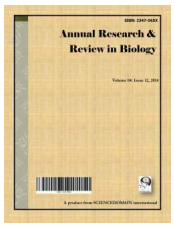

Annual Research & Review in Biology
8(3): 1-10, 2015, Article no.ARRB.14947
ISSN: 2347-565X, NLM ID: 101632869

SCIENCEDOMAIN *international*
www.sciencedomain.org

Antimicrobial Activities of *Phragmanthera incana* (schum.) Balle, a Mistletoe Species Harvested from Two Host Plants against Selected Pathogenic Microbes

O. T. Ogunmefun[1], E. A. Ekundayo[1*], T. A. Ogunnusi[1], A. H. Olowoyeye[1], T. R. Fasola[2] and A. B. Saba[3]

[1]Department of Biological Sciences, Afe Babalola University, Ado-Ekiti, Nigeria.
[2]Department of Botany, University of Ibadan, Ibadan, Nigeria.
[3]Department of Physiology, Biochemistry and Pharmacology, Faculty of Veterinary Medicine, University of Ibadan, Ibadan, Nigeria.

Authors' contributions

This work was carried out in collaboration between all authors. Author OTO designed the study and carried out the supervision of the work. Authors EAE and TAO supervised and guided the work design. Author AHO carried out the bench work. All authors read through the manuscript at the end of the write up.

Article Information

DOI: 10.9734/ARRB/2015/14947
Editor(s):
(1) Yukun Liu, Key Laboratory for Forest Resources Conservation and Utilization Southwest Mountains of China, Southwest Forestry University, China.
(2) George Perry, Dean and Professor of Biology, University of Texas at San Antonio, USA.
Reviewers:
(1) Anonymous, University of Copenhagen, Denmark.
(2) Anonymous, University of Nigeria, Nigeria.
(3) Anonymous, UCSI University, Malaysia.
(4) Anonymous, Selcuk University, Turkey.
(5) Anonymous, Regional Center of Agricultural Research, Tunisia.
Complete Peer review History: http://sciencedomain.org/review-history/11317

Original Research Article

Received 29th October 2014
Accepted 18th June 2015
Published 7th September 2015

ABSTRACT

Aims: *Phragmanthera incana*, African mistletoe popularly called "bird lime", devil's fuge, "all heal, Iscador", "mystyldene", "golden bough" etc a hemi-parasitic plant was screened for its antimicrobial properties due to its ethnomedicinal claims as a remedy for stomach disorder, diarrhoea, dysentery, wound and other infections.

**Corresponding author: E-mail: esttydayo2010@yahoo.co.uk;*

Methodology: The antimicrobial activities of aqueous and methanol extracts of *P. incana* obtained from cocoa (*Theobroma cacao*) and kolanut (*Cola nitida*) were tested *In vitro* against five Gram negative pathogenic bacteria; *Escherichia coli, Aeromonas popoffi, Proteus mirabilis, Klebsiella pneumoniae*, *Tatumella ptyseos* and five Gram positive bacteria; *Bacillus cereus, B. firmus, Paenibacillus assamensis, P. apiarius, Corynebacterium accolens;* and seven pathogenic fungi; *Candida albicans*, *Aspergillus niger, A. flavus, Sclerotium rolfsii*, *Dinemasporium* species, *Mycotypha microspora* and *Harposporium* species using agar diffusion method and food poisoning techniques. The antimicrobial activity was assessed by the presence or absence of inhibition zones, minimum inhibitory concentration (MIC), minimum bactericidal concentration (MBC) and minimum fungicidal concentration (MFC) values.
Results: *Phragmanthera incana* kolanut methanol extract showed a higher zone of inhibition for both bacterial and fungal isolates used than *P. incana* cocoa methanol extract which could be due to the phytochemical constituents. Phytochemical investigation of the mistletoe from cocoa and kolanut indicated the presence of alkaloid, phenolics, flavonoids, tannins, and cardiac glycosides. *Phragmanthera incana* aqueous extract from both plants (cocoa and kolanut) showed no antimicrobial activities towards the organisms used except for the food poisoning techniques for antifungal assay.
Conclusion: This study showed a moderate antimicrobial potential of the extracts of the mistletoe, *P. incana. Phragmanthera incana* growing on kolanut was found to be more effective than *P. incana* growing on cocoa.

Keywords: Antimicrobial activity; cola nitida; mistletoe (Phragmanthera incana); methanol; Theobroma cacao.

1. INTRODUCTION

Researches into herbal drugs are on-going and increasing on daily basis due to the need for safer and affordable drugs which subsequently lead to the formulation of new herbal products for preventive or curative purposes [1]. The use of herbal medicine as alternative therapy has become prevalent throughout the world due to the growing resistance of pathogens to conventional antibiotics [2]. Medicinal plants and their products are in an increasing demand as alternative therapy to orthodox medicines most especially in developing countries where resources are meagre [3]. Medicinal plants are frequently used as remedies for many infectious diseases [4]. It's interesting to note that the search for new antimicrobials from the medicinal plants is still increasing with the emergence of antibiotic resistance development in the pathogens [5].

