

***In-vitro* effects of aqueous and ethanolic extracts of *Parkia biglobosa* (Jacq.) Benth on selected microorganisms**

Osemwegie O. O. and Dahunsi S. O.

Department of Biological Sciences, Landmark University, Omu-Aran, Kwara State, Nigeria

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Abstract

The *in-vitro* effects of aqueous and ethanol extracts of *Parkia biglobosa* (Jacq.) R.Br. ex G.Don synonymous to *Parkia africana* (common name: African locust bean) were investigated against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Aspergillus niger*, *Microsporum gypseum*, *Aspergillus fumigatus*, *Microsporum audoni*, and *Aspergillus flavus*. *Staphylococcus aureus* growth was inhibited by different concentrations of aqueous and ethanol infusion extracts with a Minimum Inhibitory Concentration (MIC) range of 3.125-200mg/ml. All target fungal species except *Microsporum gypseum* were unaffected by the extracts and had the lowest MIC range of 12.2 – 50 mg/ml. Results further showed that the unfiltered ethanol extract of the stem had the best zone of inhibition (31 mm) against *Staphylococcus aureus* while the unfiltered aqueous extract of the root had the least zone of inhibition value of 5 mm. Observed inhibitory activity of the extracts is directly proportional to increase concentration in gradient. Phytochemical analyses revealed the presence of flavonoid in all the extracts while terpenoids and saponins were limited to stem and root extracts respectively. Effective performances of filtered and unfiltered extracts of both the stem and the roots against target microbes are not markedly different from each other. This alludes to the relatively heavy presence of flavonoids, tannins and terpenoids observed in the extracts. The result obtained from this study further corroborates the antimicrobial qualities of *P. biglobosa* and recognizes it as an accessible plant resource for experimentations that focus on tackling the emergence of resistant pathogenic microbes of plants, animals and humans.

Keywords: antimicrobial, clinical pathogens, phytochemicals, locust bean, infusion, extraction.

Correspondence: osemwegie.omorefosa@landmarkuniversity.edu.ng

Introduction

The plant kingdom represents a resource pool of species with potent anti-microbial and medicinal potentials. They constitute the richest source of pharmaceuticals, nutraceuticals and folk medicine products across the globe. Increasing bad publicity of the side-effects of synthetic drugs on humans and their influence on the evolution of resistant microbial strains triggered research into plant genetic resources or their derivatives as suitable alternative therapeutics (Wink, 2012). It has also improved folk knowledge of medicinal plants and other natural products, and their preference in

revolutionizing pharmaceuticals (WHO, 2002; Abdallah, 2011).

Parkia biglobosa (Jacq.) R. Br. ex G. Don commonly referred to as African Locust bean tree is a multipurpose tree used directly or indirectly for a wide range of purposes e.g. timber, food, folk medicine, ornamentals or household trees, soil amendment, fire-woods, ornamentals and animal feeds, especially in the West African region (Obajuluwa and Ayokunumi, 2010). It equally provides small scale businesses to rural communities through the commercial sales of its fermented seeds as flavoring and fruits as snack. Recently, the plant ranked with other trees as putative remedy for soil erosion, desertification

and windbreak in West African regions. The most popular and significant use of *P. biglobosa* is as food (Okunola et al., 2011). The bark was reported as viable remedy for toothache, leprosy, eye sores, fever, hypertension, wounds such as ulcer and snake bite (Asuzu and Harvey, 2003; Alabi et al., 2005). Esenwah and Ikenebomeh (2008) observed the presence of anti-nutritional factors which Udobi and Onaolapo (2012) linked to the antimicrobial and other phytochemical potentials in many plant parts. It is a perennial dicotyledonous angiosperm that belongs to the family Fabaceae (-Mimosoideae) along with other tree legumes (Thiombiano et al., 2014). Geographically, they are distributed largely in the savannah regions of the world. The tree was reportedly introduced to West Africa during the transatlantic slave trades, it is deciduous, growing between 7 to 20 m high or 30 m in extreme cases (Ntui et al., 2012; Ikpeme et al., 2012).

