

BIOACTIVITY AND PHYTOCHEMICAL SCREENING OF NIGERIAN MEDICINAL PLANTS GROWING IN ONDO AND EKITI STATE AGAINST BACTERIAL ISOLATES FROM PEDIATRICS HOSPITAL.

ABSTRACT

The Aqueous, ethanol, hexane and ethyl acetate extracts of the leaves, bark and roots of *Acacia albida*, *Anchomanes difformis*, *Bosciasenegalensis*, *Bridelia ferruginea*, *Ficus ingens*, *Indigofera arrecta* and *Moringa oleifera* were examined for possible source of antibacterial activities against some selected clinical organisms found in the pediatric hospital in Ondo and Ekiti State, Nigeria. The selected clinical organisms used for this research work are *Salmonella typhi*, *Staphylococcus aureus*, *Klebsiella pneumoniae* and *Escherichia coli*. It was observed that all plant were potent against all the tested clinical organism. *Anchomanes difformis*, *Moringa oleifera* and *Indigofera arrecta* extracts were the most active medicinal plant during the course of this research work. *Bosciasenegalensis*, *Acacia albida*, *Bridelia ferruginea* and *Ficus ingens* extract were not as active as the first three medicinal plants. All the plants extracts shows evidence of antibacterial properties. Phytochemical screening of the four medicinal plants reveals, very vital phytochemicals like saponin, tanins, alkaloids, flavonoids, cardiac glucosides, anthraquinones, at a very appreciable quantity. It also reveals the present of minerals at mg/100g, the minerals present are sodium, potassium, calcium, iron, zinc, copper and phosphorus which are essential in biological and physiological process in human health. The main objectives of this research work is to elucidate the antimicrobial and phytochemical biological activity of seven Nigerian Medicinal plants on selected clinical organisms in Nigeria, as another remedy for the treatment of various infections and a new race in developing locally made drugs.

27 **Keywords:** Antibacterial assay, agar well diffusion method, phytochemical
28 screening; minerals composition; nutrient and anti-nutrient composition, proximate
29 composition.

30 INTRODUCTION

31 The discovery of new antibiotics is a very important aspect of medicinal plants, which is dated
32 back 500 to 700 BC. Pharmacist, Pharmacologist, Biochemist and Pharmaceutical microbiologist
33 are taking the necessary clue from the advent of medicinal plants in the discovery of production
34 of new antibiotic for the cure of different diseases(Iwu 1999).

35 Medicinal Plants are good source of antimicrobial agent which can be used as a form of
36 chemotherapy in the cure of different illness. Some are used for chewing sticks, some are used
37 for soup condiment and some plants are use even for spiritual purpose in some part of the
38 western part of Nigeria (Osuntokun 2014).Major health hazard may not be cure by the
39 conventional medicine, but medicinal plants are found to be useful in such areas,such as the
40 pediatric hospital which is under study in the scope of this research work. Some poor people may
41 not even have enough money to buy the conventional drug, but nature has blessed us with the
42 medicinal plants for food and also for the cure of major health hazard.(Osuntokun 2014,
43 Sofowora (2008).

44 The use of medicinal plants for therapy predates the introduction of synthetic antibiotics. It
45 also predates the religious, social and economic barriers (Akinyemiet al.,2000). Antibiotics are
46 naturally occurring or synthetic organic compounds which inhibit or destroy bacteria,
47 generally at low concentration (Brooks et al.,2007). Many lives have been saved by the use
48 of synthetic antibiotics but in recent times, these disease causing microbes have developed
49 resistance to some of these antibiotics and have become difficult to treat in our various
50 health care homes ,taking an example of our pediatric hospital. This situation may lead to
51 increased cost of medication and in the long run leads to increased morbidity and mortality
52 (Osuntokun 2015).

53 The medicinal plants is taking a place of pride in the world economy knowing fully well
54 that,develoing country can make a lot of foreign exchange in this aspect of drug production,

55 drug discovery, development of new drugs and antibiotic through medicinal plants, especially
56 in Nigeria, where God has blessed us abundantly with various degree of medicinal
57 plants. (Nmorsi & Osuntokun 2014), (Okwu 2005).

58 Traditional medicine like orthodox medicine has its own methods and techniques of applications,
59 which however aims at healing diseases (Wurochekke, *et al.*, 2008). The treatment and
60 control of diseases by the use of the available medicinal plants in a locality will continue to
61 play a significant roles in medical healthcare implementation in the developing countries of
62 the world. Nearly all cultures and civilizations from ancient times to the present day have
63 depended fully or partially on herbal medicine because of their effectiveness, affordability,
64 availability, low toxicity and acceptability. Our empirical belief on herbal drugs, gives us an
65 edge on the use of medicinal plants which is on the high increase in our daily lives especially in
66 Nigeria.