The mistletoe plant is commonly referred to as "Afomo onisana" in the South-western part of Nigeria (Yoruba Land) because the flowers resemble a match stick, sometimes yellow-flowered with red or purple corolla tips such as in *P. capitata* or purple flowered with yellow corolla tips as in some other mistletoe.

The mistletoe plant is an obligate parasite that depends partly on its host to obtain water and minerals but can carry out photosynthesis [1]. It is an evergreen hemi-parasite that can grow in most parts of the globe. It has different families and species that are well known worldwide. They grow primarily on deciduous trees such as oak, elm, fir, pine and apple.

They are widely distributed throughout Africa, Europe, North America, and Asia. In the South-western Nigeria, mistletoe is commonly found growing especially on tree crops like *Cola nitida* (Vent.) Schott & Endl. or *Cola acuminata* (*P. Beauv*.) Schott & Endl. (kola), *Theobroma cacao* L. (cocoa), *Irvingia gabonensis* (Aubrey-Lecomte ex O. Rorke) Baill. (bush mango) and *Coffea arabica* L. (coffee). Orange (*Citrus sp*.) and guava (*Psidium guajava* L.) can also serve as host trees to mistletoe.

They are highly specialized angiosperms of the *Loranthaceae* and *Viscaceae* family. They cause important damages to their hosts with great economic loss [6]. They consist of about 75 genera and 1,000 species of woody plants, many of them are hemi parasites, all of them except three having the mistletoe habit [7,8].

A decoction of the leaves of mistletoe is traditionally used in the treatment of hypertension to alleviate symptoms of such as headache, dizziness, palpitation, etc. For centuries, mistletoe also served as a folk medicine for cancer treatment and the plant has sometimes been used in Europe to treat tumors [9] and as a

digestive aid, heart tonic and to treat arthritis, amenorrhea, wounds, asthma, bed wetting, infection, hysteria and other mental disturbances [6].

Mistletoe is a diuretic and depurative, that is, urine production is increased and metabolic toxic wastes such as uric acid are eliminated. It is used whenever blood is to be purified. Mistletoe is employed to correct menstrual disorder, excessive menstruation and uterine hemorrhages. It is an excellent and effective remedy for epilepsy [10].

Plant materials continue to play a major role in primary health care as therapeutic remedies in many developing countries [11]. The control of bacterial infection has been remarkably effective since the discovery of antibacterial drugs [12]. However, some of the pathogens rapidly become resistant to many of the first discovered effective drugs [12]. The search for new antibacterial agents in particular from medicinal plants has been warranted due to the development of drug resistance and undesirable side effects of certain antibiotics [13]. The screening of plant extracts has been of great interest to the researchers for the discovery of new drugs effective in the treatment of several diseases.

Phragmanthera incana is a woody parasitic shrub, stems to 2m long; of secondary jungle and bush savanna areas; from Sierra Leone to West Cameroons and Fernando Po Island (in the gulf of Guinea that forms part of Equatorial Guinea), and extending across the Congo basin to Zaire and Angola [14]. Young parts and perianth more or less densely covered with brown hairs; berries red. The plant is very variable in the shape and size of the flowers and leaves. The plant is very variable in form, common and widely distributed [15]. *P. incana* can be found on *Anacardium occidentale, Alchornea castaneifolia*, *Bauhinia monandra, Bombax sessile, Aleurites molluccana* etc [16].

Information on *Phragmathera incana* is very rare and since plants that are apt to produce mutagens could perhaps produce antibiotics, therefore it is imperative to investigate *Phragmanthera incana*.

2. MATERIALS AND METHODS

2.1 Collection of Plant Samples

Phragmanthera incana harvested from cocoa (*Theobroma cacao*) and kolanut (*Cola nitida*) trees were collected from an area about 8km away from Owo, Ondo State, Nigeria. Identification and authentication was done at the Forestry Research Institute of Nigeria (FRIN) herbarium while a voucher specimen of *Phragmanthera incana* with Forestry Herbarium Index (FHI) 108925 and University of Ibadan herbarium (UIH) number 22332 was submitted at the Botany Department herbarium of the University of Ibadan.

2.2 Collection of Test Organisms

Five Gram positive bacteria; *Bacillus cereus, B. firmus, Corynebacterium accolens, Paenibacillus assamensis, P. apiaries* and five Gram negative bacteria; *Aeromonas popoffi, Klebsiella pneumoniae, Proteus mirabilis, Escherichia coli, Tatumella ptyseos*. Seven fungi; *Aspergillus flavus, A. niger, Sclerotium rolfsii, Mycotypha microspora, Candida albicans, Dinemasporium* species *and Harposporium* species were collected from the Department of Biological Sciences (Microbiology), Afe Babalola University, Ado-Ekiti. Biochemical tests were done for confirmation of microorganisms based on microscopic appearance and characterization was carried out as described by [17]. The isolates were then stored on slants at 4℃ and on plates at 25℃ for bacteria and fungi respectively.