Natural herbal medicine practitioners in many African nations have employed different parts e.g. seed, stem bark, root, leaves, pod, pulp, seed and flower of the locust bean tree as remedy to many human infections and ailments. A study conducted in Togo confirmed the medicinal values of *P. biglobosa* and its popularity in the nation's traditional healthcare services (Karou et al., 2011). The efficient wound-healing property of the plant in the south-western region of Nigeria was reported by Adetutu et al. (2011) while Traore et al., (2013) reportedly used the plant extracts as anti-malaria in Guinea. The anti-bacterial property of the plant was observed by Abioye et al. (2013) to compare with synthetic streptomycin in action and potency. Similarly, the crude extract from different parts of *P. biglobosa* showed positive inhibitory effect on the growth of methicillin resistant *Staphylococcus aureus* (MRSA). Stem bark at various concentrations (10 - 25mg/ml) was observed to be most active against MRSA isolates from orthopedic patients (Ajaiyeoba, 2002) suggesting strong antimicrobial potentials. The current state of information on the ethnobotanical values of this plant underscores the need for further research on their antimicrobial properties. Many other reports on the medicinal and antimicrobial values of the different parts of the plant abound in literature (Booth and Wickens, 1988; Abbiw, 1990; Millogo-Kone et al., 2008; Udobi et al., 2008; Adeshina et

al., 2009; Abioye et al., 2013). These lend credence to their widespread traditional uses in many African nations as therapeutics of bacterial and fungal infections. This study therefore aims at investigating the antimicrobial and phytochemical attributes of the stem and root of *P. biglobosa*.

Materials and Methods

Samples collection and preparation of extracts: The stem bark and root of *Parkia biglobosa* were harvested within Landmark University environ, Omu Aran, Nigeria. The samples were sliced separately into irregular pieces using a kitchen knife, washed under a flowing tap water to remove adhering sands, and then air-dried for 14 days. They were later macerated with a hammer mill machine and pulverized to powder using an electric blender for extraction and phytochemical screening.

Aqueous and ethanol extraction: Ethanol and aqueous extracts from the stem bark and the root of *P. biglobosa* were prepared. One hundred grams (100 g) each of dry powder of the stem bark and root were soaked in 1000 ml of 60% ethanol and left on a laboratory bench for 24 hrs from where they were manually agitated twice at irregular interval within the period. Each of the samples was extracted to exhaustion using a Soxhlet apparatus. The mixture of each sample was sieved using a 9 mm Whatman filter paper (No.1) with 2.5 μ m retention, and the filtrates concentrated using the method of Akintobi et al. (2013) after which it was modified for the removal of the residual ethanol by a rotary evaporator (ROVA 100 model). The dry crude extracts (filtered and unfiltered) were kept in desiccators until required.

Similarly, 100 g each of dry pulverized samples of the stem bark and root samples of *P. biglobosa* were soaked in 1000 ml of distilled water, left for 48 hrs in a shaker and agitated for 6 hrs daily to increase the surface area of extraction. The mixture was thereafter sieved using a Whatman filter paper (No.1) with 2.5 μ m retention after which the filtrates were concentrated using a rotary evaporator. The extracts (filtered and unfiltered) were kept in desiccators until required.

Phytochemical screening of plant extract: The phytochemical screening was done according to Sofowora (2008). Clean sterile test tubes were used for the analyses. Each test tube was filled

with 2 ml of the plant extract, 7 ml of distilled water and mixed thoroughly using a Vortex flow-meter for further tests.

Test for alkanoids (True alkaloids): Two (2) ml of aqueous and ethanol extracts from the stem bark and root of *P. biglobosa* were each mixed in 8 ml of 1% HCl, and filtered. Two milliliter of the filtrates were treated separately with both Dragendorff's and Mayer's reagents and observed for the presence or absence of precipitate and turbidity.

Test for saponin (Foaming test): Two (2) ml each of the distilled water diluted extracts was heat to boil in test tube, shaken vigorously and observed for a persistent stable froth (mass of small bubbles on liquid surface).

Test for terpenoids (Salkowski test): Five (5) ml of each of the extract was treated with 0.5 ml of acetic anhydride and ethanol respectively and each observed for a layer of reddish brown coloration.

Test for steroids: Two (2) ml each of the aqueous extract was treated with 2 ml acetic anhydride while the process was modified with the use of 2ml chloroform in the ethanol extract. Both solutions were then treated with 2 ml of concentrated sulphuric acid respectively and observed for a color change that indicates the presence of steroids (Venkatesan et al., 2009)

Test for flavonoid: Four (4) ml of each of the extracts was measured into separate test tubes and treated with 1.5 ml of 50% ethanol solution. Each resulting solution was warmed using a heating mantle and afterwards magnesium was added. To the resulting solutions, about 5-6 drops of concentrated hydrochloric acid (HCl) was added and observed for a red coloration indicating the presence of flavonoids.

