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Phytochemical Composition and Antimicrobial Activity of *Daniella oliveri* Extracts on Selected Clinical Microorganisms

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Authors' contributions

This work was carried out in collaboration among all authors. Author OOT is the leading author, who designed, analyzed, interpreted and prepared the manuscript for publication. Author OOT is a researcher who researched the antimicrobial and phytochemical properties of various medicinal plants in Nigerian and Africa. Authors OAF, AFO and BOO did the proof reading of the entire manuscript to prepare it for publication and also helped during the antimicrobial assays. Author AOT helped to proof read the first and the final manuscript before publication. Authors AMYB and OOI helped during the phytochemical procedure and the chemical analysis of the plant extracts. All authors read and approved the final manuscript.

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ABSTRACT

Daniella oliveri (Caesalpiniacea) is a plant found in the Amazon region and other parts of South America and Africa. The tree may reach a height of 100 feet and trunk diameter of 4 feet. *Daniella oliveri* (Rolfe) Fabiaceae is commonly known as Ilorin balsam (eepo-iya) or *Copaihu africana*. It is an indigenous African tree, found extensively in Benin, Cameroon, Gambia and Nigeria. The basic objective of this project is to determine the antimicrobial activity and phytochemical composition of *Daniella oliveri.* The leaf and bark of the *Daniella oliveri* were obtained from a location in the south western part of Nigeria, in the tropical rainforest of Ikare Akoko, Ondo state and Ile Ife, Osun state, Nigeria. The plants were authenticated by a certified botanist at the herbarium unit of Department of Plant science and Biotechnology, Adekunle Ajasin University, Akungba Akoko, Ondo State, Nigeria and Obafemi Awolowo University, Ile Ife, Osun State, Nigeria. The leaf and bark were prepared using ethanol. The antimicrobial assay was carried out on leaf and bark using Agar diffusion method and the zone of inhibition was measured. The test organisms used for this research work are *Escherichia coli, Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella pneumonia, Bacillus cereus, Proteus vulgaris, Streptococcus viridians, Shigella dysenteriae, Bacillus subtilis* and *Salmonella typhi.* All the extracts (leaf and bark) of the highest concentration (60 mg/ml) has the highest zone of inhibition while the lowest concentration (7.5 mg/ml) has the lowest zone of inhibition. The phytochemical analysis revealed the presence of alkaloids and phenol, tannin, phylate, oxalate, saponin and steroid. The presences of these compounds were inferred as being responsible for the anti-microbial properties of the extracts. Mineral elements composition content present in the plant are sodium (Na), potassium (K), calcium (Ca), Magnesium (Mg), Zinc (Zn), Iron (Fe), Lead (Pb), therefore, the use of medicinal plants should be studied in detail and encouraged for use worldwide to reduce the level of resistance of microorganisms to drugs and make drugs available for the populace.

Keywords: Antimicrobial activity; phytochemical activity; proximate composition; mineral elements constituent; anti-nutrient composition.

1. INTRODUCTION

Daniella oliveri (Caesalpiniacea) is a plant found in the Amazon region and other parts of South America and Africa. The tree may reach a height of 100 feet and trunk diameter of 4 feet [1]. It produces liquid oleoresin which has been used as medicine by indigenous people for more than 400 year [2]. The oleoresin is produced in the tree's trunk, stem, and leaves and it consists of large but varying amounts of volatile oils (primarily composed of sesquiterpene hydrocarbons usually including caryophyllene), non volatile resinous substances and small quantities of acids. The oleoresin is traditionally used as an anti-inflammatory agent and in the treatment of a variety of genito-urinary tract diseases and skin ailments. Moreover, it is used as an anti-rheumatic, antiseptic, antibacterial, diuretic, and hypotensive agent, and also as an expectorant, laxative, purgative, vermifuge and vulnerary [3]. The leaves are also used in folk medicine as an anti-diabetic agent. Modern scientific studies have authenticated some of these medicinal uses of oleoresin such as its effectiveness as an antibacterial, anti-inflammatory, and anti-oxidant agent [4].

Daniella oliveri (Rolfe) Fabiaceae is commonly known as Ilorin balsam (eepoiya) or *copaihu africana*. It is an indigenous African tree found extensively in Benin, Cameroon, Gambia and Nigeria [5]. The leaves are used traditionally to treat diabetes and yellow fever [6]. The leaves were found to contain quercitrin, quercameritrin, rutin and the rare flavoured glycoside quercitin-3-methoxy 3-o-rhamnosylpranosyl [7,8]- β- d-Glycopyranoside (Narssine) isolated from n-butanol extract [9].