2.3 Extraction of Plant Sample

Phragmanthera incana leaves harvested from kolanut (430 g) and cocoa (400 g) were air-dried and extracted with methanol employing the method of cold maceration for 72 hours. The solvent was distilled over using Rotary Evaporator (model number RE300DB) at a low temperature of about 45℃ and further concentrated on water bath at a low temperature of 40℃. Other portions of *P. incana* leaves from kolanut (400 g) and cocoa (380 g) were soaked in clean water and left at room temperature for 72 hours using the method of cold maceration [18,19]. The extracts were decanted and concentrated on water bath at a low temperature of 45℃ to give the crude extracts used for the antimicrobial investigation.

2.4 Phytochemical Analysis of *P. incana*

The extracts were subjected to both qualitative and quantitative phytochemical tests for plant secondary metabolites namely; alkaloids, flavonoids, tannins, saponins, cardiac glycosides

and phenolics according to the method described by Harborne [20] and Trease and Evans [21].

2.5 Antibacterial Activities of Aqueous and Methanol Extracts of *P. incana*

An overnight culture of each bacterial isolate was prepared by taking two wire loops of the organism from the stock and inoculating each into 5 mL sterile nutrient broth and incubated for 24 hours at 37℃. From overnight culture, 0.1 ml of each organism was taken and put into the 9.9 ml of sterile distilled water to get (1:100) of the dilution of the organism. An aliquot (0.1 ml) was taken from the dilution into a sterile Petri dish, prepared Mueller Hinton agar was then poured into the plate, swirled and allowed to solidify. A cork borer (6 mm) that was already sterilized was used to create wells inside the sterile agar plates. The crude extracts were reconstituted with dimethyl sulphoxide (DMSO) and introduced into the well. The DMSO served as the negative control and was introduced into a separate well while standard antibiotic disc served as positive control. The plates were made to remain on the bench for 2hours before incubation at 37℃ for 24 hours. The tests were conducted in triplicates. The antimicrobial activity was determined by measurement of zone of inhibition around each well.

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

Minimum inhibitory concentration (MIC) for each of the plant extract showing antimicrobial activity against the test isolates was determined using broth micro dilution method [22]. The lowest concentration of the extracts in the well of the test tube that showed no turbidity after incubation was taken as the MIC values. The visible growth of microorganisms was taken by the turbidity of the wells in the test tube. Sub culturing from each well showing no apparent growth was used to determine the minimum bactericidal concentration (MBC). MBC was determined as the least concentration of the extract showing no visible growth on sub culturing [23,24].

2.7 Antifungal Activities of Aqueous and Methanol Extracts of *P. incana*

Two techniques were used to determine the antifungal activities of *P. incana* viz agar well diffusion and food poisoning methods. In agar well diffusion method, aliquots of spore were prepared by mixing loopful of fungal spores in sterile distilled water. Spore suspension (0.1 ml) was put into a sterile Petri dish, prepared Potato Dextrose agar was then poured into the plate, swirled and allowed to solidify. A cork borer (6 mm) already sterilized was used to create wells inside the sterile agar plates. The crude extracts were reconstituted with Dimethyl sulphoxide (DMSO) and introduced into the well. The DMSO served as the negative control and was introduced into a separate well as appropriate. The plates were left on the bench for 2hours before incubation at 27℃ for 48 hours. The tests were conducted in triplicates. The antifungal activity was determined by measurement of zone of inhibition around each well. In food poisoning technique, sterile potato dextrose agar was mixed with crude aqueous extracts of the test plants after cooling. Quantity measuring about 15 ml of the medium was poured into each of the petriplates and allowed to solidify. 7-day-old culture of the test fungi (6 mm disc) were placed at the centre of the petriplates and incubated at a temperature of about 25±2℃ for a period of seven days. The colony diameter was measured in millimetre after incubation. Two replicates were maintained for each treatment and potato dextrose agar medium having no aqueous extract was used as control. The same procedure was repeated for methanolic extracts of *P. incana*. The effects of the extracts in inhibiting fungi growth was measured in terms of mycelial growth inhibition (%) i.e. fungitoxicity of the extracts was calculated by using: the formula:

$$\% \text{ inhibition} = dc - dt/dc \times 100$$

Where

dc = Average increase in mycelial growth in control,

dt = Average increase in mycelial growth in treatment [25]

2.8 Determination of Minimum Fungicidal Concentration (MFC)

The fungicidal activity was determined for fungi that exhibited clearance in the minimum inhibitory concentration. After 72 h of incubation, 1 ml was subcultured from the cleared test tubes into PDA plates and was incubated at 27℃ for 48h. The MFC was the lowest concentration that showed either no growth or less growth [26].