Test for tannin: The extracts were diluted with distilled water in separate test tubes that were heated over a water bath. To each of the solution was added about 3-4 drops of 10% ferric chloride and observed for a blue coloration.

Test for polysaccharides (Reducing Sugars): A portion of plant extract (0.5 ml) of the plant extracts was added to 1 ml of water followed by 5-8 drops of Fehling's solution and heated over water bath. The mixture was later observed for a brick red precipitate that indicates the presence of reducing sugars.

Viable counts for bacterial isolates: The viable count of target bacterial isolates was

carried out by titration method which involved serial dilution of bacteria, plating out of each diluent and counting of obtrusive colonies on each plate and recorded as colony forming unit per ml (CFU/ml). A measure of 0.1 ml of each diluent was aseptically dispensed into previously dry-heated sterilized Petri dishes containing gelled Nutrient Agar (NA) and left on an aseptic laboratory bench. Thereafter, sterile swab sticks were used to streak each plate after dipping it in the emulsified titrant. The plates were then each sealed with two strips of adhesive tapes bottom-to-lid, incubated at 37° C for 24 h in inverted position on a clean laboratory work bench. The resultant colonies were obtained by calculating the average CFU for each dilution multiplied by the dilution factor. This process was repeated for *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

Susceptibility testing: This was carried out using the agar well diffusion method, Nutrient agar was prepared aseptically after which approximately 20ml of it was dispensed into labeled sterile Petri dishes and was properly mixed by swirling before allowing to set. The entire surface of the Nutrient agar plate was swabbed properly with a sterile cotton swab containing the target bacterial isolates by rotating approximately 60° C between streaking for even distribution from a stock culture. After this, a 5 mm diameter sterile cork-borer was used to bore equidistant wells (4 per medium) in the agar. The wells were filled with 1 ml of the extracted samples and allowed to set for 60 minutes to ensure proper diffusion of samples. The plates were then incubated on a laboratory work bench at room temperature (37°C) for 24 h without overturning them for observation and clear zones of inhibition measured with a meter rule.

Minimum Inhibitory Concentration (MIC) evaluation: Antimicrobial activities against *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Microsporum gypseum* were done using the broth dilution method. Cultures of *S. aureus* and *P. aeruginosa* were diluted to density of 10² CFU/ml in normal physiological saline. The plain tubes containing two-fold dilution of each test sample were inoculated by the diluent. *Staphylococcus aureus* was incubated aerobically at 37° C for 24 hrs while *Microsporum gypseum* was incubated at room temperature for 5 days.

The MICs were determined by turbidity detection (Ochei and Kolhatkar, 2000).

Minimum Bactericidal Concentration (MBC) evaluation: The MBC was obtained by sub-culturing 0.01ml of the **highest concentration of the agent** which shows visible growth. Tubes without visible signs of growth in the minimum inhibitory concentration (MIC) tube dilution test were subjected to fresh antibiotics-free media. The MBC was taken as the lowest concentrations that result in the killing of 99.9% bacteria (Ochei and Kolhatkar, 2000).

Results

The results showed that unfiltered ethanol extracts of the stem recorded the highest zone of inhibition measuring 31 mm for

Staphylococcus aureus while the unfiltered aqueous extract of the root had the least zone of inhibition of 9 mm diameter for *Pseudomonas aeruginosa* (Table 1). It also observed that only *Microsporium gypseum* was vulnerable to the crude extracts (filtered and unfiltered) derived from different parts (root and stem) of the plant except the filtered aqueous extract of the root (Table 2). The filtered ethanol extract of the stem had the best inhibition value of 18 mm (highest) compared to the lowest inhibitory zone value of 5 mm obtained from the filtered aqueous extract of the root against *Microsporium gypseum*. The extracts showed no inhibitory activity against other target fungal isolates used for the study.

Table 1: Zone of inhibition of crude extracts of root and stem of *Parkia biglobosa* against target bacterial isolates (mm).