The roots, stem, bark and leaves of *Daniella oliveri* are used in the treatment of scrotal elephantiasis, dysentery, ring worms, syphilis, typhoid fever, eye sore and ear ache and the stem bark of *Daniella oliveri* was used in the treating fever, boil and back ache. The plants are found in both temperate and tropical regions of the world with 630 genera and species. Substances derived from medicinal plants remain the basis for a large proportion of commercial medications for the treatment of various diseases like heart diseases, high blood pressure, pains, asthma, malaria, typhoid fever, snake bites, arrow poison and other problems. These

substances are available in a variety of forms. [10].

The use of plant parts in the treatment of human disease is as old as the disease themselves, and herbal medicine was the major form of medicine in Nigeria. Infectious diseases are major cause of death in developing countries and today according to WHO as many as 80% of world population depends on traditional medicines for their primary health care needs and that 25% of the drugs are based on plants and their derivatives [11,12].

Moreover, the plant is used as an anti-rheumatic, antiseptic, antibacterial, diuretic, and hypotensive agent, and also as an expectorant, laxative, purgative, vurmifuge, and vulnerary. The leaves are also used in folk medicine as an anti- diabetic agent. Modern scientific studies have authenticated some of these medicinal uses of oleoresin such as its effectiveness as an antibacterial, anti- inflammatory, and anti- oxidant agent [13,14].

In Cote d'Ivoire, the gum is considered aphrodisiac and diuretic, and it is chewed to treat cough, headache, tachycardia and painful menstruation. The gum is externally applied to itching skin and skin diseases. The gum and bark are taken in various preparations, internally and externally, and sometimes with other plant parts, to treat veneral diseases, ulcers and sores, circumcision wounds, leprosy, dysentery, colic, menstrual problems, cough, cold, angina bronchitis, tuberculosis, kidney problems, appendicitis, headache, backache, rheumatism, fever pains, hernia, tooth-ache, and snakebites [15]. The root is considered diuretic and a decoction is taken to treat veneral diseases, absence of menses, anxiety, insanity, food poisoning, and skin diseases. Leafy twigs are put in baths to treat fever and jaundice, and also as a tonic. A decoction of the leafy twigs with salt is taken as a purgative, to treat constipation and stomach-ache [16,17].

2. MATERIALS AND METHODS

2.1 Collection and Identification of Samples

The leaf and bark of the *Daniella oliveri* were obtained from a location in the south western part of Nigeria, in the tropical rainforest of Ikare Akoko, Ondo state and Ile Ife, Osun state, Nigeria. The plant were authenticated by a certified botanist at the herbarium unit of Department of Plant science and Biotechnology, Adekunle Ajasin University, Akungba Akoko, Ondo State, Nigeria and Obafemi Awolowo University, Ile Ife, Osun state, Nigeria. The leaves, bark and stem were washed thoroughly with distilled water, stored in air tight containers and kept at room temperature prior to use [18].

2.2 Test Organisms

The test bacteria used in this study were *Salmonella typhi, Klebsiella pneumoniae, Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa, Shigella dysenteriae, Bacillus cereus, Streptococcus viridians, Proteus vulgaris, Bacillus subtilis*. They were obtained from Adekunle Ajasin University, Akungba Akoko, Ondo State, Nigeria and Obafemi Awolowo University, Ile Ife. Osun State, Nigeria They were isolated on sterile nutrient agar slants and taken to the microbiology laboratory of the Adekunle Ajasin University, Akungba Akoko, Nigeria. All slants of test organisms were kept at -4℃ prior to bioassay of the extracts. Extensive biochemical tests were carried out to further confirm all the test bacterial strains [19,20].

2.3 Preparation of Plant Extracts for Extraction

All the plant materials obtained were first washed thoroughly with sterile distilled water and air dried at room temperature for about two weeks to ensure that the samples lose most of their moisture content. The following extractions were carried out: aqueous and Ethanol. For each extraction, 400 g of each dried plant material was weighed separately into conical flasks containing 1200 ml each of distilled water and ethanol, The mixtures were initially shaken rigorously and left for 9 days. All mixtures were filtered using sterile Whatman filter papers and the filtrates were collected directly into sterile crucibles. All filtrates obtained were introduced into sterile reaction tubes and heated continuously in water bath at the following temperatures: 78℃ for ethanol extraction, and 105℃ for distilled water. The residues obtained were kept at room temperature [21].

