



PHYTOCHEMICAL AND ANTIBACTERIAL ACTIVITY OF LEAVE EXTRACTS OF *GUIERA SENEGALENSIS LAM* ON SELECTED SPECIES OF GRAM POSITIVE AND GRAM NEGATIVE BACTERIA

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Abstract

The aim of this study was to determine the preliminary phytochemical component and antibacterial activity of the leaves extract of *Guiera senegalensis lam* (Combretaceae) against three clinical isolates (*Staphylococcus aureus*, *E. coli* and *Klebsiella species*) using standard method of analysis. The test for phytochemical component revealed the presence of alkaloid, anthraquinolones, tannin and phlobatanins. The result of antibacterial activity showed that the ethanolic extract exhibit higher zone of inhibition against all the clinical isolates, with *E. coli* and *Klebsiella species* showed zone of inhibition of 35mm followed *Staphylococcus aureus* 30mm. Similarly, both aqueous and methanolic extract were sensitive to all the clinical isolate except *Klebsiella species* which showed resistance to the methanolic extract of the leaves. The results obtained in this research imply that the leaves extract of *Guiera senegalensis lam* could be useful in the treatment of infections caused by *Staphylococcus aureus*, *E. coli* and *Klebsiella species*.

Key words: Phytochemical, Antibacterial, *Guiera senegalensis* and Extraction

Introduction

Guiera senegalensis Gmel (Combretaceae) is a shrub of savannah region of West and Central Africa (Fiot *et al.*, 2006). The leaves are bitter-tasting and have widely recognized in African medicine as a “cure-all” in herbal concoctions (Hiermann and Bucar, 1994; Jigam *et al.*, 2011). Its stem consist of numerous knots that send out branches. The ash-grey stem and branches have fibrous or pubescent bark and bear opposing, short petiolated oval leaves, sometimes mucronate, sometimes even cordate at their base, about 2 to 4 cm long by 1 to 2 cm wide. These grey-green leaves, darker on their upper surface, display black spots on their lower surface and are slightly downy on both sides. These features lend the plant an overall silver green colour that is conspicuous in brush land (Silva *et al.*, 2008; Somboro *et al.*, 2011). In most part of the Northern Nigeria powdered leaves are combined with food as a general tonic and blood restorative and also to women as a galactagogue (Koumare *et al.*,

1968; Jigam *et al.*, 2011). The usual form of preparation for internal use is in decoctions or mixed with food preparations. *G. senegalensis* leaves are widely used for pulmonary and respiratory diseases, for coughs, as a febrifuge, colic and diarrhea, syphilis, beriberi, leprosy, impotence, rheumatism, diuresis and expurgation (Hutchinson and Dalziel, 1965; Zeljan *et al.*, 1998). Many previous studies indicated the presence in leaves of two alkaloids, flavonoids, naphthopyrans, tannins, and a naphthyl butenone; in roots, were only obtained tannins and the same beta-carboline alkaloids than in leaves (Koumare *et al.*, 1968; Combiere *et al.*, 1977; Bucar *et al.*, 1996; Mahmoud and Sami, 1997; Bouchet *et al.*, 2000; Ancolio *et al.*, 2002; Silva and Gomes, 2003, Fiot *et al.*, 2006). The branches, leaves, bark and roots of *G. senegalensis* are also used for the treatment of stomach pain and dysenteric diarrhea (Kerharo *et al.*, 1948; Aniagu *et al.*, 2005), syphilis, beriberi, leprosy and impotence (Kerharo *et al.*, 1948). It is also used in veterinary medicine among the Tukolor people in diets designed to increase body weight, reproductive capacity and milk secretion in animals (Kerharo and Adam, 1974; Somboro *et al.*, 2011). The literature reports several recorded uses for *G. senegalensis* in traditional medicine to treat various illnesses (Fiot *et al.*, 2004). It is recognized as being active against cough, respiratory congestion and fever (Kerharo and Adam, 1974), and is prescribed as an antitussive (Negrevergne, 1968; Faye *et al.*, 1980; Sanogo *et al.*, 1998; Diatta *et al.*, 2007), to ease breathing and to treat lung and bronchial disorders. It is also used against malaria (Benoit *et al.*, 1996; Ancolio *et al.*, 2002; Azas *et al.*, 2002; Somboro *et al.*, 2011). Some populations mix galls from *G. senegalensis* (gall nuts are frequently formed on the above-ground parts of the plant) with charcoal to make a highly diuretic powder prescribed in serious cases of oligouria and even anuria, and in particular for cerebral malaria. It has also been demonstrated that preparations made from galls possess antiviral properties (Lamien *et al.*, 2005; Aniagu *et al.*, 2005). Powdered dried leaves associated with *Melanthera scandens* are administered by the nasal route to treat headaches and sinusitis (Berhaut, 1967). The leaves are also used as a poultice on tumours and against the Guinea worm (Berhaut, 1967). Besides these uses in traditional medicine, some extracts of this plant have been found to possess pharmacological properties: antimicrobial (Le Grand, 1989; Bosisio *et al.*, 1997; Sanogo *et al.*, 1998), antifungal (Silva and Elsa, 2003) and antioxidant (Bucar *et al.*, 1998; Bouchet, 1998). Therefore, the present was to determine the phytochemical and antibacterial activity of *Guiera senegalensis* among some selected species of gram positive and gram negative bacteria.

Materials and Method

The leaves of *Guiera senegalensis* were collected from various part of Maiduguri metropolis. The plant was identified and authenticated by a plant taxonomist in the department of Biological Sciences University of Maiduguri, Borno state. The plant leaves stored were dried under laboratory condition and homogenized to fine powder.