67 Due to ineffectiveness of most drugs as a result of microbial resistance to available agents most
68 especially in developing countries, more patients in Nigerian are seen in medical centers than
69 ever. The intractable problem of antimicrobial resistance has led to the resurgence of interest in
70 herbal products as sources of novel compounds to suppress or possibly eradicate the ever
71 increasing problems of emergence of newer diseases. Udekwe, (2010).

72 Phytochemicals are plants produce chemical compounds as part of their normal metabolic
73 activities. These phytochemicals are divided into (1) **primary metabolites** such as sugars and
74 fats, which are found in all plants; and (2) **secondary metabolites**—compounds which are found
75 **in a smaller range of plants**, serving a more specific function. It is these secondary metabolites
76 and pigments that can have therapeutic actions in humans and which can be refined to produce
77 drugs—examples **are inulin** from the roots of **dahlia**, **quinine** from the cinchona, morphine and
78 codeine from the poppy, and digoxin from the foxglove. Carrubba, & Scalenghe (2012).

79 Alkaloids are a class of chemical compounds containing a nitrogen ring. Alkaloids are produced
80 by a large variety of organisms, including bacteria, fungi, plants, and animals, and are part of the
81 group of natural products (also called secondary metabolites). Many alkaloids can be purified
82 from crude extracts by acid-base extraction. Examples are the local anesthetic and stimulant
83 cocaine; the psychedelic psilocin; the stimulant caffeine; nicotine; the analgesic morphine; the

antibacterial berberine; the anticancer compound vincristine; the anti-hypertension agent reserpine; the choline-mimetic galatamine; the spasmolysis agent atropine; the vasodilator vincamine; the anti-arrhythmia compound quinidine; the anti-asthma therapeutic ephedrine; and the antimalarial drug quinine. **Springbob *et al.*, 2009.**

Polyphenols (also known as phenolics) are compounds that contain phenol rings. The anthocyanins that give grapes their purple color, the **isoflavones**, the **phytoestrogens** from soy and the **tannins** that give tea its astringency are **phenolics**. **Cravotto *et al.*, 2010.**

Glycosides are molecules in which a sugar is bound to a non-carbohydrate moiety, usually a small organic molecule. Glycosides play numerous important roles in living organisms. Many plants store chemicals in the form of inactive glycosides. These can be activated by enzyme hydrolysis, which causes the sugar part to be broken off, making the chemical available for use. Many such plant glycosides are used as medications. In animals and humans, poisons are often bound to sugar molecules as part of their elimination from the body. **Cravotto *et al.*, 2010.**

Terpenes are a large and diverse class of organic compounds produced by a variety of plants, particularly conifers, which are often strong smelling and thus may have had a protective function. They are the major components of resins, and of turpentine produced from resin. Terpenes are major biosynthetic building blocks within nearly every living creature. Steroids, for example, are derivatives of the triterpene squalene. Terpenes and terpenoids are the primary constituents of the essential oils of many types of plants and flowers. Essential oils are used widely as natural flavor additives for food, as fragrances in perfumery, and in traditional and alternative medicines such as aromatherapy. Vitamin A is an example of a terpene. The carotenoids produce the reds, yellows and oranges of pumpkin, corn and tomatoes. **Edgar *et al.*, 2002.**

In view of this, it is therefore very important to search for effective, efficient, low cost and reliable traditional therapeutic agents, for the development of locally made **drugs (antibiotics)**, to reduce abuse of drugs and drug resistant organisms which is a common factor in some conventional drugs. **(Trease 1989).** Plant profile, local and botanical names and its uses, are

111 elucidate in the table 1 below, to show that this plant are been in use for centuries.
 112 **Sofowora(2008).**

113 **Table 1;Profile of the Medicinal Plants under study**

Botanical Name	Family	Local Names	Part used	Medicinal uses
1. <i>Acacia albid.a</i>	Leguminosae	Gawo	Root	Skin infections
2. <i>Anchomanesdifformis.</i>	Araceae	Chakara	Roots	1.Cough. 2.Respiratory diseases.
3. <i>Bosciasenegalensis.</i>	Capparidaceae	Anza	Roots	1.Anticancer. 2.Ulcer swellings.
4. <i>Brideliaferruginea.</i>	Euphorbiaceae	Kirni	Leaf	1Antibiotic. 2.Skin infection, 3.Diuretic, 4.Febrifuge urethral discharge.
5. <i>Ficusingens.</i>	Moraceae	Kawuri	Root	1.Piles. 2.diarrhea. 3.laxative. 4.diuretic.
6. <i>Indigoferaarrecta.</i>	Papilionoideae	Ba-ba	Leaf	1.Infections stomach. 2.Pain. 3.Snakebite. 4.Epilepsy.
7. <i>Moringaoleifera.</i>	Moringacceae	Zogale	Leaf	1Asthma. 2.Eye infection. 3.Migraine. 4.Headache.

