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PHYTOCHEMICAL SCREENING AND ANTIMICROBIAL ASSESSMENT OF EXTRACTS OF THE PODS OF PARKIA BIGLOBOSA (JACQ. BENTH)

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Abstract

The phytochemical screening and antimicrobial assessment of the extracts of the pods of Parkia biglobosa were investigated. Qualitative phytochemical analysis revealed the presence of active principles such as the tannins, anthraquinones, flavonoids, saponins, steroidal nuclei, combined reducing sugar, reducing sugar, carbohydrates, cardiac glycosides and alkaloids, mostly in the aqueous and ethanolic extracts, although the petroleum ether extract was positive for Salkowskii and Liebermann - Burchard's tests. The antimicrobial tests indicated that both the aqueous and the ethanolic extracts were strongly active against Salmonella typhi, Pseudomonas aeruginosa, Staphylococcus aureus, Escherichia coli, Streptococcus pygenes and Proteus vulgaris. These results showed that this plant pods could be a potential source for new antimicrobials against enteric organisms.

Key words: Ethnomedicine,Phytochemical, antimicrobial, Parkia biglobosa

INTRODUCTION

For quite a long time now, the plant family *Mimosaceae* has been the subject of investigation in terms of the nutritional value of its seeds (Ajaiyeoba, 2002). The seeds of *Parkia biglobosa* are used for food seasoning especially in the West African region (Odunfa, 1986).The fruits pulp and seeds of this plant are also known to be rich in protein and amino acids (Ajaiyeoba, 2002).Hence, these seeds have been serving as a good source of protein for the low income earners of these countries. In fact, the local condiment, daddawa, is being referred to as "the poor man's meat" in some parts of northern Nigeria. Apart from proteins, the seeds of this all important plant have been found to contain oils whose predominant fatty acid is the arachidonic acid (Ajaiyeoba, 2002).

In addition, *Parkia biglobosa* is known as Dorowa in Hausa; Origili in Igbo; Igba or Irugba in Yoruba; Lonchi in Nupe and Lai in Gwari (Ajaiyeoba, 2002). It is popularly known as the African locust bean (Osundina, 1995), fern leaf or monkey cutlass tree (Motar and Carol, 2007). It is a small but widely spread genus in the savannah region of Africa, Brazil, Java and Surinan. Among the African countries where *Parkia biglobosa* can be found includes Senegal, Gambia, Guinea, Sierra Leon, Togo, Benin and Nigeria (Man *et al.*, 2003)

Besides its medicinal value, *Parkia biglobosa* has been used as food. For instance, the green matured pods are roasted and eaten by man. The dried seeds are fermented and used in the preparation of a protein and fat rich food condiment called daddawa while the yellow starchy pulp surrounding the seeds is an important food supplement rich in vitamin C and carbohydrates (Moctar and Carol, 2007). This dried powder from the pulp is also mixed with water to produce a drink or porridge (elonuwa) in Nupe land (Man *et al*, 2003). In the Gambia, Ghana and some parts of Nigeria, the boiled decoctions obtained from the pods are used to impart water resilience to floors, walls and ceramic pots (Keay, 1989). Apart from providing shades in the hot climates, the plant is used for the production of charcoal and firewood. Medicinally, at the local level, this plant finds a wide range of applications. For example, the decoction of its stem bark is used in the treatment of asthma, wounds, fever, toothache and as eye lotion (Odunfa, 1986; Man *et al.*, 2003). This decoction is also used to cure diarrhoea, leprosy sores, ulcers and skin infections (Johnson, 1983). In addition, the leaves of this plant can be made into lotions for curing sore eyes, haemorrhoids, toothache and

bronchitis. The flowers are gritted, macerated and used for curing hypertension while the infusion of the flower buds is used in curing lumbago and leprosy prophylactics (Johnson, 1983). The red tannins, obtained from the bark of this tree, are used for dyeing leather. In addition, the seed coat can be used, with indigo dye, to improve the luster of fabrics. The seeds, when smoked and powdered, can be made into infusion and taken as a drink to reduce blood sugar in diabetic patients (Johnson, 1983).
In agro forestry, *Parkia biglobosa* is used to fix atmospheric nitrogen in the soil, reduce soil erosion and increase soil fertility (Aliero, 2004).

