91	5.0	0.42	5.7	0.24	0.39

Key:- E.E = Ethanol extract, E.A.E = Ethyl acetate extract, N-H.E = N- Hexane extract, W.W.E = Warm water extract.

Table 1 represent Quantitative Phytochemical screening of ethanolic extract, Ethyl acetate extract, N- Hexane extract and warm water extract of *Piper guineense*, *Ocimum gratissimum* and *Cassytha filiformis*.

From *Piper guineense*, tannins (7.01mg/100g) of warm water extract had the highest yield followed by steroids (6.88mg/100g) of ethanolic extract of *Piper guineense*.

From *Ocimum gratissimum*, flavonoids (6.1mg/1000g) of exthanolic extract *Ocimum gratissimum* had highest yield followed by flavonoids (5.77mg/100g) of warm water extract of *Ocimum gratissimum*.

From *Cassytha Fillformis*, Saponins (6.79mg/100g) and tannins(5.66mg/100mg) of warm water extract of *Cassytha Fillformis* had highest yield and Steroids (5.7mg/100g) of Ethyl acetate extract *Cassytha Fillformis*.

Table 2: Qualitative Phytochemical Screening of Plant Extracts of *Piper guineense*, *Ocimum gratissimum* and *Cassytha filiformis*.

Secondary (Test) Metabolites	<i>Piper guineense</i>				<i>Ocimum gratissimum</i>				<i>Cassytha filiformis</i>			
	E.E.	E.A.E.	N-H.E.	W.W.E.	E.E.	E.A.E.	N-H.E.	W.W.E.	E.E.	E.A.E.	N-H.E.	W.W.E.
Alkaloids	-	-	-	-	-	-	-	-	+	-	-	+
Flavonoids	+	++	-	+	+++	+	+	++	++	++	+	+
Saponins	+	-	-	++	+	-	-	+	+	-	-	++
Tannins	++	+	-	++	+	+	+	++	+	+	-	++
Phlobatannins	+	-	-	++	+	-	-	++	+	-	-	++
Steroids	++	++	+	+	+	++	-	+	-	++	-	-
Glycosides	+	+	++	+	+	+	-	+	+	+	+	+
Quinones	+	+	+	++	+	+	-	+	+	-	-	+

Key:-E.E = Ethanol extract, E.A.E = Ethyl acetate extract, N-H.E = N- Hexane extract, W.W.E = Warm water extract

(-) = Not detected, (+) = Low concentration, (++) = Moderate concentration, (+++) = High concentration.

Table 2 represent the qualitative phytochemical screening of ethanolic extract, ethyl acetate extract, N-Hexane extract and warm water extract of piper guineense, *Ocimum gratissimum* and *Cassytha filiformis*.

Saponins, tannins, Phlobatannins and Quinones of warm water extract of piper guineense have the moderate concentration when compare to saponins, tannins, phlobatannins and Quinones of ethanolic extract, Ethyl acetate extract, N- Hexane extract of piper guineense except for tannins of piper guineense which also had moderate concentration, followed by steroids from ethanolic extract, ethyl acetate extract plus Glycosides from N- Hexane extract of piper guineense.

Apart from flavonoids, tannins and Phlobatannins of warm water extract of *Ocimum gratissimum* that had moderate concentration and steroids of N- Hexane extract, is the flavonoids from ethanolic extract of *Ocimum gratissimum* that had the highest concentration.

Saponins, tannins, Phlobatannins and Quinones of warm water extract of *Cassythia filiformis* had the moderate concentration when compare to saponins, tannins and phlobatannins of ethanolic extract, Ethyl acetate extract, N- Hexane extract of the same *Cassythia filiformis* except for flavonoids of ethanolic and ethyl acetate extract of *Cassythia filiformis* which had moderate concentration also.

Table 3: Preliminary stage of the mean inhibition zone (MM)

Bacterial Organisms	<i>Piper guineense</i>				<i>Ocimum gratissium</i>				<i>Cassythia filiformis</i>				Susceptibility Disc
	E.E	E.A.E	N-H.E	W.W.E	E.E	E.A.E	N-H.E	W.W.E	E.E	E.A.E	N-H.E	W.W.E	VA ₃₀
MRSA	7.5±1.5	6.3±3.0	6.7±1.3	5.7±0.1	0.0±0.0	5.3±2.1	7.0±3.0	0.0±0.0	5.7±3.1	6.9±2.0	5.7±2.1	4.7±1.5	6.3±1.5
MSSA	6.5±1.0	0.0±0.0	4.0±0.5	0.0±0.0	5.0±2.0	7.1±0.5	6.0±1.0	7.0±0.5	5.0±3.5	6.0±2.1	5.5±0.5	0.0±0.0	0.0±0.0

Key: E.E = Ethanol extract, E.A.E = Ethyl acetate extract, N-H.E= N-hexane extract, W.W.E = Warm water extract, VA₃₀= Vancomycin, MM=Millimeter, MRSA =Methicillin Resistant *Staphylococcus aureus*, MSSA= Methicillin Sensitive *Staphylococcus aureus*,

Note: The results are the values of triplicate tests measured after 24hr incubation at 37⁰C.