3. RESULTS

The results of both qualitative and quantitative phytochemical analyses represented in Table 1

and Table 2 show the presence of medicinally active constituents in *P. incana*. Both *P. incana* from cocoa (A) and *P. incana* from kolanut (B) had tannins, alkaloids, flavonoids, cardiac glycosides and phenols but anthraquinones and saponins were absent for the qualitative analysis. The quantitative estimation of the chemical constituents of the plant studied showed that the leaf of *P. incana* obtained from two different host trees *T. cacao* (cocoa) and *C. acuminate* (kolanut) was rich to some extent in alkaloid, flavonoids, cardiac glycosides, tannin, and phenolic compound. *Phragmanthera incana* from cocoa was very rich in alkaloid, tannins, saponins and anthraquinones than *P. incana* obtained from kolanut. Likewise, *P. incana* from kolanut also had high content of flavonoids, cardiac glycosides and phenolic than cocoa.

Table 1. Qualitative phytochemical properties of *P. incana* growing on cocoa and kolanut

Parameters (mg/100g)	A	B
Alkaloids	+	+
Flavonoids	+	+
Tannins	+	+
Saponins	-	-
Cardiac glycosides	+	+
Phenolics	+	+
Anthraquinones	-	-

Keys: A = Phragmanthera incana harvested from cocoa, B =Phragmanthera incana harvested from kolanut

Table 3 shows the antibacterial activities of the leaf extract from two different hosts. The methanol extract exhibited antibacterial activities while the aqueous extract had no effect on the test organisms. Generally, *Bacillus cereus* was the most inhibited by *P. incana* from cocoa and kolanut with zones of inhibition of 21.00 mm and 14.00 mm respectively. *Bacillus cereus* was most susceptible to *P. incana* harvested from kolanut tree with zones of inhibition of 21.00±3.00 followed by *Paenibacillus assamensis, Aeromonas popoffi* while *Proteus mirabilis* was least sensitive to *P. incana* harvested from cocoa tree zones of inhibition of 10.00±2.00. However, *Bacillus firmus, Corynebacterium accolens, Paenibacillus apiaries, Klebsiella pneumoniae, Escherichia coli* and *Tatumella ptyseos* were not sensitive to *P. incana* harvested from both kolanut and cocoa trees.

Figs. 1 and 2 expressed the antibiotic susceptibility of test organisms. It was observed that all the Gram positive organisms were resistant to Cotrimoxazole, Cloxacillin and Augmentin while *Corynebacterium accolens* was resistant to Erythromycin (5 µg), Gentamicin (10 µg), Augmentin (30 µg), Streptomycin (10 µg), Tetracycline (10 µg) and Chloramphenicol (10 µg), Cloxacillin 5 µg, and Cotrimoxazole 25 µg. Generally, all the tested Gram negative bacteria were most susceptible to Ofloxacin but resistant to Augmetin and Amoxycillin. The result also shows *Proteus mirabilis* was most susceptible to Ofloxacin with zone of inhibition of 26.0 mm.

Table 4 shows the minimum inhibitory concentration (MIC) where it was also observed that at higher concentration there was a stronger activity but there no activity at lower concentration against the test bacteria. The minimum inhibitory concentration of the plant extracts were evaluated between 25–200 mg/ml. The result shows that MIC on PKME for *Bacillus cereus, Proteus mirabilis* and *Aeromonas popoffi* was 100 m/ml while that of *Paenibacillus assamensis* was 200 mg/ml. Similar MIC value was obtained for *B. cereus* on PCME while the MIC of 200 mg/ml was obtained for *Proteus mirabilis* and *Aeromonas popoffi*. The minimum bactericidal concentration (MBC) of the extracts was evaluated between 25- 200 mg/ml as shown in Table 5. The MBC was 200 mg/ml for *Bacillus cereus and Proteus mirabilis* on PKME and PCME respectively while it was 100 mg/ml on PKME and 200 mg/ml on PCME for *Aeromonas popoffi*.

Table 2. Quantitative phytochemical properties of *P. incana* growing on cocoa and kolanut

Parameters	A	B
Alkaloids (mg/100 g)	1745.00±15.00	1545.00±25.00
Flavonoids (mg/100 g)	1265.00±15.00	1458.30±30.00
Tannins (mg/100 g)	1516.60±18.30	1455.00±40.00
Saponins (mg/100 g)	56.70±25.00	23.30±5.00
Cardiac Glycosides (mg/100 g)	373.30±20.00	513.30±2.00
Phenolics (GAE/g)	54.00±2.00	73.50±1.30
Anthraquinones (g/100 g)	45.00±5.00	23.30±1.60