The increasing resistance of most synthetically derived antimicrobial agents is of utmost concern. The search for suitable medicinal plants with potent active principles against microorganisms becomes imperative. Since the antimicrobial activity of *Parkia biglobosa* has been established from previous studies. The objective of this study is to identify the secondary metabolites present in the pod of this plant and examines the antimicrobial activities of petroleum ether extract, ethanolic extract and aqueous extract against pathogenic enteric isolates.

MATERIALS AND METHODS

Sample Collection and Pretreatment

The dried fruits of the plant were obtained around the suburb of Minna using a sickle. The plant and the fruits were then authenticated by Dr. Jonathan Gana of Department of Biology, Niger State College of Education, Minna. They were dried at room temperature in the laboratory for a week. The seeds, with the yellow pulp were removed by peeling and the dried husks were further dried for three days in the laboratory, crushed and pounded into powder using pestle and mortar. The powder sample was packed in air tight and waterproof polyethene bags for further use.

Extraction of the Pods

The powder sample (500g) was extracted with petroleum ether (40-60°C) for 24 hours by Soxhlet extraction. The extract was evaporated on a water bath, then scraped and kept in screw caped bottles, labeled as petroleum ether extracts (PE) and the defatted sample was air dried and further extracted with ethanol for 48 hours. This was then concentrated on a water bath and then stored in screw capped bottles, labeled ethanol extract (EE),
Another portion of the defatted sample was extracted with water by maceration, filtered, and concentrated by evaporation to dryness on a water bath. The extract was labeled aqueous extract (AE).

Phytochemical Analysis

The three extracts (PE,EE and AE) were subjected to phytochemical screening using various standard methods (Sofowora, 1982;Trease and Evans,1983;Harbone,1993; and Ajaiyeoba,1998) to test for the presence of alkaloids, cardiac glycosides,saponin, tannins,flavonoids, carbohydrates, terpenoids and steroids,anthraquinones,phlobatannins, reducing sugar combined reducing sugar.

Microorganisms

The microorganisms used in this study were *Salmonella typhi*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Staphylococcus aureus*, *Streptococcus pygenes* and *Proteus vulgaris*.These organism were obtained from stock cultures in the Department of Microbiology, Federal University of Technology,Minna,Nigeria.

Antimicrobial Screening

The antimicrobial screening of the various extracts was carried out using the Agar dilution method (Adeniyi *et al.*, 1996).This was used to determine the antimicrobial activity of the various extracts against the tested micro organisms. The nutrient agar used to dilute the sample solution to the required concentration was inoculated by surface streaking using sterile wire loop with the test organisms. The plates were incubated at 37°C overnight and observed for growth inhibition, after which the diameter of zones of inhibition was measured. Plates that had growth of the test organism inhibited at 2.0mg/ml were further diluted to determine the Minimum Inhibitory Concentrations (MIC) and Minimum Bactericidal Concentration (MBC).

Determination of Minimum Inhibitory Concentration (MIC) and Minimum Bacterial Concentration (MBC) of Plant extracts

The MIC of the extracts against the test organisms was determined using the broth dilution method. 0.5cm^3 of each extract solution at a concentration of 100mgcm^{-3} was added to 1.5cm^3 of nutrient broth. The test organisms were inoculated into each test tube mixed thoroughly and were then incubated at 37°C for 24 h. The tube with the lowest dilution which had no detectable growth was considered as the MIC.

To determine the MBC, a loopful of broth was collected from those tubes which showed no turbidity and inoculated on nutrient agar. The inoculated agar plates were incubated at 37°C for 24h. The concentration at which no visible growth was observed was noted as the MBC.