Table 3 represent Preliminary stage of the mean inhibition zone of Methicillin Resistant *Staphylococcus aureus* (MRSA) and Methicillin Sensitive *Staphylococcus aureus* (MSSA).

Ethanolic extract of *Piper guineense* (7.5±15) had the highest mean inhibiton on Methicillin Resistant *Staphylococcus aureus* (MRSA) followed by others. Ethyl acetate extract of *Ocimum gratissium* (7.1±0.5) had the highest mean inhibition on Methicillin Sensitive *Staphylococcus aureus* (MSSA) followed by others.

Table 4: Minimum inhibitory concentration of plants extracts of *Piper guineense*, *Ocimum gratissimum* and *Ocimum gratissimum*on different concentration of mg/ml of Methicillin Resistant *Staphylococcus aureus*(MRSA).

Conc. Mg/ml.	<i>Piperguineese</i>				<i>Ocimum gratissimum</i>				<i>Cassythia filiformis</i>			
	E.E	E.A.E	N-H.E	W.W.E	E.E	E.A.E	N-H.E	W.W.E	E.E	E.A.E	N-H.E	W.W.E
200	5.5±1.6	3.7±1.0	6.5±0.3	5.0±1.5	6.7±0.5	6.3±0.1	4.5±1.5	2.7±0.3	6.7±1.5	5.0±1.0	6.5±1.5	7.0±0.5
100	4.0±1.5	6.5±0.5	7.0±1.0	6.8±1.5	1.9±0.5	1.3±1.0	5.5±1.5	1.0±0.1	5.5±1.5	6.0±1.0	7.0±1.6	6.7±1.0
50	5.6±1.6	1.6±1.0	3.9±0.5	4.0±1.5	5.4±0.3	7.0±0.2	1.0±1.5	3.0±0.6	6.0±0.5	4.0±05	7.5±1.5	4.5±0.5
25	6.1±0.3	6.0±1.5	3.5±1.5	4.0±1.0	7.0±0.5	4.6±0.5	1.0±0.5	4.0±1.5	5.0±1.0	6.7±1.5	4.0±1.0	5.3±0.4

Key: E.E =Ethanol extract, E.A.E =Ethyl acetate extract, N-H.E =N-hexane extract, W.W.E =Warm water extract, MRSA =Methicillin Resistant *Staphylococcus aureus*,

Note: The results are the values of triplicate tests measured after 24hr incubation at 37⁰C

Table 4 represent minimum inhibitory concentration of plants extracts of *Piper guineese*, *Ocimum gratissimum* and *Ocimum gratissimum*on different concentration of mg/ml of Methicillin Resistant *Staphylococcus aureus* (MRSA).

N-hexane extract (7.0±1.0), warm water extract(6.8±1.5), ethyl acetate extract (6.5±0.5) of *Piper guineese* had the highest minimum inhibitory concentration respectively at 100mg/ml concentration of Methicillin Resistant *Staphylococcus aureus* and also that of ethanolic extract of *Piper guineese* at 25mg/ml concentration of MRSA. Ethanolic extract of *ocimum gratissium* (7.0±0.5) and ethyl acetate extract of *Ocimum gratissium* had the highest minimum inhibitory concentration at 50mg/ml and 25mg/ml concentration of MRSA.

N-Hexane extract of *cassythia filliformis* (7.0±1.5) had the highest minimum inhibitory concentration at 50mg/ml concentration of MRSA compared to other extract of *cassythia filiformis*.

Table 5: Minimum inhibitory concentration of plants extracts of *Piper guineese*, *Ocimum gratissimum* and *Ocimum gratissimum* at different concentration of mg/ml of Methicillin Sensitive *Staphylococcus aureus* (MSSA).

Conc. Mg/ml	<i>Piper guineese</i>				<i>Ocimum gratissimum</i>				<i>Cassythia filiformis</i>			
	E.E	E.A.E	N-H.E	W.W.E	E.E	E.A.E	N-H.E	W.W.E	E.E	E.A.E	N-H.E	W.W.E
200	6.0±1.0	4.0±0.7	2.0±0.6	7.0±0.4	5.0±1.0	6.5±1.0	4.5±0.5	6.0±1.5	2.5±1.5	4.5±0.5	6.5±0.6	3.8±0.5
100	7.0±1.5	5.0±1.5	6.0±1.0	2.0±0.7	6.5±0.5	3.0±1.5	5.8±1.5	7.0±1.5	6.0±1.0	4.0±0.5	6.0±0.7	5.0±1.6
50	4.0±1.7	3.0±1.5	5.5±1.5	7.0±0.3	4.5±1.6	6.5±0.5	1.0±0.5	3.0±0.7	6.5±1.5	4.0±1.3	5.0±0.7	2.3±0.5
25	5.1±1.7	6.0±1.5	5.7±1.5	3.0±1.0	6.0±1.5	4.7±1.5	3.0±0.3	2.7±1.4	6.0±0.5	2.0±0.7	3.0±1.5	1.0±0.5

Key: E.E = Ethanol extract, E.A.E = Ethyl acetate extract, N-H.E= N-hexane extract, W.W.E = Warm water extract, MSSA= Methicillin Sensitive *Staphylococcus aureus*,

Note: The results are the values of triplicate tests measured after 24 hours incubation at 37⁰C.