Keys: A = Phragmanthera incana from cocoa, B = Phragmanthera incana from kolanut

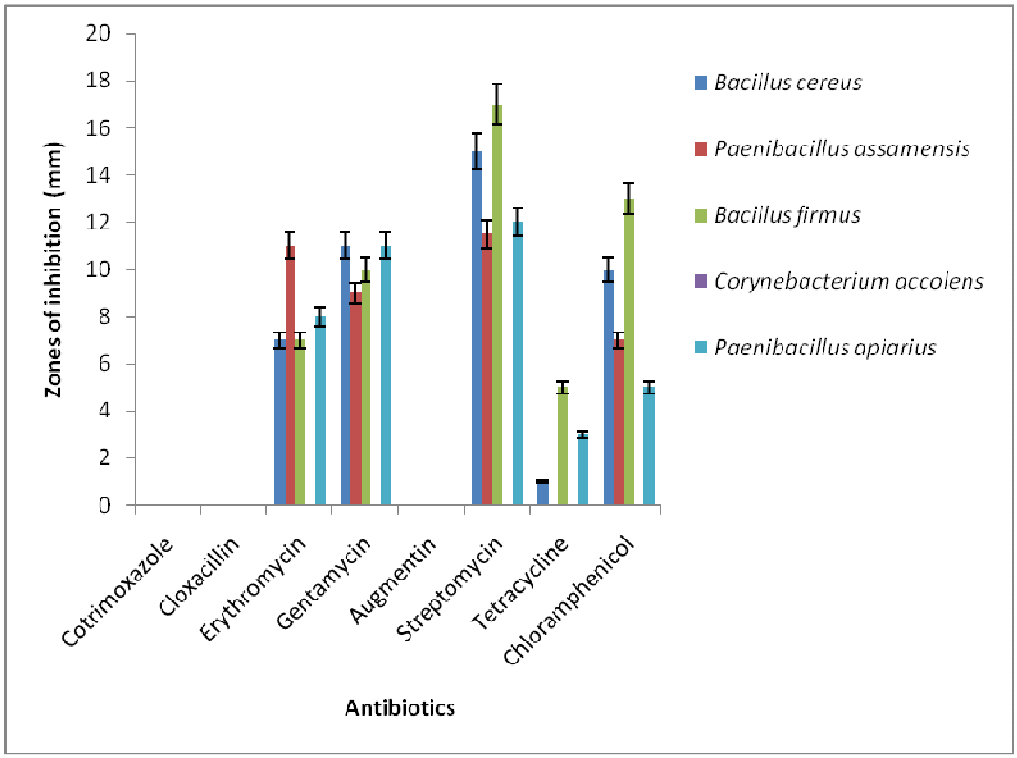

Fig. 1. Antibiotic susceptibility patterns of selected Gram positive bacteria

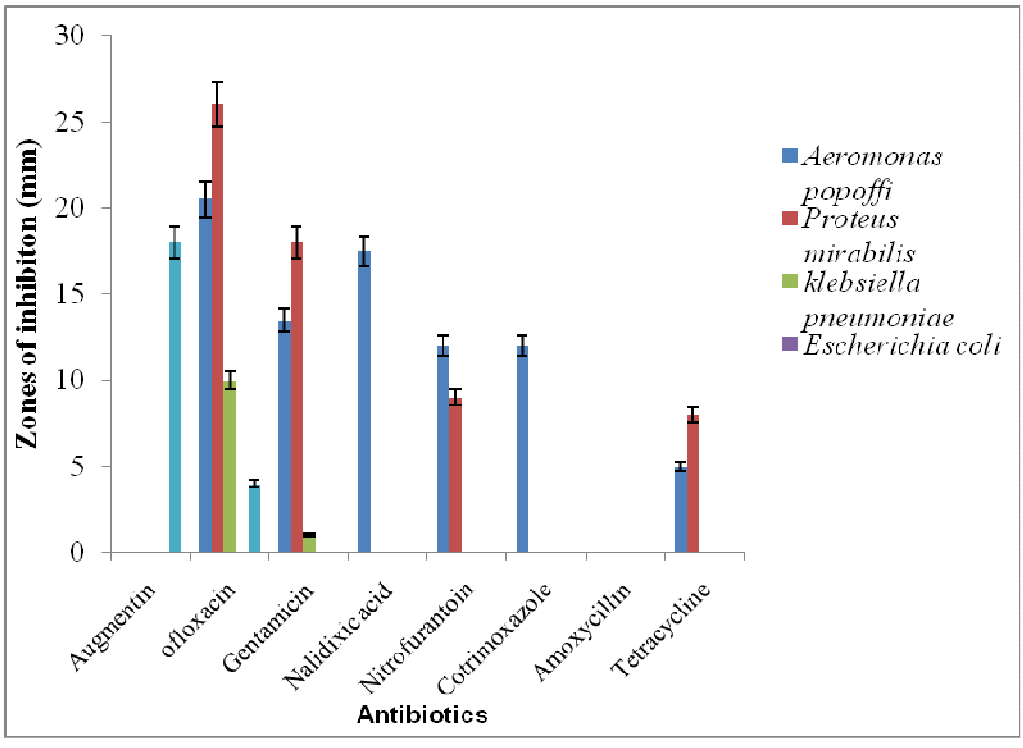

Fig. 2. Antibiotic susceptibility patterns of selected Gram negative bacteria

Table 3. Antibacterial activity exhibited by extracts of *Phragmanthera incana* growing on cocoa and kolanut