2.4 Standardization of Extracts

Using aseptic condition, the extract is reconstituted by adding 1.2 g of each extract with 5 ml of dimethylsulphoxide (DMSO) and 15 ml of sterile distilled water making it 60 mg/ml. For

each extract, 7.5 ml of distilled water is measured into three sterile bijou bottle. In bijou bottle A 7.5 ml from 60 mg/ml extract was added and in bijou bottle B 2.5 ml from 60 mg/ml extract was added and bijou bottle C 2.5 ml from bijou bottle A was added. A is 30 mg/ml, B is 15mg/ml, C is 7.5 mg/ml respectively [22].

2.5 Standardization of Inoculum

Slants of the various organisms were reconstituted using an aseptic condition. Using a sterile wire loop, approximately one isolated colony of each pure culture was transferred into 5 ml of sterile nutrient broth and incubated for 24 hours. After incubation, transfer 0.1 ml of the isolated colony using a sterile needle and syringe into 9.9 ml of sterile distilled water contained in each test tube and then mixed properly. The liquid now serve as a source of inoculum containing approximately 10^6cfu/ml of bacterial suspension [23].

2.6 Antimicrobial Assay of Plants Extracts Using Agar Well Diffusion Method

All antibacterial assays for the plant extracts were carried out by well diffusion technique. All the test organisms were sub-cultured onto sterile Mueller Hinton Agar plates and incubated at 37℃ for 18-24 hour. Five distinct colonies for each organism were inoculated onto sterile Mueller Hinton broth and incubated for 3-4 h. All innocula were standardized accordingly to match the 0.5 McFarland standard and this standard was used for all susceptibility tests. All the extracts were reconstituted accordingly into the following concentrations: 60, 30, 15 and 7.5 mg/ml; using the Dimethyl Sulphoxide (DMSO) [24].

The susceptibility testing was investigated by the Agar well diffusion method. A 0.1 ml of 1:10,000 dilutions (equivalent to 10^6cfu/ml) of fresh overnight culture of the clinical isolates grown in Muller Hilton agar was seeded into 40 ml of Mueller Hilton agar, and properly mixed in universal bottles. The mixture was aseptically poured into sterile Petri dishes and allowed to set. Using a sterile cork borer of 4 mm diameter, equidistant wells were made in the agar. Drops of the re-suspended, (2 ml per well) extracts with concentrations between 60 to 7.5 mg/ml were introduced into the wells till it was filled. Ciprofloxacin (2 mg/ml) was used as the control experiment.

The plates were allowed to stand on the bench for an hour, to allow pre-diffusion of the extracts before incubation at 37℃ for 24 hours. The zones of inhibition were measured to the nearest millimetre (mm) using a standard transparent meter rule. All experiments were performed in duplicates [25].

2.7 Phytochemical Screening, Mineral Elements Composition, Anti-Nutrient and Proximate Composition of *Daniella oliveri*

The determination of quantitative, quantitative, micro-nutrient composition, anti-nutrient and proximate compositions of Daniella olivera were elucidated below.

2.8 Qualitative Method of Analyses

2.8.1 Preliminary test / preparation test

Plant filtrates were prepared by boiling 20 g of the fresh plant in distilled water. The solution was filtered through a vacuum pump. The filtrate were used for the phytochemical screening for flavonoids, tannins, saponins, alkaloids, reducing sugars, anthraquinones and anthocyanosides. [26].

(i) Test for Alkaloids -About 0.2gram was warmed with 2% of H_2SO_4 for two minutes, it was filtered and few drops of Dragendoff's reagent were added. Orange red precipitate indicates the present of Alkaloids [26].

(ii) Test for Tannins -One milliliter of the filtrate was mixed with 2 mi1 of Fec13, A dark green colour indicated a positive test for the tannins [27].

(iii) Test for Saponin -One milliliter of the plant filtrate was diluted with 2 mil of distilled water; the mixture were vigorously shaken and left to stand for 10 minutes, during which time, the development of foam on the surface of the mixture lasting for more than 10 minutes, indicates the presence of Saponin [28].

(iv) Test for Anthraquinones- One milliliter of the plant filtrate was shaken with 10 mil of benzene; the mixture was filtered and 5 mil of 10% (v/v) ammonia were added, then shaken and observed. A pinkish solution indicates a positive test [13,14].

(v) Test for Flavonoid- About 5 mL of each aqueous extracts was added with 1% NH_3 solution. A positive test result was

confirmed by the formation of a yellow coloration or turbidity [29].