Table 5 represent minimum inhibitory concentration of plants extracts of *Piper guineense*, *Ocimum gratissimum* and *Ocimum gratissimum* at different concentration of mg/ml of Methicillin Sensitive *Staphylococcus aureus* (MSSA).

Ethanol extract of piper guineense had highest minimum inhibitory concentration (7.0±1.5) at 100mg/ml concentration of Methicillin Sensitive *Staphylococcus aureus* while that of warm water extract of Piper guineense had minimum inhibitory concentration (7.0±0.4) and (7.0±0.3) at 200mg/ml and 50mg/ml concentration of MSSA respectively.

Warm water extract of *Ocimum gratissimum* had the highest minimum inhibitory concentration (7.0±1.5) at 100mg/ml concentration of MSSA followed by others.

Ethanol extract of *Cassipoupa filiformis* had the highest minimum inhibitory concentration of (6.5±1.5, 6.0±1.0, 6.0±0.5) at 50, 100 and 25mg/ml concentration of MSSA respectively and N-Hexane extract of *Cassipoupa filiformis* also had the highest minimum inhibitory concentration of (6.5±0.6 and 6.0±0.7) at 200 and 100mg/ml concentration of MSSA respectively.

IV. Discussion

Importance of this study is the interest in plants with antibacterial properties has revived as a result of current problems associated with the use of antibiotic. In this study, three medicinal plants are popular used in Nigeria for the treatment of several ailments of bacterial and non-bacterial infection. Infections have increased to great extent and antibiotics resistance effects become an ever-increasing therapeutic problem. This study showed that the test plants; *Piper guineense*, *Ocimum gratissimum* and *Cassipoupa filiformis* are good sources of antimicrobial property. The bioactive compounds on the medicinal plants employed contain various secondary metabolites such as Alkaloids, flavonoids, saponins, tannins, phlobatannins, steroids, glycosides and quinines in appreciable quantities. The effective inhibitory potency observed with the plants parts proof that the inhibitory compounds were extractable by the employed solvents against the tested pathogenic bacterial isolates. This observation as reported correlates with De and James (2002) who emphasized that these compounds are known to show medicinal activity as well as exhibiting physiological activity. However, 1g of plants extracts in difference solvent (Ethanol, Ethyl acetate, N-hexane, and warm water) were prepared in 1ml of aqueous dimethyl sulphoxide (DMSO). This concentration was visible active on the combinative therapeutic actions of the various secondary metabolites contained in the plants. Some of the tested bacteria isolates such as MRSA and MSSA reported to be associated with nosocomial and community acquired infection were found susceptible to the plants crude extracts used in this study. This proof emphasized that some nosocomial and community acquired infections could be prevented or alleviated with the use of N-hexane, ethanol and ethyl acetate extract of the investigated plants.. Healthy fresh leaves of these plants could as well be prepared as food. The MIC values of the plants were determined by using different concentrations (200, 100, 50 and 25mg/ml) of ethanol, ethyl acetate, n-hexane and warm water extracts on the tested bacterial isolates. This perhaps helps to interpret the fact that differences in inhibitory diameters (mm) could result in the same therapeutic potency when concentration varies, depending on the organism's susceptibility to the antibacterial components present in the extracts. The presence and the phytochemical components of the studied plants, the inhibitory zones and the MIC concentrations at which values were effective on the tested organisms, highlights that there were variations in the antimicrobial potency of the plants extracts. The variations in the sensitivity could also be attributed to the differences in growth rate of the tested organisms, nutritional requirements, temperature and inoculum size (Gaill and Jon, 1995). It has been reported that antibiotics are not the only antibacterial agents and this study observed the effective potency of the studied plants extracts on the selected pathogenic bacteria isolates than some highly rated antibiotics (reference drug) in disease cure and prevention. One problem in the use of medicinal plants is the quantity desired to effect cure hence most times, medication is basically on unspecified quality of decoctions and infusion (Manesh, 2008).

V. Conclusion

In conclusion, the results provide justification for the use of these plants in folk medicine to treat various infectious diseases. But the problem in the use of medicinal plants is the quantity desired to effect cure hence most times, medication is basically on unspecified quality of decoctions and infusions. The plants parts showed antibacterial effectiveness on the tested bacterial isolates resulted within 24 hours of incubation in both the crude extract screening and Minimum inhibitory concentration values. The ethyl acetate and ethanol extracts of the plants displayed extensively a competitive inhibitory potency with the more effective N-hexane extracts of the plants parts on the tested isolates which majority are gram negative bacterial known for their ability to

form resistance to drug. The plants though effective on all the bacterial isolates, there were variation in inhibitory potency resulting from variations in the secondary metabolites concentration in the plants parts. Meanwhile warm water and ethanol extract showed the highest level reaction in the secondary metabolites screening test.

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