Plant extract	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>
Root ethanol filtered	25	16
Root ethanol unfiltered	23	15
Stem ethanol filtered	26	17
Stem ethanol unfiltered	31	14
Root aqueous filtered	25	10
Root aqueous unfiltered	27	09
Stem aqueous filtered	-	-
Stem aqueous unfiltered	-	-
Distilled water (Control)	-	-

- = zero value

Table 2: Zone of Inhibition values of crude extracts of root and stem of *Parkia biglobosa* against fungal isolates (mm).

Target Organism	FEER	UEER	FEES	UEES	FAER	UAER	UAES	FAES
<i>Microsporium gypseum</i>	16	11	18	17	-	5	12	14
<i>Microsporium audonii</i>	-	-	-	-	-	-	-	-
<i>Aspergillus niger</i>	-	-	-	-	-	-	-	-
<i>Aspergillus fumigatus</i>	-	-	-	-	-	-	-	-
<i>Aspergillus flavus</i>	-	-	-	-	-	-	-	-

FEER= Filtered ethanol extract of root; UEER= Unfiltered ethanol extract of root; FEES= Filtered ethanol extract of stem; UEES= Unfiltered ethanol extract of stem; FAER= Filtered aqueous extract of root; UAER= Unfiltered aqueous extract of root; FAER= Filtered aqueous extract of stem; UAES= Unfiltered aqueous extract of stem; - = No inhibition; - = zero value.

The results for the MIC (MBC and MFC) are presented in tables 3, 4 and 5. The extracts that showed antimicrobial activity were selected for the MIC test. The ethanol extracts showed different levels of clear zones from 3.125 to 200

mg/ml for *Staphylococcus aureus* while the root aqueous extracts was observed to affect the bacterium at a narrower concentrations of 50 to 200 mg/ml.

Table 3: Minimum Bactericidal Concentration (MBC) for *Staphylococcus aureus*

Sample quantity (mg/ml)	Root ethanol filtered	Root ethanol unfiltered	Stem ethanol filtered	Stem ethanol unfiltered	Root aqueous filtered	Root aqueous unfiltered	Stem aqueous filtered	Stem aqueous unfiltered
200	NG	NG	NG	NG	NG	NG	++	++
100	NG	NG	NG	NG	NG	NG	+++	+++
50	NG	NG	NG	NG	NG	NG	+++	+++
25	NG	NG	NG	NG	+	++	+++	+++
12.5	NG	NG	NG	NG	++	+++	+++	+++
6.25	NG	NG	+/-	NG	+++	+++	+++	+++
3.125	+/-	+/-	+	+/-	+++	+++	+++	+++
1.56	+	++	++	++	+++	+++	+++	+++
0.78	++	+++	+++	+++	+++	+++	+++	+++
0.39	++	+++	+++	+++	+++	+++	+++	+++
0.19	+++	+++	+++	+++	+++	+++	+++	+++

NG= No growth; +/- = Scanty growth; += Moderately normal growth; +++ = Borderline growth; ++++ = Profuse growth

The ethanol extracts of the root and stem showed inhibitory activity against *Pseudomonas aeruginosa* at Minimum Inhibitory Concentrations greater than 50 mg/ml while the effect of the aqueous extracts range from scanty to profuse growth of the target microorganisms at even the highest MIC value of 200 mg/ml. The MBC results showed that *S. aureus* is more susceptible to the plant ethanol extracts over a wider concentration gradient compared to *P. aeruginosa*. The extracts were equally observed to be less effective against *P. aeruginosa* at concentrations lower than 25 mg/ml. All the extracts except the filtered aqueous extract of the root of *P. biglobosa* inhibited the growth of human pathogenic

Microsporium gypseum resulting in the appearance of clear zones at various concentrations between 50 and 200 mg/ml (Tables 4 and 5).

The results show stronger inhibitory activity for ethanol extracts against target isolates compared to aqueous extracts. Similarly, the extracts of the plant roots were more microbicidal at wider concentrations compared to stem extracts. In addition, the antimicrobial capacity of both filtered and unfiltered extracts used in this experimentation were not significantly different despite the expectation that the unfiltered extracts would be more microbicidal.