(vi) Test for Cardiac Glycoside- About 5 mil of the extract was mixed with 2 mil of glacial acetic acid containing one drop ferric chloride solution. To this, 1 mil of concentrated sulphuric acid was slowly underplayed to the sample mixture. A positive test result was confirmed by the presence of a brown ring at the Interface [30].

(vii) Test for steroids -10 mil of each ethanol extract are evaporated to insipient dryness over a steam bath and cooled to room temperature. It was then defatted repeatedly with hexane. The defatted aqueous layer was then warmed over a steam bath to remove the residual hexane. To this, 3 mil of $Fecl_3$ reagent was added and 1 mil of concentrated sulfuric acid was then slowly added. A positive test was evident when a reddish brown coloration occurred [30].

(viii) Total Phenol -(Spectrophotometric Methods) 2 g of each sample, 1 ml of diethyl ether was added for defatting. The fat free samples were boiled with 50 ml of ether for 15 min to obtain the phenolic components which were measured at 505 nm following the standard method [30].

2.9 Quantitative Method of Analyses

2.9.1 Saponin

About 20 grams 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 were heated using a hot water bath. At about 55℃, 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℃. The concentrate was 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 m1 of 5% aqueous sodium chloride. The remaining solution were 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 [31].

2.9.2 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 weigh [32,33].

2.9.3 Cardiac glucosides

Legal test and the killer-kilianiwwas adopted, 0.5 g of the extract were added to 2 ml of acetic anhydrate plus H_2S0_4.

2.9.4 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 $FeCl_3$ in 0.1 M Hcl_3 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. [26].

2.9.5 Alkaloids

Five grams of the plant sample were weighed into a 250 ml beaker and 200 ml of 10% acetic acid in ethanol was then be added, the reaction mixture were covered and allowed to stand for 4 hour. These were filtered and the extract 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 [29].

2.9.6 Phlobatannin

About 0.5 grams of each plant extracts were dissolved in distilled water and filtered. The filtrates were boiled in 2% HCl, red precipitate show the present of phlobatannin [34].

2.10 Total Phenol (Spectrophotometric Method)

2 g each of the samples were defatted with 1 mL of diethyl ether using a soxhlet apparatus for 2 hours. The fat free samples were boiled with 50 mL of ether for the extraction of the phenolic components for 15 minutes. 5 mL of the extracts were pipetted into 5 mL flask and then 10 mL distilled water was added. 2 mL of ammonium hydroxide solution and 5 mL of concentrated amyl alcohol were also added. The samples were made up to mark and left to react for 30 minutes for colour development. This was measured at 505 nm [35].

2.11 Determination of Proximate Analysis of *Daniella oliveri*

The proximate parameters (moisture, dry matter, ash, crude fats, proteins and fibers, nitrogen, carbohydrates and energy values) were determined using Association of Official Analytical Chemists Methods [36,18].

1. Determination of moisture content was done by drying samples in oven (WiseVen, WON-50, Korea) at 110℃ until constant weight was attained [37].
2. Nitrogen estimation was carried out by the micro-Kjeldahl (BUCHI, KjelFlex K-360, Switzerland) method with some modification [21].
3. The crude proteins were subsequently calculated by multiplying the nitrogen content by a factor of 6.25 [21]. The energy value estimation was done by summing the multiplied values for crude protein,
4. Crude fat and carbohydrate respectively at Water Factors. Crude fats were determined by Soxhlet apparatus using *n*-hexane as a solvent.
5. The ash values were obtained by heating samples at 550℃ in a muffle furnace (Wise Therm, FHP-03, Korea) for 3 hour [38].
6. The carbohydrate content was determined by subtracting the total crude protein, crude fibre, ash content and crude fat from the total dry matter [38].
7. Crude fibre was estimated by acid-base digestion with 1.25% H_2SO_4 (v/v) and 1.25% NaOH (w/v) solutions [39].

3. RESULTS

The leaf and bark of *Daniella oliveri* was soaked with ethanol and left for 7 days. Table 1 shows the yield of the extracts after separation and evaporation.

Table 2 shows the antimicrobial activities of ethanolic leaf extract of *Daniella oliveri* at concentrations 60, 30, 15 and 7.5 mg/ml tested against the some selected clinical isolates. There was a greater zone of inhibition when tested against *Shigella dysentariae* (15 mm), *Proteus vulgaris* (14 mm), and *Pseudomonas aeruginosa* (13 mm) at 60 mg/ml.