				5.Febrifuge. 6.Abortifacient.
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114 Sofowora(2008),Osuntokun (2014).

115 MATERIALS AND METHOD

116 The experiment involved seven types of Nigerian medicinal plants, with four different
117 extracting solvent, four clinical organisms, and three concentrations of medicinal plants
118 extracts, the four clinical isolates are Bacteria.

119 Source of plant materials

120 The Nigerian Medicinal plants selected were obtained from the roots and the branch of
121 *Acacia albida*, *Anchomansdifformis*. *Bos senegalenss*, *Brideliaferruginea*,
122 *FicusingensIndigoferaarrecta* and *Moringaoleifera*. The plant were collected in
123 IkareAkoko, a tropical rainforest, Ondo State, Nigeria with latitude (7.21692 North) and
124 longititude (5.21561 East) The plant parts were authenticated by a Botanist at the
125 Department of Botany, Ekiti State University Ado Ekiti, and AdekunleAjasin University,
126 AkungbaAkoko, Ondo State ,Nigeria.

127 Source of microorganisms

128 Pure cultures of *Salmonella typhi*, *Staph aureus*, *KlebsiellapnumoniaeEscherichia*
129 *coli* isolated from children stools attending the Pediatrics from the Federal Medical
130 Center (Pediatrics) (FMC), Owo, Ondo State, and Ekiti State Teaching Hospital
131 (Pediatrics),Ekiti State, Nigeria Bacterial cultures were maintained on Blood agar slant
132 (Osuntokun 2015).

133 Extract preparation

134 The leaf and root of the tested plant were well dried and then grinded to powder. About
135 200g of the powder were separately soaked in 400ml of 95% Ethanol, Distilled water,
136 Ethyl acetate and Hexane in a 500ml reagent bottle and stoppered. This was allowed to
137 stand for 14 days to permit full extraction of the active ingredients. The fluids were then
138 filtered using Whatman No1 filter paper. The extracts were rotary dried to obtain the
139 concentrate. It was then kept in refrigerator at -4⁰c prior to use. (Appelbaum 1992),
140 (Osuntokun 2015).

141 **Antimicrobial Assay (Determination of Zones of Inhibition) using Agar Well**
142 **Diffusion method.**

143 Twenty five milliliters of Kliger iron agar were poured into sterile petri dishes and
144 allowed to set. Standardized test bacterial cultures were inoculated into the sterile Kliger
145 iron agar plate using sterile cotton swab. A sterile cork borer of 6mm diameter were
146 used to punch wells on the agar on each of the petri dishes. Three holes were made
147 on the surface of the plate with one in the center to serve as the control. Each hole was
148 labeled (A,B,C, D) representing a particular concentration of 20mg/ml, 40mg/ml and
149 60mg/ml. The dishes were then filled into the wells with three drop of their
150 respective type of extract according to the labeled format. The central well containing
151 the diluent reagent **Streptomycin** was used as control. The process was carried out for
152 each extract and the inoculated petri dishes were left for few minutes for extract
153 of diffuse into agar. The plates were incubated at 37°C for 18-24 hours after which the
154 zones of inhibition (if any) were measured (**Osuntokun&Olajubu 2014**).

155 **PHYTOCHEMICAL SCREENING**

156 The extract were analysed for the presence of Alkaloid, Glycosides, Tannins, Saponins,
157 Anthraquinones, Anthocyanosides, Flavonoids, Reducing sugars, Cyanogenic(**Edeogaet**
158 **al.,2005, Osuntokun&Ajayi 2014, Harbone 1998,Sofowora 2008**).

159 **Qualitative Method of Analyses**

160 Plant filtrates were prepared by boiling 20 g of the fresh plant in distilled water. The solution was
161 filtered through a vacuum pump. The filtrate were used for the phytochemical screening for
162 flavonoids, tannins, saponins, alkaloids, reducing sugars, anthraquinones and anthocyanosides.

163 **(i).Test for Alkaloids**

164 About 0.2gram was warmed with 2% of H₂SO₄ for two minutes, it was filtered and few drops of
165 Dragendoff's reagent were added. Orange red precipitates , indicate the present of Alkaloids
166 (**Trease and Evans 1989**).

167 **(ii) Test for Tannins**

168 One milliliter of the filtrate was mixed with 2ml of **FeCl₃**. A dark green colour indicated a
169 positive test for the tannins.