Table 4: Minimum Bactericidal Concentration (MBC) for *Pseudomonas aeruginosa*

Sample quantity (mg/ml)	Root ethanol filtered	Root ethanol unfiltered	Stem ethanol filtered	Stem ethanol unfiltered	Root aqueous filtered	Root aqueous unfiltered	Stem aqueous filtered	Stem aqueous unfiltered
200	NG	NG	NG	NG	+/-	+/-	+++	++
100	NG	NG	NG	NG	+	+	+++	+++
50	NG	NG	NG	+	++	++	+++	+++
25	+	++	+/-	++	+++	+++	+++	+++
12.5	++	+++	++	++	+++	+++	+++	+++
6.25	+++	+++	++	+++	+++	+++	+++	+++
3.125	+++	+++	+++	+++	+++	+++	+++	+++
1.56	+++	+++	+++	+++	+++	+++	+++	+++
0.78	+++	+++	+++	+++	+++	+++	+++	+++
0.39	+++	+++	+++	+++	+++	+++	+++	+++
0.19	+++	+++	+++	+++	+++	+++	+++	+++

NG= No growth; +/- = Scanty growth; += Moderately normal growth; +++ = Borderline growth; ++++ = Profuse growth

Table 5: Minimum Bactericidal Concentration (MBC) for *Microsporium gypseum*

Sample quantity (mg/ml)	Root ethanol filtered	Root ethanol unfiltered	Stem ethanol filtered	Stem ethanol unfiltered	Root aqueous filtered	Root aqueous unfiltered	Stem aqueous filtered	Stem aqueous unfiltered
200	NG	NG	NG	NG	++	NG	NG	NG
100	NG	NG	NG	NG	++	+	NG	NG
50	NG	+	NG	NG	+++	+++	++	NG
25	+	++	++	+/-	+++	+++	++	+
12.5	++	+++	+++	++	+++	+++	+++	++
6.25	+++	+++	+++	++	+++	+++	+++	++
3.125	+++	+++	+++	+++	+++	+++	+++	+++
1.56	+++	+++	+++	+++	+++	+++	+++	+++
0.78	+++	+++	+++	+++	+++	+++	+++	+++
0.39	+++	+++	+++	+++	+++	+++	+++	+++
0.19	+++	+++	+++	+++	+++	+++	+++	+++

NG= No growth; +/- = Scanty growth; += Moderately normal growth; +++ = Borderline

The phytochemical analysis of the stem and root extracts of the plant revealed the qualitative presence of flavonoids, saponins, tannins, terpenoids and polysaccharides (Table 6). The

aqueous extracts of the stem was observed to lack tannins, terpenoids and polysaccharides and have relatively low incidence of flavonoids and saponins.

Table 6: Results of Phytochemical Screening of Extracts Used for the Study.

Phytochemical	Extracts			
	EES	EER	AES	AER
Flavonoids	++	++	++	++
Tannins	+++	+++	-	+
Saponins	-	-	++	++
Terpenoids	++	++	-	-
Polysaccharide	+++	+++	-	++
Steroid	-	-	-	-
Alkaloids	-	-	-	-

Absent = -, present = +, moderately abundant = ++, very abundant = +++.

Discussion

The result from this study supports existing reports on widespread antimicrobials derived from African continental plant genetic resource pool that included *P. biglobosa*. The antimicrobial, medicinal, flavor, odor/scent, microhabitat and ecological niche properties of tropical plants are largely influenced by the nature and diversity of inherent phytochemicals (Udobi and Onalapo, 2012). It is the degree of solubility of these phytochemicals and the polarity of solvents used per time that informed the observed variations in different experimentations' results and the resultant antimicrobial effects on the target isolates used for the study. This observation is in concordance

with Taylor (2004) who extracted different ordinary compounds of the various parts of the same plant using solvents of various polarities. Additionally, these phytochemicals are implicated in the plants' adaptive fitness, protection and signal-induced interspecific associations within the ambient ecosystem (Heil, 2014). The level of antimicrobial potency was observed to be directly proportional to the concentration and diversity of soluble phytochemicals of the plant parts used (Abioye et al., 2013). This result attests to the socio-economic utility of *P. biglobosa* in the tropics especially as a veritable source of food for humans and livestock; income generation among rural communities and therapeutics in folk healthcare practices. Besides its use as

ornamentals, shade trees, timber for furniture and other works of art, ethnobotanical reports implicated it in the treatment of various human ailments such as colic, wounds, diarrhea, hemorrhoid, sore eyes, toothache, mouth ulcer and skin infections (El-Mahmood and Ameh, 2007; Idu et al., 2007). *P. biglobosa* ranks as one of the foremost ethnobotanically utilized trees in sub-saharan Africa. Its utility is indirectly linked to the composition of the secondary metabolites (Banwo et al., 2004; Millogo-Kone et al., 2006; Udobi et al., 2008).