Table 3 shows the antimicrobial activity of ethanolic bark extract of *Daniella oliveri* at concentrations 60, 30, 15 and 7.5 mg/ml tested against the same clinical isolates. The extract has greater inhibition zones against *Staphylococcus aureus* (14 mm), *Shigella dysentariae* (14 mm) and *Bacillus subtilis* (14 mm) at 60 mg/ml.

Table 4 shows minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of *Daniella oliveri* extracts. The Minimum Inhibitory Concentration of *Daniella oliveri* plant extracts at concentrations 60, 30, 15, 7.5, 3.75, 1.88 and 0.94 mg/ml. It was observed that the organisms grow at concentrations 7.5, 3.75, 1.88 and 0.94 mg/ml indicating that the plant is not effective at those concentrations, while at concentration 15, 30, and 60 mg/ml there was no growth. This indicates that the plant is effective at higher concentrations. At 60 mg/ml, the minimum inhibitory concentration is recorded while the minimum bactericidal concentration is recorded at 15 mg/ml

Table 5 shows the quantitative analyses of the phytochemicals present in the plant. It shows the presence of phytochemicals such as alkaloid, saponin, tannin, glycoside, flavonoid, phenol, steroid present in the leaf and shows that alkaloid, steroid and flavonoid is not present in the bark of the plant.

Table 6 shows all the mineral elements present in the leaf and bark extract and they include sodium, potassium, calcium, magnesium, zinc, iron, lead, copper, manganese and phosphorus. The result shows that there is a large amount of Potassium, magnesium and sodium in both leaf and bark while both has a low amount of iron.

Table 7 shows the analysis of the anti-nutrient present in the plant extract calculated in percentage (%). Saponin (10.54%) and flavonoid (7.92%) has the highest value for leaf while alkaloid (1.98%) and oxalate (3.92%) has the lowest anti-nutrient composition. The result

indicates that saponin is more present in the bark (9.34%) and leaf (10.54%) of the plant.

Table 8 shows the proximate nutrient composition of bark and leaf which has their highest nutrient composition in carbohydrate with 42.59 and 45.29 for bark and leaf respectively. They have their lowest value for the two extract in their fat with 6.59 and 7.21 respectively.

Table 1. Percentage yield of *Daniella oliveri* extracts

Percentage yield	Initial weight of the sample (g)	Volume used (ml)	Percentage yield of final weight (g)
Percentage yield of *Daniella oliveri* leaf extract	400	1200	8.7
Percentage yield of *Daniella oliveri* bark extract	400	1200	8.2

Table 2. Antimicrobial activity of ethanolic leaf extracts of *Daniella oliveri*

Clinical organism	Concentration 60 mg/ml	Concentration 30 mg/ml	Concentration 15 mg/ml	Concentration 7.5 mg/ml	Concentration Ciprofloxacin 30 mg/ml
Escherichia coli	12.0	7.0	3.0	1.0	20.0
Salmonella typhi	11.0	7.0	2.0	0.0	19.0
Proteus vulgaris	14.0	9.0	4.0	1.0	20.0
Klebsiella pneumonia	12.0	10.0	2.0	0.0	28.0
Bacillus cereus	12.0	7.0	2.0	0.0	19.0
Staphylococcus aureus	10.0	6.0	1.0	0.0	20.0
Pseudomonas aeruginosa	13.0	6.0	1.0	0.0	22.0
Streptococcus viridians	12.0	7.0	4.0	1.0	18.0
Bacillus subtilis	10.0	5.0	3.0	0.0	19.0
Shigella dysentariae	15.0	8.0	4.0	1.0	20.0

Zone of inhibition is measured in mm

Table 3. Antimicrobial activity of ethanolic bark extracts of *Daniella oliveri*

Selected clinical organism	Concentration 60 mg/ml	Concentration 30 mg/ml	Concentration 15 mg/ml	Concentration 7.5 mg/ml	Ciprofloxacin 30 mg/ml
Escherichia coli	11.0	5.0	3.0	1.0	20.0
Salmonella typhi	12.0	4.0	1.0	0.0	19.0
Proteus vulgaris	11.0	6.0	2.0	1.0	20.0
Klebsiella pneumonia	10.0	9.0	4.0	0.0	28.0
Bacillus cereus	8.0	5.0	1.0	0.0	19.0
Staphylococcus aureus	14.0	8.0	3.0	1.0	20.0
Pseudomonas aeruginosa	13.0	7.0	3.0	0.0	22.0
Streptococcus viridians	13.0	6.0	2.0	1.0	18.0
Bacillus subtilis	14.0	6.0	3.0	1.0	19.0
Shigella dysentariae	14.0	7.0	4.0	1.0	20.0