170 **(iii) Test for Saponins**

171 One milliliter of the plant filtrate were diluted with 2 ml of distilled water; the mixture were
172 vigorously shaken and left to stand for **10min** during which time, the development of foam on
173 the surface of the mixture lasting for more than 10mm, indicates the presence of saponins.

174 **(iv) Test for Anthraquinones**

175 One milliliter of the plant filtrate was shaken with **10ml** of benzene; the mixture was filtered and
176 5 ml of 10 % (v/v) ammonia were added, then shaken and observed. A pinkish solution indicates
177 a positive test

178 **(v).Test for Anthocyanosides**

179 One milliliter of the plant filtrate were mixed with 5 ml of dilute HCl; a pale pink colour
180 indicates the positive test.

181 **(vi)Test for Flavonoids**

182 One milliliter of plant filtrate were mixed with 2 ml of 10% lead acetate; a brownish precipitate
183 indicated a positive test for the phenolic flavonoids. While for flavonoids, 1 ml of the plant
184 filtrate were mixed with 2ml of dilute NaOH; a golden yellow colour indicated the presence of
185 flavonoids

186 **(vii)Test for Reducing Sugars**

187 One milliliter of the plant filtrate was mixed with Fehling A and Fehling B separately; a brown
188 colour with Fehling B and a green colour with Fehling A indicate the presence of reducing
189 sugars.

190 **(viii) Test for Cyanogenicglucosides.**

191 This was carried out subjecting 0.5g of the extract 10ml sterile water filtering and adding sodium
192 picrate to the filtrate and heated to boil (**Trease and Evans 1989**).

(ix) Test for Cardiac glucosides.

Legal test and the killer-kiliani was adopted, 0.5g of the extract were added to 2ml of acetic anhydrate plus H₂SO₄ (**Trease and Evans 1989**)

Quantitative Method of Analyses

(i) Saponins

About 20grams each of dried plant samples were ground and, put into a conical flask after which 100 ml of 20 % aqueous ethanol were added. The mixture was heated using a hot water bath. At about 55°C, for 4 hour with continuous stirring, after which the mixture were filtered and the residue re-extracted with a further 200 ml of 20% ethanol. The combined extracts were reduced to 40 ml over a water bath at about 90°C. The concentrate were transferred into a 250 ml separatory funnel and 20 ml of diethyl ether were added and then shaken vigorously. The aqueous layer were recovered while the ether layer was discarded. The purification process was repeated three times. 60 ml of n-butanol were added. The combined n-butanol extracts were washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath. After evaporation, the samples were dried in the oven to a constant weight, the saponin content was calculated as percentage of the starting material (**Osuntokun 2015**).

(ii) Flavonoids

About 10 g of the plant sample were extracted repeatedly with 100 ml of 80% aqueous methanol, at room temperature. The whole solution were filtered through Whatman filter paper No 42. The filtrate were later transferred into a crucible and evaporated into dryness over a water bath; the dry content were weighed to a constant weight (**Edeogaet al.,2005**).

(iii) Cardiac glucosides

Legal test and the killer-kiliani was adopted, 0.5g of the extract were added to 2ml of acetic anhydrate plus H₂SO₄ (**Trease and Evans 1989**).

(iv) Tannins

About 500 mg of the plant sample were weighed into a 50 ml plastic bottle. 50 ml of distilled water was added and shaken for 1 hour on a mechanical shaker. This was filtered into a 50 ml volumetric flask and made up to the marked level. Then, 5 ml of the filtrate was transferred into a test tube and mixed with 2 ml of 0.1 M FeCl₃ in 0.1 M HCl and 0.008 M potassium ferrocyanide. The absorbance were measured at 120 nm within 10 minutes. The tannins content was calculated using a standard curve of extract.

(v) Alkaloids

Five grams of the plant sample were weighed into a 250 ml beaker and 200ml of 10% acetic acid in ethanol was then be added, the reaction mixture were covered and allowed to stand for 4 hour. This were filtered and the extract will be concentrated on a water bath to one-quarter of the original volume. Concentrated ammonium hydroxide was added drop-wise to the extract until the precipitation is complete. The whole solution were allowed to settle and the precipitate was collected, washed with dilute ammonium hydroxide and then filtered; the residue being the alkaloid, which was dried and weighed to a constant mass (Trease and Evans 1989).

(vi) Phlobatannins

About 0.5 grams of each plant extracts were dissolved in distilled water and filtered. The filtrate were boiled in 2% HCl, red precipitate show the present of phlobatannins.

Total Phenol (Spectrophotometric methods): 2g each of the samples were defatted with 1ml of diethyl ether using a soxhlet apparatus for 2 h. The fat free samples were boiled with 50ml of ether for the extraction of the phenolic components for 15 minutes. 5ml of the extracts were pipetted into 5ml flask and then 10ml distilled water was added. 2ml of ammonium hydroxide solution and 5ml of concentrated amyl alcohol were also added. The samples were made up to mark and left to react for 30 min. for colour development. This was measured at 505 nm. (Osuntokun 2015), (Obadoni et al., 2001), (Boham, & Kocipai-Abyazam, 1974).