Selected bacteria	Zone of inhibition (mm)			
	PKME	PKAE	PCME	PCAE
Bacillus cereus	21.00±3.00	-	14.00±0.00	-
Bacillus firmus	-	-	-	-
Corynebacterium accolens	-	-	-	-
Paenibacillus assamensis	19.90±4.50	-	11.20±0.50	-
Paenibacillus apiarius	-	-	-	-
Aeromonas popoffi	19.40±1.50	-	11.50±1.00	-
Klebsiella pneumoniae	-	-	-	-
Proteus mirabilis	14.00±4.00	-	10.00±2.00	-
Escherichia coli	-	-	-	-
Tatumella ptyseos	-	-	-	-

Keys: PKME = P. incana kolanut methanol extract, PKAE = P. incana kolanut aqueous extract, PCME = P. incana cocoa methanol extract and PCAE = P. incana cocoa aqueous extract

Table 4. Determination of minimum inhibitions concentration (MIC) of the extracts

Organisms	PKME (mg/ml)	PCME (mg/ml)
Bacillus cereus	100	100
Paenibacillus assamensis	200	-
Aeromonas popoffi	100	200
Proteus mirabilis	100	200

Key ~ KME = P. incana kolanut methanol extract and PCME = P. incana cocoa methanol extract

Table 5. Minimum bactericidal concentration (MBC) of *P. incana* extract determination

Organisms	PKME (mg/ml)	PCME (mg/ml)
Bacillus cereus	200	200
Aeromonas popoffi	100	200
Paenibacillus assamensis	200	-
Proteus mirabilis	200	200

Keys: PKME= P. incana Kolanut methanol extract and PCME= P. incana Cocoa methanol extract

It was observed that PKAE and PCAE were not inhibitory to the test fungi. Only *C. albicans* was susceptible to PKME and PCME with zones of inhibition of 32.00±4.00 and 30.00±2.00 respectively using agar well diffusion method (Table 6). However, *Sclerotium* spp, *Dinemasporium* spp, *Harposporium* spp were susceptible to the extracts in varying degrees while *A. flavus*, *A. niger*, *Mycotypha microspora* and *C. albicans* were resistant to all the extracts using food poisoning technique (Table 7). The minimum inhibitory concentration on *C. albicans* was 50 mg/ml and 100 mg/ml for *P. incana* kolanut methanol and *P. incana* cocoa methanol respectively. The minimum fungicidal concentration (MFC) of the plant extracts was evaluated between 100 and 200 mg/ml. The MFC value of *P. incana* kolanut methanol extract on *C. albicans* was 100 mg/ml while *P. incana* cocoa methanol extract was 200 mg/ml.

4. DISCUSSION

Many literatures have reported Multiple Drug Resistance (MDR) development due to the indiscriminate use of commercial antimicrobial drugs commonly used in the treatment of diseases caused by pathogenic organisms. Adding to this problem, antibiotics are sometimes associated with adverse effects on the host, including hypersensitivity, immune suppression and allergic response [27]. Measures to overcome such situations has led to this study of antimicrobial potency of *P. incana* for possible clue as an alternative to replacing some antibiotics known for their side effects and inconsistent effects in the purposes they were manufactured. Plants serve as raw materials for useful structures that have great potentials in the synthesis of new chemotherapeutic agents. To achieve this development of new antimicrobial drugs was found in the *In vitro* antibacterial activity assay as reported by [28]. ''Many reports are available on the antiviral, antibacterial, antifungal, anthelmintic, anti-mollusca and anti-inflammatory properties of plants'' [29,30]. The reports of some of these assays have led to the isolation and characterization of the active chemical compounds (principle) that confer on the plants its potentials as antimicrobial and other therapeutic agents leading to the development of new drugs for administration in humans. Crop protection however, is yet to find popularity in utilising antifungal or antibacterial property of plants for the development of new drug formulations for applications commercially.

Table 6. Antifungal activities exhibited by extracts of *Phragmanthera incana* leaves harvested from cocoa and kolanut

Selected fungi	Zone of inhibition (mm)			
	PKME	PKAE	PCME	PCAE
Candida albicans	32.00±4.00	-	30.00±2.00	-
Aspergillus niger	-	-	-	-
Aspergillus flavus	-	-	-	-
Sclerotium species	-	-	-	-
Dinemasporium spp	-	-	-	-
Harposporium spp	-	-	-	-
Mycotypha microspora	-	-	-	-

Keys: PKME = P. incana kolanut methanol extract, PKAE = P. incana kolanut aqueous extract, PCME = P. incana cocoa methanol extract and PCAE = P. incana cocoa aqueous extract

Table 7. Antifungal activities exhibited by extracts of leave of *Phragmanthera incana* growing on cocoa and kolanut using the food poisoning technique

Plant extracts	*Aspergillus niger*	*A. flavus*	*Sclerotium* sp	*Candida albicans*	*Dinemasporium* spp	*Harposporium* spp	*Mycotypha microspora*
PKAE	-	-	12.50±2.00	-	-	12.10±1.00	-
PKME	-	-	15.20±0.50	-	12.50±6.00	16.10±2.00	-
PCME	-	-	11.60±1.50	-	15.80±0.10	-	-
PCAE	-	-	11.30±1.00	-	-	13.40±3.00	-