Zone of inhibition is measured in mm

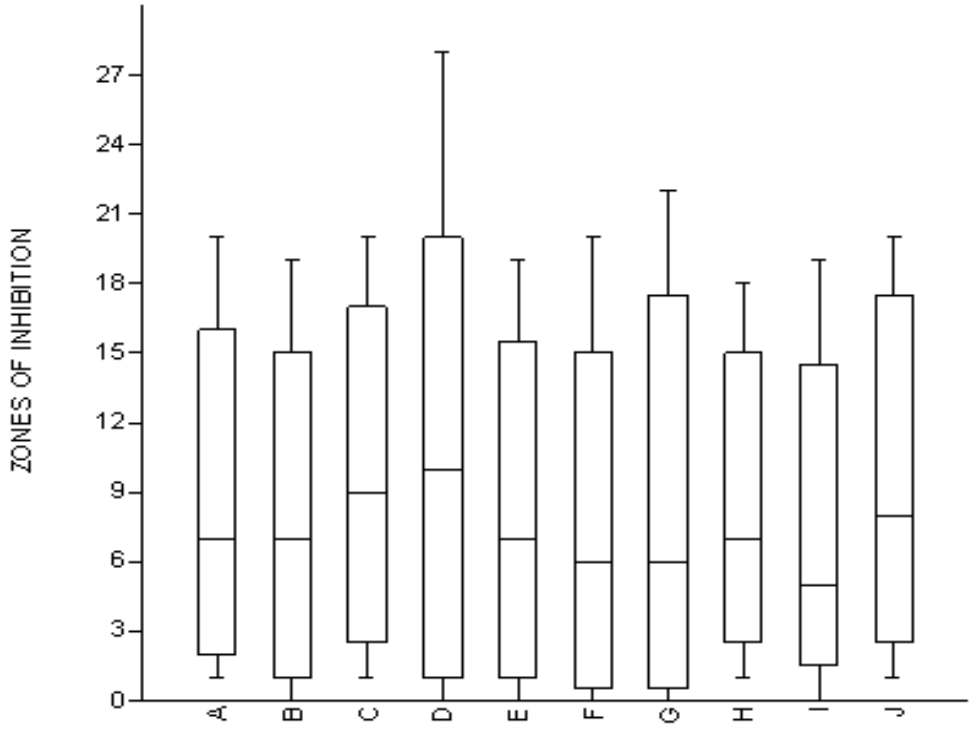

Fig. 1. A graphical representation showing the activity of *Daniella oliveri* leaf extract on some selected clinical isolates

Table 4. Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of *Daniella oliveri* extracts

Concentration mg/ml	Leaf ethanol	Bark ethanol
60	NG	NG
30	NG	NG
15	NG	NG
7.5	G	G
3.75	G	G
1.88	G	G
00.94	G	G

Key: G: Growth; NG: No Growth
Note: At 60 mg/ml, the minimum inhibitory concentration is recorded while the minimum bactericidal concentration is recorded at 15 mg/ml

4. DISCUSSION

Several investigations had reported that *Daniella oliveri* and other medicinal plants contain antimicrobial substances [40]. The result of this present study agrees with the reports of these previous workers, who reported that the plant has a wide range of some active plant substances against certain organisms. *Daniella oliveri* extracts have been used for many thousands of years in food preservation [41,42], pharmaceuticals, alternative medicine and natural therapies [41].

It is necessary to investigate those plants scientifically which have been used in traditional medicine to improve the quality of healthcare. *Daniella oliveri* extracts are potential sources of novel antimicrobial compounds especially against *Escherichia coli, Pseudomonas aeruginosa* [30] *Staphylococcus aureus*, *Klebsiella pneumonia, Bacillus cereus, Proteus vulgaris, Streptococcus viridians, Shigella dysenteriae, Bacillus subtilis* and *Salmonella typhi* [29,2,1].

Antimicrobial properties of plants are desirable tools in the control of undesirable microorganisms especially in the treatment of infectious diseases which correlates with research of Aboaba and Efuwape, [1] and also in correlation with this present study. The active components usually interfere with growth and metabolism of microorganisms in a negative manner [42].