RESULTS AND DISCUSSION

Table 1: Phytochemical analysis of extracts of *Parkia biglobosa* pods

Constituents	PE extract	EE extract	AE extract
Alkaloids	-	+	+
Saponins	-	+	+
Flavonoids	-	+	+
Tannins	-	+	+
Cardiac glycosides	+	+	-
Phlobatannins	-	-	+
Anthraquinones	-	+	-
Steroids	+	-	+
Carbohydrates	-	+	+
Reducing sugars	-	+	+
Combined reducing sugars	-	+	+

+ : Present -: Absent

The results obtained in the phytochemical screening as presented in Table 1 showed the medicinal importance of the pods of this plant which various parts have been reported to be useful for the treatment of various ailments (Johnson, 1983; Odunfa, 1986; Ajaiyeoba, 2002 and Mann *et al.*, 2003). In conformity with Ajaiyeoba's finding, alkaloids was also found to be present in the pods. The pods were found to contain saponins, tannins, flavonoids, steroids and anthraquinones. These constituents were known to exhibit medicinal activity as well as physiological activity (Sofowora, 1993). Cardiac glycosides were found to be present in all the extracts. However, there is no difference between ethanolic and aqueous extract test conducted. This is due to the fact that they are both polar solvent. Both ethanol and aqueous extracts were positive for saponins, a class of compound known to be effective for the treatment of syphilis and other veneral diseases (Sofowora, 1993).

Flavonoids and tannins are known to have antibacterial and antifungal properties in ethanol and aqueous extract but absent in petroleum ether. These findings gave credence to the traditional medicinal application of the crude extract of the plants as remedies for asthma, internal and external wounds, fever, and toothache and as eye lotion (Odunfa, 1986; Man *et al.*, 2003).

Table 2: A ntimicrobial activities of the various extracts of *Parkia biglobosa* Pods

Organisms	Zone of inhibition(mm) P.E.	E.E.	A.E.
Salmonella typhi	-	16	10
Pseudonomas aerugnosa	-	15	8
Escherichial coli	-	19	12
Staphylococcus aureus	-	20	15
Proteus vulgaris	-	17	-
Staphylococcus pyogene	-	14	2

The results for antimicrobial screening indicated as the diameter of zone of inhibition is given in Table 2. The results showed the antibacterial activity of the extracts against the tested organisms. The ethanol extract was found to be the most active against all the test organisms, this could be due to the presence of the bioactive constituents present in the extract. These components have been reported by the researchers to be active against microorganism (Buwa and Staden, 2006). The demonstration of activity against the pathogenic bacteria by the plant is an indication that the plant can be used to develop new antibacterial agents to treat ailments caused by these organisms (Banso and Olutumayin, 2001). The zone of inhibition was measured by the use of a transparent plastic ruler and was observed to be above 14mm for ethanolic extract. It has been found that for Enterobactoriaceae to be regarded as sensitive to any antimicrobial agent, it must produce a zone of inhibition of greater or equal to 14mm. Consequently, this plant is a potential anti-microbial agent (Barry *et al.*, 1985).The petroleum ether extract showed no inhibitory effect against the microorganisms tested. The antimicrobial activity of these extracts may be due to the presence of some phytochemical constituents (Sofowora, 1982; Finar, 1983; Anna, 1990; Bep, 1996).

Table 3: **The Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentrations (MBC) of the extracts of *Parkia biglobosa* pods in (mg/L)**

Organisms	MIC	MBC
Salmonella typhi	0.8	1.0
Pseudonomas aerugnosa	0.1	0.2
Escherichial coli	0.6	0.8
Staphylococcus aureus	1.5	2.0
Proteus vulgaris	2.8	3.0
Staphylococcus pyogene	1.9	2.0

The MIC and MBC of the extracts ranged from 0.1 to 2.8mg/ml and 0.2 to 3.0 mg/ml respectively (Table 3). The antimicrobial potency of plants is believed to be due to tannins, saponins, alkaloids, anthraquinones and flavonoids. The MIC values for most of the extracts were lower than their MBC values, suggesting that these extracts inhibited growth of the test microorganisms while being bactericidal at higher concentrations.