Keys: PKME = P. incana kolanut methanol extract, PKAE = P. incana kolanut aqueous extract, PCME = P. incana cocoa methanol extract and PCAE = P. incana cocoa aqueous extract

The phytochemical screening of *Phragmanthera incana* leaves exhibited the presence of phytochemicals such as alkaloids, tannins, flavonoids, phenols and cardiac glycosides which are similar to the findings of [31]. The aqueous extract was found to be least effective in exhibiting antimicrobial effects on the test bacteria in the present study. This can be corroborated by Koduru et al. [32] and Nyembo et al. [33] also who stated that aqueous extracts always show little or no antibacterial activity and this may be connected with the fact that water is least effective in extracting the active antimicrobial component(s) present in the leaves of *P. incana*. However, the aqueous extract showed antifungal activity against some of the test fungi and this could be ascribed to the anionic components such as thiocyanate, nitrate, chlorides and sulphate apart from other water soluble constituents that are present naturally in the plant material [34].

Phragmanthera incana kolanut methanol extract showed a higher inhibition than *Phragmanthera incana* cocoa methanol extract which could be due to the phytochemical constituents which are known to be biologically active and exert antimicrobial properties through different mechanisms [35]. The inhibitory activity of plant extract is also largely dependent on the concentration, parts of the plant used and the microbes tested [36]. The differences in the sensitivity of pathogenic microorganisms may be due to the differences in concentrations, methods of extraction used in each study [37] and the little diffusion properties of these extracts in the agar and soil composition and water availability [38].

5. CONCLUSION

The present study showed a moderate antimicrobial potential of the extracts of the mistletoe, *Phragmanthera incana. P. incana* growing on kolanut was better in effectiveness than *P. incana* growing on cocoa. It may be concluded from the present studies that both extracts can be used as a potential source of natural antimicrobial compound. Further research is required for the identification of bioactive molecule present in the two extracts.

COMPETING INTERESTS

Authors declare that no competing interests exist.

REFERENCES

1. Griggs P. Mistletoe, myth, magic and medicine. The Biochemistry. 1991;13:3-4.
2. De Smet PAG. Herbal remedies, New England. Journal. Medicine. 2002;347: 2046-2056.

3. Murray NB. African indigenous medicinal research functions, Nairobi. 2008;37-38.
4. Ahmad I, Mehmood Z, Mohammad F. Screening of some Indian medicinal plants for their antimicrobial properties. Journal. Ethnopharmacol. 1998;62:183-193.
5. Fyhrquist P, Mwasumbi L, Haeggstrom, C.A, Vuorela H, Hiltunen R, Vuorela P. Ethnobotanical and antimicrobial investigation on some species of Terminalia and Combretum (Combretaceae) growing in Tanzania. Journal Ethnopharmacology. 2002;79:169-177.
6. Boussim IJ, Guinko S, Tuquet C, Salle G. Mistletoes of the agro forestry Parklands of Burkina Faso. Agro forestry System. 2004; 60:39-49.
7. Watson DM, Herring M. Mistletoe as a keystone resource: An experimental test. Proceedings of the Royal Society B: Biological Sciences. 2012;279(1743):3853-3860.
DOI:10.1098/rspb.2012.0856
8. Efuntoye MO, Ayodele AE, Thomas BT, Ajayi TO. Does host plant affect the antibacterial activity of *Tapinanthus bangwensis* Danser (Loranthaceae)? Journal Medicinal Plants Research. 2010; 4(13):1281-1284.
9. Deeni YY, Sadiq NM. Antimicrobial properties and phytochemical constituentsof the leaves of African mistletoe (*Tapinanthus dodoneifolius* (DC) Danser) (Loranthaceae): An etnomedicinal plant of Hausaland, Northern Nigeria, Journal of Ethnopharmacology. 2002;83: 235-240.
10. Polhill R, Wiens D. Mistletoes of Africa. The Royal Botanic Garden, Kew, U.K. 1998; 370.
11. Dimayuga RE, Garcia SK. Antimicrobial screening of medicinal plants from Baja California Sur. Mexico. Journal of Ethnopharmacology. 1991;181-192.
12. Ríos JL, Recio MC. Medicinal plants and antimicrobial activity. Journal of Ethnopharmacology. 2005;100:80-84.
13. WHO. Traditional Medicine Strategy (2002-2005) WHO Publications. 1-6. Zakaria M. Isolation and characterization of active compounds from medicinal plants. Asia Pacific Journal of Pharmacology. 2002;6: 15-20.
14. Bright EO, Okusanya BA. Infestation of economic plants in Badeggi by *Tapinanthus dodoneifolius* (DC) Danser and *T. globiferus* (A. Rich) Van Tiegh. Niger. Journal. Weed Science. 1998;11:51-56.
15. Burkill HM. The useful plants of West Tropical Africa. 1985;3.
16. Flora of West Tropical Africa (F.W.T.A.) ed. 1. 466, *Phragmanthera incana* in Kew Bull 1956;168.
17. Cowan MM, Steel. Manual for the identification of medical bacterial. Cambridge University Press, London. 1991; 331.
18. Ruttoh EK, Bil C, Tarus PK, Machocho A, Karimi LK, Okemo P. Antifungal activity of *Tabernaemontana stapfiana* Britten (Apocynaceae) organic extracts. Pharmacognosy Research. 2009;1(6):387-391.
19. Ogbole OO, Adeniji JA, Ajaiyeoba EO, Adu DF. Anti-polio virus activity of medicinal plants selected from the Nigerian ethno-medicine. Academic Journals. 2013; 12(24):3878-3883.
20. Harborne JB. Phytochemical methods - A guide to modern techniques of plant analysis. Chapman and Hall, London, 1998;182- 190.
21. Trease GE, Evans WC. Phamacognosy. London: Baillvera Tindall. 2004;13:309–707.
22. Basri DF, Fan SH. The potential of aqueous and acetone extracts of galls of *Quercus infectoria* as antibacterial agents. Indian Journal Pharmacology. 2005; 37(1):26-29.
23. Vollokova AD, Kostalova, Sochorova. Isoquinoline alkaloid from mahonia aquifolium stem bark is active against Malseizia species. Journal of Microbiology. 2001;46:107–111.
24. Ogbulie JN, Ogueke CC, Okoli IO, Anyawu BN. Antibacterial activities and toxicological potencial of crude ethanolic extracts of *Euphorbia hirta*. African Journal of Biotechnology. 2007;6(13):1544–1548.
25. Akinpelu DA, Kolawole DO. Phytochemicals and antimicrobial activity of leaf extract of *Piiostigma thoriningi* (Schum). Science Focus. 2004;4:64-70.
26. Singh J, Tripathi NN. Inhibition of storage fungi of blackgram (*Vigna mungo*) by some essential oils. Flavour and Fragrance Journal. 1991;14:1-4.
27. Dey SK, Banerjee D, Chattapadhyay S. Karmakar KB. Antimicrobial activities of some medicinal plants of West Bengal. Int. J. Pharma Bio. Sci. 2010;1:1-10.