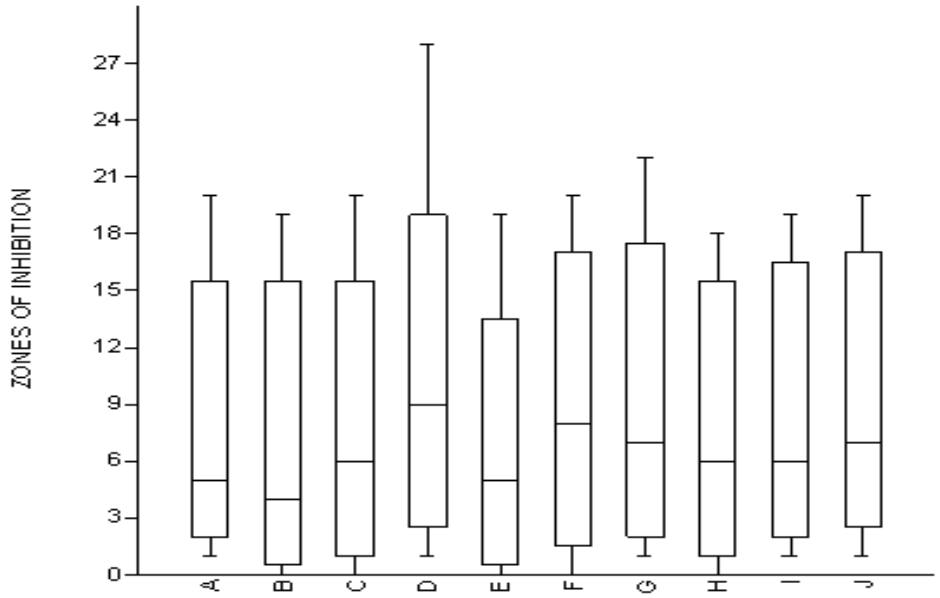

Fig. 2. A graphical representation showing the activity of *Daniella oliveri* bark extract on some selected clinical isolates

Key- Escherichia coli - A; Salmonella typhi- B; Proteus vulgaris- C; Klebsiella pneumonia- D; Bacillus cereus- E; Staphylococcus aureus- F; Pseudomonas aeruginosa- G; Streptococcus viridians- H; Bacillus subtilis- I; Shigella dysentariae - J

Table 5. Qualitative analysis of the phytochemical screening of *Daniella oliveri extracts*

Sample	Alkaloid	Cardiac glycoside	Steroid	Anthraquinone	Phenol	Tannin	Saponin	Flavonoids
BARK *Daniella oliveri*	- ve	+ ve	-ve	+ ve	+ ve	+ ve	+ ve	- ve
LEAF *Daniella oliveri*	-+ve	+ ve	+ve	+ ve	+ ve	+ ve	+ ve	+ ve

Table 6. Quantitative analysis of Mineral elements composition present in *Daniella oliveri* extracts (mg/100 g)

Plant sample used	Na	K	Ca	Mg	Zn	Fe	Pb	Cu	Mn
BARK *Daniella oliveri*	20.33	41.24	15.53	25.38	18.71	4.38	ND	ND	15.34
LEAF *Daniella oliveri*	23.47	42.46	17.47	21.43	16.92	5.20	ND	ND	16.00

Preliminary phytochemical screening showed that ethanolic extracts of *Daniella oliveri* contain most of the phytochemicals like alkaloids, tannins, saponins, quinine and anthraquinone. It was discovered that the presence of alkaloid, saponin, steroid and phenol explain why the leaves can be used in the treatment of malaria, ring worms, syphilis, typhoid fever, eye sore and ear ache, head ache and other forms of fever and the stem bark of *Daniella oliveri* were used in the treating fever, boil and back ache while the tannin explain why it is used for treatment of dysentery according to [28] which explain the antimicrobial efficacy of saponin and steroid against clinical organisms.

Table 7. Quantitative analysis of anti –nutrient present in *Daniella oliveri* extracts (result in percentage (%))

Parameters	Bark *Daniella oliveri*	Leaf *Daniella oliveri*
Tannin	2.34	2.73
Phenol	3.21	3.94
Phylate	2.97	2.03
Oxalate	2.31	3.92
Saponin	9.34	10.54
Flavonoid	8.43	7.92
Alkaloids	1.87	1.98

The presence of these bioactive components in the crude extracts has been linked to their activities against disease causing microorganisms and also offering the plant themselves protection against infection by pathogenic organisms [12].