CONCLUSION

The ethanolic extracts of *Parkia biglobosa* pods could be a possible source to obtain new and effective herbal medicines to treat infections caused by multi-drug resistant strains of microorganisms. However, it is necessary to determine the toxicity of the active constituents, their side effects and pharmaco-kinetic properties.

References

Adeniyi, B.A.; Odelola, H.A.; Oso, B.A. 1996. Antimicrobial Potentials of *Diospyros mesiliformis* (Ebenaceae). *African Journal of Medicinal Science*. 225:221-224

Ajaiyeoba, E.O. 1998. Comparative Phytochemical and Antimicrobial studies of *Solanum macrocarpum* and *Solanum toro* leaves. Fitoterapia. 70:184-186.

Ajaiyeoba, E.O. 2002. Phytochemical and Antimicrobial Properties of *Parkia biglobosa* and *Parkia biciolor* leaf Extracts. *African Journal of Biomedical Research*, 5(3): 125-129.

Aliero, B.L. 2004. Effects of Sulphuric acid, mechanical Sacrification and wet heat treatments on germination of seeds of African locust bean tree,(*Parkia biglobosa*). *African Journal of Biochemistry*, 3(3): 179-181.

Anna, L. 1990. Plants for people. Natural History Meseum Publications,. London. Pp.135-167.

Banso, A; Olutumayin, O.O. 2001. Phytochemical and antibacterial evaluation of aqueous extracts of *Daniella oliveri* and *Nauclea latifolia*. *Nigerian Journal of Biotechnology*. 1: 114-115.

Bary, A.L; Thornsberry, C. 1985. Susceptibility tests. Diffusion test procedure. *Journal of Chemical Pathology*. 19:492-500.

Bep, O.B. 1996. Medicinal plants in tropical West Africa. Published by the Press Syndicate of the University of Cambridge, London, New York, New Rochelle, Melborne and Sidney. Pp.124-127, 160-169.

Buwa, L.V.; Staden, J.V. 2006. Anti microbial and antifungal activities of traditional medicinal plants used against veneral diseases in South Africa. *Journal of Ethnopharmacology* 103:139-142.

Finar, I.L. 1983. Organic Chemistry, Stereochemistry and Chemistry of Natural Products. 5th Edition, Vol. 2. Published in the U.S.A. by Longman Inc. New York. Pp.513, 517-519, 696-697, 781-782, 784 and 793.

Harborne, J.B. 1993. Phytochemical Methods, 2nd Edition, Chapman and Hull, London, Pp. 288.

Johnson, H. 1983. Herbal Medicine Database in Africa, Stuttgart-Medpharm scientific publishers, New York Pp. 67-79.

Keays, R.J.W. 1989. Trees of Nigeria. Published in the U.S.A. by Oxford University Press, New York. Vol. 1, Pp.132-142 and 195-196.

Mann, A; Gbate M; Abdulkadir, N.U. 2003. Medicinal and economic plants of Nupe Land. Jube Evans Books Publications Bida, Nigeria pp. 137.

Motar, S; Carol, C. 2007. Seed Leaflet. A Macmillan Publication on Seedbank-Project. Publication No. 124 of September, West Sussex, U.K.

Odunfa, S.A. 1986. Dawadawa In Legume Based Fermented Foods. Reddy, N.R.; Pierson, M.D. and Salunkhe, D.K. (eds) CRC Press Inc., Florida pp.183-189.

Osundina, M.A. 1995. Response of seedlings of *Parkia biglobosa* (African locust bean) to drought and inoculation with Vesicular Arbuscular Mycorrhiza. *Journal of Botany*, 8: 1-11.

Sofowora, A. 1982. Medicinal Plants a Traditional Medicine in Afri Published by John Wiley and So Ltd, New York, Pp.6-55, 142-177.

Sofowora, A. 1993. Medicinal Plants a Traditional Medicine in Africa Edn. John Wiley and Sons Ltd, N York, Pp.6-65.

Trease, E; Evans, W.C.1983. Pharmacogno Baillere Twidall, London; Pp 21-205, 378 and 540.