28. Tona L, Kambu K, Ngimbi N, Cimanga K. Antiamoebic and phytochemical screening of some Congolese medicinal plants. Journal of Ethnopharmacol. 2003;61:57-65.
29. Samy RP, Ignacimuthu S. Antibacterial activity of some folklore medicinal plants used by tribals in Western Ghats in India. Journal Ethnopharmacology. 2000;69:63-71.
30. Govindarajan R, Vijayakumar M, Singh M, Rao CHV, Shirwaikar A, Rawat AKS, Pushpangadan P. Antiulcer and antimicrobial activity of *Anogeissus latifolia.* Journal Ethnopharmacology. 2009;106:57-61.
31. Ogunmefun OT, Fasola TR, Saba AB, Oridupa OA. The ethnobotanical, phytochemical and mineral analyses of *Phragmanthera Incana* (Klotzsch), A species of mistletoe growing on three plant hosts in South-Western Nigeria. Int J. Biomed Sci. 2013;9(1):33-40.
32. Koduru S, Grierson DS, Afolayan AJ. Antimicrobial activity of *Solanum aculeastrum* (Solanaceae). Pharmacol. Biol. 2006;44:284-286.
33. Nyembo K, Kikakedimau N, Mutambel H, Mbaya N, Ekalakala T, Bulubulu O. *In vitro* antibacterial activity and phytochemical screening of crude extracts from *Jatropha curcas* Linn. Eur. J. Med. Plants. 2012; 2(3):242-251.
34. Darout I, Cristy A, Skaug N, Egeberg P. Identification and quantification of some potential antimicrobial anionic components in miswak extract. Industrial Journal Pharmacology: 2000;32:11-14.
35. Ekundayo FO, Adeboye CA, Ekundayo EA. Antimicrobial activities and phytochemical screening of pignut (*J. curcas* Linn.). J. Med. Plants Res. 2011;5: 1261-1267.
36. Kalimuthu K, Vijayakumar S, Senthilkumar R. Antimicrobial activity of the biodiesel plant, *Jatropha curcas*. Intern. J. Pharm. Bio. Sci. 2010;1:1-5.
37. Kumar S, Bahchi GD, Darokar MP. Antibacterial activity observed in the seeds of some coprophilus plants. International Journal Pharmacology. 1997;35:179-184.
38. Romero CD, Chopin SF, Buck G, Martinez E, Garcia M, Garcia I. Antimicrobial properties of common herbal remedies of the southwest. Journal of Ethnopharmacology. 2005;99:253-257.

Peer-review history:
The peer review history for this paper can be accessed here:
http://sciencedomain.org/review-history/11317