RESULTS-

Table 2: Antibacterial Activity of Aqueous Extract of Medicinal Plants at 20mg/mL, 40mg/mL, and 60mg/mL Concentrations.

Selected Clinical organisms	<i>Salmonella typhi</i>			<i>Staphylococcus aureus</i>			<i>Klebsiella pneumoniae</i>			<i>Escherichia coli</i>			Streptomycin (control)
Extracts	20 mg/mL	40 mg/mL	60 mg/mL	20 mg/mL	40 mg/mL	60 mg/mL	20 mg/mL	40 mg/mL	60 mg/mL	20 mg/mL	40 mg/mL	60 mg/mL	20 mg/mL
<i>Acacia albida</i>	10.0	12.0	13.0	10.0	12.0	14.0	10.0	12.0	13.0	4.0	7.0	10.0	2.50
<i>Anchomanes difformis</i>	10.0	11.0	12.0	12.0	15.0	16.0	10.0	11.0	12.0	3.0	5.0	7.0	2.40
<i>Bosciasenegalensis</i>	7.0	8.0	10.0	10.0	12.0	13.0	8.0	10.0	11.0	7.0	8.0	10.0	2.0
<i>Bridelia ferruginea</i>	9.0	12.0	13.0	13.0	14.0	15.0	5.0	7.0	8.0	3.0	6.0	8.0	3.0
<i>Ficus ingens</i>	7.0	14.0	15.0	10.0	15.0	19.0	5.0	8.0	10.0	6.0	7.0	8.0	2.7
<i>Indigofera arrecta</i>	6.0	10.0	13.0	10.0	11.0	12.0	7.0	9.0	9.5	2.0	6.0	7.0	3.5
<i>Moringa oleifera</i>	6.0	7.0	10.0	7.0	10.0	12.0	4.0	5.0	7.0	5.0	8.0	10.0	4.0

246 Unit of zone of inhibition- mm

247 Table 3: Antibacterial activity of Ethanol Extract of Medicinal Plants. At

248 20mg/mL, 40mg/mL, and 60mg/mL Concentrations.

Selected Clinical organisms	<i>Salmonella typhi</i>	<i>Staphylococcus aureus</i>	<i>Klebsiella pneumoniae</i>	<i>Escherichia coli</i>	Streptomycin (control)
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Extracts	20 mg/ mL	40 mg/ mL	60 mg/ mL	20 mg/ mL	40 mg/ mL	60 mg/ mL	20 mg/ mL	40 mg/ mL	60 mg/ mL	20 mg/mL	40 mg/mL	60 mg/mL	20 mg/mL
<i>Acacia albida</i>	2.0	5.0	6.0	1.0	2.0	4.0	3.0	5.0	7.0	2.0	3.0	4.0	3.0
<i>Anchomanes difformis</i>	3.0	5.0	7.0	1.0	4.0	6.0	2.0	5.0	8.0	1.0	3.0	8.0	2.9
<i>Boscia senegalensis</i>	2.0	2.0	5.0	1.0	3.0	5.0	2.0	4.0	5.0	2.0	3.0	6.0	4.5
<i>Bridelia ferruginea</i>	5.0	6.0	6.0	2.0	4.0	6.0	3.0	3.5	5.0	2.0	4.0	7.0	4.0
<i>Ficus Ingens</i>	8.0	10. 0	11. 0	10.	12. 0	13.	5.0	6.0	9.0	8.0	11.0	10	5.0
<i>Indigofera arrecta</i>	10.0	11. 0	13. 0	10. 0	11. 0	12. 0	7.0	8.0	10	2.0	7.0	8.0	4.0
<i>Moringa oleifera.</i>	8.0	1.0	12. 0	10. 0	11. 0	13. 0	4.0	5.0	8.0	1.0	5.0	7.0	3.5

249 Unit of zones of inhibition-mm

Selected Clinical organisms	<i>Salmonella Typhi</i>			<i>Staphylococcus saureus</i>			<i>Klebsiella pneumoniae</i>			<i>Escherichia coli</i>			Streptomycin (control)
Extracts	20 mg /mL	40mg g/mL	60 mg /mL	20 m g/ mL	40 mg /mL	60 mg/ mL	20mg g/mL	40mg /mL	60mg g/mL	20mg/ mL	40mg /mL	60mg/ mL	20mg/mL
<i>Acacia albida</i>	4.0	7.0	9.0	1. 0	2.0	3.0	4.0	7.0	9.0	4.0	7.0	9.0	1.0
<i>Anchomanes difformis</i>	1.0	1.0	8.0	1. 0	3.0	5.0	6.0	3.0	6.0	1.0	1.0	8.0	5.0