The varying concentrations of both ethanol and aqueous extracts (filtered and unfiltered) of the root and stem respectively showed no inhibitory activity on all the target fungal isolates used in the study except *Microsporum gypseum*. This result, although inconsistent with reports from previous studies, suggests that most of the fungal isolates are insensitive to the different concentration range of the extracts used in the study (El-Mahmood and Ameh, 2007; Akintobi et al., 2013; Olabinri et al., 2013). Differences in the extraction method, nature and selection of solvents used across studies may have directly influenced the composition of phytochemicals in the extracts. This could have accounted for the observed inconsistency. The mechanism of enzymes exudation by fungi though unclear also have implication for the resultant antimicrobial effect of the infusion extracts of the plant root and stem against the dermatophyte; *Microsporum gypseum* compared to the more recalcitrant *Aspergillus* spp. Also, the difference in the suite of target fungi used may putatively have caused the variation noticed across studies. While further investigation is required in determining the optimal fungicidal concentration of *P. biglobosa* extracts obtained by infusion, it is logical to assume that the nature of the liquid extractor coupled with fungal innate enzyme mechanisms may have negative influence on MFC values (Hsouna et al., 2011). The hypersensitivity reaction of fungi to phytochemical toxicity viz-a-viz their relatively advanced cellular nature is not yet fully understood but may underlie the extreme polarity in inhibition values observed between bacteria and fungi. Hypothetically, it could be inferred that most bacterial isolates are by the reason of their cellular simplicity likely to be more susceptible to the plant's phytochemicals and its cytotoxicity effects (Udobi

and Anaolapo, 2012; Akintobi et al., 2013; Olabinri et al., 2013). Therefore an understanding of the morpho-physiology of microorganisms exposed to plant extracts' toxicity and the mode of action of either a suite of infused bioactive compounds or individual phytochemical will provide better understanding on the susceptibility and/or resistance of microorganisms to infusion extracts of plants.

Crude ethanol extracts of *P. biglobosa* were observed to be active against *Staphylococcus aureus* and *Pseudomonas aeruginosa* respectively. Comparatively, unfiltered ethanol extracts of the stem showed the best antimicrobial activity against these two bacterial isolates and this agreed with the work of Obajuluwa and Ayokunumi (2010) that evaluated the antibacterial effect of the methanolic extracts of leaves, stem bark and root of locust bean plant against wound isolated methicillin resistant *Staphylococcus aureus* (MRSA).

The ethanol extracts of both the stem and root of *P. biglobosa* were more effective against the target bacterial isolates than observed for aqueous infusion extracts. Consequently, ethanol extracts had wider MIC values that range from 3.125 to 200 mg/ml for target bacterial isolates (Akintobi et al., 2013). It was observed that all the plant extracts used for the study had flavonoid which according to Iwashina (2003) is responsible for many biological activities of plants notable among which is pollinator attractant, allelopathy and feeding attractant. This could have accounted for its presence in the different parts of the plant. Polysaccharides, tannins and terpenoids were moderately to abundantly present in the ethanol extracts of the stem and root of the test plant compared to saponins. This may be attributed to differential solubility characteristics of the phytochemicals. While the study did not examine the biological characteristics of each of the identified phytochemicals and their microbicidal role, it recognizes their mediation in antibiosis (Kennedy and Wightman, 2011). The result showed no alkaloids and steroids presence in the various aqueous and ethanol infusion extracts analyzed. Consequently, the qualitative presence of phytochemicals such as tannins, polysaccharides and terpenoids in the ethanol extracts underscore their antimicrobial property especially against bacterial isolates (Nascimento

et al., 2000; Gershenzon and Dudareva, 2007). This study though preliminary, is one of the numerous attempts at sourcing a suitable biologically-derived alternative antibiotic to resolve the rapidly emerging drug-resistant strains challenge of the world. While they result corroborates existing body of literature on and recognized the potentiality of *P. biglobosa* as good for exploration of quality antimicrobials, more studies involving larger spectrum of diverse target strains is key to discerning the large-scale pharmacological value of the test plant.

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