The high percentage of saponin among plant parts used leaf has the highest antinutrient which is (10.54%), and the phytochemicals result also suggest the activities present in *Daniella oliveri* extract of leaf and bark may be due to presence of tannin and this finding correlate with the observation of [23] who reported the antibacterial efficacy of flavonoids and tannin against clinical organisms and these findings relate with the observations of [27] who related that these bioactive components are known to show medicinal activity as well as exhibiting physiological activity [13] and exhibit anti-inflammatory, anti-oxidant, and membrane stabilizing property [39].

The presence of terpenoid has been reported to be useful in herbal medicines [9]. These phytochemicals also have some strong antimicrobial significance against some potential enteric pathogens; the presence of Alkaloids in significant quantities may be used as anti malaria, analgesics and stimulants [22].

The other phytochemicals present in the plant are known to inhibit tumour growth, treatment of intestinal disorder like diarrhea and dysentery, Tannins are used in treating wounds, sprains, bruises and arresting bleeding [6].

In general, the levels of medicinal plants and their compounds necessary to inhibit microbial growth are higher in foods than in culture media. This is due to interactions between phenolic compounds and the food matrix [38,30] and should be considered for commercial applications.

The plant extracts presented positive antimicrobial activity for *Escherichia coli, Pseudomonas aeruginosa* [29] *Staphylococcus aureus*, *Klebsiella pneumonia, Bacillus cereus, Proteus vulgaris, Streptococcus viridians, Shigella dysenteriae, Bacillus subtilis* and *Salmonella typhi* [29,8,1,2]. The ethanolic extracts of *Daniella oliveri* are very potent in terms of activity because of the presence of bioactive components in large quantity [43].

Several phenolic compounds like tannins, flavonoids present in the cells of plants are potent inhibitors of many hydrolytic enzymes such as proteolytic macerating enzymes used by plant organisms. Other preformed compounds like Saponins also have antifungal properties. Many plants contain non-toxic glycosides that can get hydrolyzed to release Phenolics that are toxic to microbial pathogens [43]. Therefore, the compounds detected may be responsible for the antibacterial activity according to [44].

The proximate analysis showed that the plant extracts were rich in carbohydrate which the basic source of energy. it is always very difficult to survive without the energy, and this is produce in abundant in carbohydrate. it is also an empirical believe of traditional medicine provided that daily consumption of the medicinal plant like *Daniella oliveri* can lead to life longevity [45].

Table 8. Quantitative analyses of proximate composition of *Daniella oliveri* extracts in percentage

Plants	Ash	MC	CP	Fat	Fibre	CHO
BARK *Daniella oliveri*	10.56	9.12	14.45	6.59	10.33	42.59
LEAF *Daniella oliveri*	10.21	8.24	13.34	7.21	11.32	45.24

KEY: ASH- ash content; MC-mineral content; CP- crude protein; FAT- fat content; FIBRE- fibre content; CHO- carbohydrate content

The Mineral elements composition of *Daniela olivera* in mg/100 g indicated that leaves and bark contained vital micro-nutrient such as Potassium, Calcium, Magnesium, and Iron while Lead show minimal occurrence. Antimicrobial inhibition of bacteria by plant extracts had been extensively reported [46]. This confirms the report of [18,47] that the active principles of plant drugs are more concentrated in storage organs such as leaves, roots, seeds, bark while flowers and woody parts of herbaceous stem are relatively inert.

Similarly various other compound present in several plants showed such antimicrobial activity against disease causing organisms like *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus*, *Klebsiella pneumonia, Bacillus cereus, Proteus vulgaris, Streptococcus viridians, Shigella dysenteriae, Bacillus subtilis* and *Salmonella typhi* [45].

Providing effective health care is a challenge under the best of economic circumstances. In the world's poorest countries, where infectious diseases are rife and resources limited, that challenge can assume over-whelming proportions, hence the resurgence in the use of herbal preparations to treat diseases.

5. CONCLUSION

After the experiment, it was concluded that *Daniella oliveri* plant can be effective in the treatment of Dysentery, typhoid fever, headache and series of infections caused by microorganisms. This can be used in the production of more antibiotics in Nigeria and Worldwide to avoid and combat resistance of microorganisms to antibiotics. It also shows that the use of local herbs or medicinal plant should be encouraged by Government agencies and individuals worldwide.

6. RECOMMENDATION

It is thereby recommended that crude extract of *Daniella oliveri* which is a medicinal plant is safe for the treatment of clinically cause infection like diarrhea and dysentery and should be encourage for use.

ETHICAL APPROVAL

It is not applicable.

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COMPETING INTERESTS

Authors have declared that no competing interests exist

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