EXTRACTION OF GUIERA SENEGALENSIS LEAVES

Seventy gram (70g) of the powdered plant leaves was soaked in 350ml of methanol, ethanol and distilled water in separate conical flask, which was thereafter stopper with a rubber cork and left for 24 hour. The resultant suspension was then filtered using muslin cloth. The filtrate was evaporated at room temperature (Oladunmoye, 2007).

PHYTOCHEMICAL SCREENING

Test of Alkaloid

The plant filtrate was treated with Mayer's reagent (Potassium mercuric iodide). Formation of yellow coloured precipitate indicates the presence of alkaloid (Tiwari *et al.*, 2011).

Test of Anthraquinolones

Five (5ml) of plant extract is shaken with 10ml benzene and 5ml of 10% ammonia solution is added. The mixture is shaken and the presence of pink, red or violet colour in the ammonical (Lower) phase indicates the presence of anthraquinolones.

Test of Flavonoid

The plant extract were extract treated with few drops of sodium hydroxide solution. Formation of intense yellow colour, which becomes colorless on addition of dilute acid, indicates the presence of flavonoid.

Test for Tannin

1% of gelatin solution containing sodium chloride was added to the plant extract. Formation of white precipitate indicates the presence of tannins (Tiwari *et al.*, 2011).

Test for Saponin

The plant extract were diluted with distilled water to 20ml and this was shaken in a graduated cylinder for 15minutes. Formation of 1cm layer of foam indicates the presence of Saponin (Tiwari *et al.*, 2011).

Test for Phlobotanins

Dilute hydrochloride acid was added to 2ml of leaves extract in a test tube. The appearance of red precipitate indicates the presence of Phlobotanins.

Test of Isolates

The isolates used for this research work was obtained from medical microbiology laboratory, University of Maiduguri teaching hospital, Maiduguri, Borno state, Nigeria, after confirmation. The isolates include *E. coli*, *Staphylococcus aureus* and *Klebsiella species*.

Results and discussion

The result of phytochemical analysis of the leaves extract of *Guiera senegalensis* was presented in Table 1. The phytochemical screening showed that the leaves extract contained alkaloid, Anthraquinolones, tannins and Phlobotanins, while flavonoid and Saponin were not found in the leaves extract. The result of this study correlates with the finding of Williams *et al.* (2009) which showed that the root extract of *Guiera senegalensis* contained alkaloid, Anthraquinolones, Saponin, phlobatanin and tannins. The result of this study is similar to observation made by Sule *et al.* (2001) which showed that *Guiera senegalensis* contained Anthraquinolones, alkaloid, tannin, ascorbic acid. The presence of this phyto-component in is very important, because they play a vital role as antimicrobial, antidiarrheal and antihelminthetic agent. The presences of alkaloid in the leaves of *Guiera senegalensis* will make it to intercalate into cell wall and DNA of pathogens and also inhibit the release of autocoid and prostaglandis (Antimicrobial). It also possesses anti-oxidating effect, thus reduce nitrate generation which is useful for protein synthesis, and suppress transfer of sucrose from stomach to small intestine. This compound also acts as antihelminthetic by diminishing the support of glucose to the helminthes, which act on central nervous system causing paralysis (Tiwari *et al.*, 2011). Presence of tannin in the leaves make it to possess antimicrobial property (By binding to adhesion, enzymes inhibition substrate deprivation).

The result of antibacterial activity of *Guiera senegalensis* leaves (Table 2) revealed that the extract possess high bactericidal activity on the clinical isolates used in this study. The isolates tested were *E. coli*, *Staphylococcus aureus* and *Klebsiella species*. All the extract was shown to exhibit broad activity against all the clinical isolates (except *Klebsiella species* which showed resistant with methanolic extract) as compared to the sensitivity of the isolate to the standard septrin. The minimum inhibitory concentration (MIC) (Table 3) of ethanolic extract of the leaves was for *E. coli* followed by *Staphylococcus aureus* and *Pseudomonas species* and ranges from 10 to 1.25µg/l. The ethanolic extract of the leaves produced impressive antibacterial activity against the entire tested organism with zones of inhibition ranging between 30 and 35mm. *Klebsiella species* was not susceptible to methanolic extract of the leaves. Comparison of the activity of the Seprin (Positive control) with those of the extract shows that the extracts may serve an alternative source of treatment of infections caused by *E. coli*, *Staphylococcus aureus* and *Pseudomonas species*.

Conclusion

The results obtained in this research imply that the leaves extract of *Guiera senegalensis lam* could be useful in the treatment of infections caused by *Staphylococcus aureus*, *E. coli* and *Klebsiella species*.

Table 1: Phytochemical screening of leaves of *Guiera senegalensis*

Tested Phytocomponent	leaves of <i>Guiera senegalensis</i>
Alkaoid	+
Anthraquinolones	+
Flavonoid	-
Tannin	+
Phlobotannins	+
Saponins	-

Table 2: Antibacterial activity of leaves of *Guiera senegalensis lam* against some species of gram positive and gram negative bacteria

Diameter of zone of inhibition (mm)				
Isolates	Aqueous extract	Ethanolic extract	Methanolic extract	Ciprofloxacin
<i>E. coli</i>	15	35	29	20
<i>Staphylococcus aureus</i>	20	30	30	15
<i>Klebsiella species</i>	10	35	0	8

Table 3: Minimum inhibitory concentration (MIC) of ethanolic extract of the leaves of *Guiera senegalensis*

Isolates	10	5	2.5	1.25	0.125
<i>E. coli</i>	+	+	+	+	-
<i>Staphylococcus aureus</i>	+	+	-	-	-
<i>Klebsiella species</i>	+	+	-	-	-

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