<i>Boscia senegalensis</i>	1.0	1.0	4.0	1.0	4.0	8.0	2.0	3.0	4.0	1.0	3.0	4.0	8.0
<i>Brideliafer ruginea</i>	3.0	3.0	5.0	2.0	3.0	5.0	1.0	2.0	7.0	3.0	4.0	6.0	7.0
<i>Ficusingen s</i>	1.0	1.0	2.0	1.0	5.0	8.0	1.0	4.0	8.0	1.0	4.0	5.0	5.0
<i>Indigofera arrecta</i>	1.0	4.0	8.0	1.0	4.0	7.0	1.0	3.0	6.0	1.0	4.0	8.0	5.0
<i>Moringa oleifera.</i>	1.0	7.0	9.0	1.0	4.0	8.0	1.0	4.0	8.0	1.0	7.0	9.0	8.0

250 **Table 4:Antibacterial Activity of Ethyl acetate Extract of Medicinal Plants At**
 251 **20mg/mL,40mg/mL, and 60mg/mL Concentration.**

252 Unit of zones of inhibition-mm

253

254 **Table 5:Antibacterial Activity of Hexane Extract of Medicinal Plants**
 255 **at20mg/mL,40mg/mL, and 60mg/mLConcentrations.**

Selected Clinical organisms	<i>Salmonella Typhi</i>			<i>Staphylococcus aureus</i>			<i>Klebsiellapnumoniae</i>			<i>Escherichia coli</i>			Streptomycin (control)
Extracts	20 mg /m L	40 mg /m L	60 mg /m L	20m g/m L	40 mg/ mL	60m g/m L	20 mg/ mL	40m g/m L	60m g/m L	20 mg/ mL	40m g/m L	60m g/mL	20mg/ mL
<i>Acacia albida</i>	1.0	3.0	4.0	0.0	1.0	6.0	2.0	5.0	8.0	3.0	5.0	10.0	5.0
<i>Anchoman</i>	5.0	6.0	7.0	4.0	5.0	10.0	5.0	6.0	7.0	4.0	8.0	10.0	4.0

257

TABLE 6; Qualitative Analysis Of Phytochemicals Screening Of The Medicinal Plants

[illegible]

Ficusingen s	+ ve	+ ve	- ve	+ ve	+ ve	+ ve	+ ve	- ve
Indigofera arrecta	+ ve	+ ve	+ ve	+ ve	+ ve	+ ve	+ ve	- ve
Moringaol eifera.	-ve	+ ve	- ve	+ ve	+ ve	+ ve	+ ve	+ ve

259 **Key- Positive (+ve) , Negative (-ve)**

260 **TABLE 7- Quantitative Analysis Of Minerals Present In Medicinal Plants Extract**
 261 **(mg/100g).**

Medicinal Plants sample used	Na	K	Ca	Mg	Zn	Fe	Pb	Cu	Mn	P
Acacia albida	22.7 3	29.1 1	30.4 5	26.0 5	28.0 4	7.88	ND	0.0 3	6.00	24.5 1
Anchomanesdifformi s	20.5 6	22,3 4	21.8 9	22.1 0	20.8 9	5.92	ND	0.0 4	6.34	25,9 1
Bosciasenegalensis	14.5 9	14.2 0	18.7 7	22.1 0	21.5 9	22.1 0	2.2 4	1.2 0	6.23	17.9 0
Brideliaferruginea	19.0 0	23.9 8	22.1 2	23.3 4	16.8 9	22.1 2	3.5 6	2.1 6	5.79	18.7 8
Ficusingens	20.9 2	23.1 2	29.3 4	24.7 8	17.3 4	20.3 4	2.7 8	3.0 0	4.92	27.6 4
Indigoferaarrecta	21.3 3	43.2 1	16.5 0	26.3 7	17.7 5	4.36	ND	ND	14.3 3	88.4 3
Moringaoleifera.	20.0	26.1	31.4	22.0	25.0	5.77	ND	0.0	6.20	25.1

	0	4	6	5	3			1		2
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262

263 **TABLE 8- Quantitative Analysis Of Anti- Nutrient Present In Medicinal Plants Extract In**

264 **Percentage (%).**

PARAMETERS	<i>Acacia albida</i>	<i>Anchomanes difformis</i>	<i>Bosciaseneg alensis</i>	<i>Brideliafer ruginea</i>	<i>Ficusin gens</i>	<i>Indigofer aarrecta</i>	<i>Moringaole ifera</i>
Tannin	2.20	2.10	2.32	2.37	2.30	2.25	ND
Phenol	2.50	3.57	2.56	2.49	3.49	3.45	ND
Phylate	16.30	15.27	15.68	15.79	12.33	12.45	2.25
Oxalate	3.66	3.74	6.57	6.51	8.51	8.55	1.50
Saponin	13.87	14.04	9.78	9.70	7.52	7.61	1.67
Flavonoid	8.59	8.52	6.43	6.56	10.38	10.41	ND
Alkaloids	3.23	1.25	4.25	4.31	4.36	4.37	
Tannin	2.20	2.10	2.32	2.37	2.30	2.25	ND
Phenol	2.50	3.57	2.56	2.49	3.49	3.45	ND

265

266 **TABLE 9- Quantitative Analysis Of Proximate Composition Of Medicinal Plants**
267 **Extract (%)**

Medicinal plants used	% Ash	% MC	% CP	% Fat	% Fibre	% CHO
<i>Acacia albida</i>	10.67	9.31	13.33	6.43	11.49	46.00
<i>Anchomanesdifformis</i>	11.69	9.46	12.87	6.46	11.41	47.12
<i>Bosciasenegalensis</i>	9.37	3.74	14.68	7.22	4.33	60.55
<i>Brideliaferruginea</i>	9.39	3.79	14.72	6.82	4.46	60.90
<i>Ficusingens</i>	8.72	7.32	16.25	5.34	8.53	53.72
<i>Indigoferaarrecta</i>	8.78	7.33	16.19	5.40	8.50	53.79
<i>Moringaoleifera.</i>	10.53	9.00	14.45	6.50	10.37	42.00

268 **Keys: MC=Moisture content CP=Crude protein. CHO=Carbohydrate.**

269 DISCUSSION

270 The Aqueous , Ethanol, Hexane and **Ethyl acetate** extracts of the leaves, and roots of
271 *Acacia albida*, *Anchomanesdifformis*, *Bosciasenegalensis*, *Brideliaferruginea*.
272 *Ficusingens*, *Indigoferaarrecta* and *Moringaoleifera* were examined for possible properties of
273 antimicrobial and phytochemical activities against the selected clinical organisms. The clinical
274 organisms used are *Salmonella typhi*, *Staphylococcus aureus*, *Klebsiellapnumonniae* and
275 *Escherichia coli*.

276 It were observed that in Aqueous extract of *Anchomanesdifformis*,
277 *Ficusingens*, *Moringaoleifera* and *Bosciasenegalensis* shows appreciable high antibacterial
278 activity between 10 mg/mL and 19 mg/mL and *Acacia albida*, *Brideliaferruginea* and
279 *Indigoferaarrecta* show low antibacterial activity between the ranges of 6mg/mL to 12 mg/mL.
280 This result conform with the finding of Indrayanet al.,2002, Osuntokun&olajubu 2014, the
281 action of plant against microorganism.

282 In ethanol extract shows appreciable high antibacterial activity in the plant extract of
283 *Anchomanesdifformis*, *Ficusingens*, *Indigoferaarrecta* and *Moringaoleifera* while in *Acacia*
284 *albida*, *Bosciasenegalensis*, *Brideliaferruginea* shows appreciable low zones of inhibition

between the range of 6ml/mg to 12 mg/mL, this conform to the fact that plant are probable source of antimicrobial and therapeutic agent against some nosocomial organisms especially the organism found in pediatrics hospital and other hospital related organisms , if this is possible that is , plant serving as a good antibiotics , then it can be consumed in are fined form in our day to day activity, like in our coffee tea , beverages, soup ingredient ,food seasons and etc. this is a proof evident by **De and James 2002**.

In ethyl acetate extract, plant shows high zones of inhibition between 7mg/mL and 10mg/mL in the plant extract of ***Anchomanesdifformis*, *Indigoferaarrecta*while *Acacia albida*, *Bosciasenegalensis*, *Brideliaferruginea* and *Ficusingens*shows** low zone of inhibition between 1mg/mL and 5mg/mL.***Anchomanesdifformis* and *Indigoferaarrecta*are** good source of inane booster in the case of HIV patient and some researchers are in the advent of making a good HIV drugs from the duo of ***Anchomanesdifformis*, and *Indigoferaarrecta***. This another proof that medicinal plant is a good source of therapitic agent. (**Lambert, 2002; Russell, 2002**). **Hexane plant extract show s** appreciable antibacterial activity **in hexane plant extract of *Anchomanesdifformis*, *Bosciasenegalensis*, *Brideliaferruginea*, *Indigoferaarrecta*and *Moringaoleifera*and** low zone so of inhibition in ***Acacia albida* and *Ficusingens***.

The activities of all the medicinal plants extracts ,predicts the presence of antimicrobial evidence in the medicinal plants under study, it also brighten the hope for the advents of new antibiotics which can be derived from medicinal plants source but not only new antibiotics but evidence for new drugs for the cure of different infection associated with pathogen microorganism, under which gram positive and gram negative bacteria has been adequately taken into consideration(**Osuntokun 2015**).

Nigerian populace are fed-up with the foreign conventional drugs which can be , and always been adulterated by some disgruntle elements in the society, but , there is need for the herbal drugs from our natural environment must be developed. This herbal drugs can not be adulterated, but can be improved upon, by the standardizing the production and herbal practice, due to the fact that medicinal plants are all over the four geo-political zone of the countr**y.in** other word the production and the use of the medicinal plant must be standardized and encouraged which can also be a source of revenue to the Nigerian country.

The pediatrics hospital, little or nothing is known about the presence of microorganism in this health care facility, but with this present study, it is a fact that microbes can also be present and isolated in this health care facility, putting into consideration that children are prone to microbe attack even if they are still in the hospital. Adequate care must be taken to reduce the presence of pathogen in the pediatrics hospital to avoid cross contamination and reemergence of infectious diseases.

In comparing the agar well diffusion and paper disc diffusion, both are good methods but, agar well diffusion is more preferable than paper disc diffusion because of the result obtained during both processes, the result in agar well are more substantial evidence of antimicrobial activity than the paper disc diffusion. In my opinion, agar well is more recommendable for research purposes.

Ogundare *et al.*, (2006) shows that plant species of *Anchomanes difformis*, *Indigofera arrecta* and probable *V. tenoreana* containing saponins, flavonoids, tannins and anthraquinones which were found to have very potent antibacterial as well as antifungal activities. These phytochemical constituents were further reported to be responsible for many antimicrobial activities of different plant species (Ghosh *et al.*, 1996; Iwu *et al.*, 1999). Flavonoids have been reported to be synthesized by plants in response to microbial infections and are good antibacterial agents; tannins have been demonstrated to have antibacterial activities (Akiyama *et al.*, 2001), (Osuntokun 2015).

Proximate composition of the medicinal plant under study, shows that, carbohydrate, moisture, fat, fibres, ash and crude protein are all present inclusive in plant analysis. It was observed that carbohydrate has the highest proportion followed by crude protein. Scientifically, these two proximate quantities are the most important element in life. Carbohydrate manufactures our energy and protein serves as the building blocks of life, for repair of worn out tissues and other metabolic processes.

The importance of phytochemicals must be clearly emphasized in this present study. Phenols is another phytochemical that were found in the medicinal plant, they are weakly acidic, Hydroxyl group attached directly to an aromatic ring, it serves as Antiseptic, anti-inflammatory, antimicrobial, anti-tumor and also a good disinfectant in its activity which is found in table 6 Ogundare *et al.*, (2006).

343 Flavonoids is Water soluble, super antioxidant and free radical scavenger.it is used in the
 344 In prevention of oxidative cell damage, allergies free radicals, microbesits active as
 345 Antioxidant, anticarcinogens, antimicrobial, antitumor. Saponins were observe to have
 346 Bitter taste, foaming property, haemolytic effect on red blood cells and Emulsifying agent.
 347 it were also observed that saponin containing plant are good Expectorant, cough
 348 suppressant, hemolytic activity(Osuntokun&Oluwafoise 2015).

349 Tannins were observable in medicinal plant under study. they have Unpleasant taste, tans
 350 leather, used in the production of leather and ink; in treating wounds, varicose ulcers,
 351 hemorrhoids ,frostbite and burns, and it has a Soothing relief, regenerates skin, anti-
 352 inflammatory and diuretics in its activity(Osuntokun&Oluwafoise 2015).in other word
 353 phytochemical and medicinal plants plays unquantifiable role in our day to day activities, this
 354 important were x-rayed in this research work.

355 CONCLUSION

356 The medicinal plants has an antibacterial and phytochemical effectiveness on the tested bacterial
 357 isolates resulted within 24 hours of incubation of the organism to all the plant extract. The
 358 aqueous, ethanol and ethyl acetate extract display a high level of antibacterial activity compared
 359 to the extract of hexane, but all the plant tested against the clinical organism have a certain degree
 360 of antibacterial activity, the best activity were found in Anchomanesdifformis in aqueous extract,
 361 Moringaoleifera in ethanol and ethyl acetate extract and Indigoferaarrectain hexane extract
 362 have little or less activity against the organism, knowing well that the hexane extract do not
 363 really do well in this research work, but in all, all plant were proven active because of their
 364 inhibitory zone of action .this result shows that medicinal plants have a great effect on clinical
 365 organism found on pediatric hospital in Ekiti and Ondo state.

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