

Effects of Methanol Extract of *Parkia biglobosa* Stem Bark on the Liver and Kidney Functions of Albino Rats

C.I. Ezekwe, Anaya Chinenye Ada and P.C. Ugwu Okechukwu

Department of Biochemistry, University of Nigeria, Nsukka, Enugu State, Nigeria

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Abstract: Extract of *Parkia biglobosa* stem bark is used in the treating of malaria, diarrhoea, pains and host of other diseases in Nigerian traditional medicine (NTM). The aim of this research was to determine the beneficial properties of *Parkia biglobosa* as drug and its effect on the vital organs (such as liver and kidney). To achieve this, parameters such as ALP, ALT, AST, catalase, urea, PCV, RBC count, MDA and creatinine concentrations in the blood were evaluated as well as the phytochemical screening. The results revealed that the extract did not induce significant changes in some of the hepatic and hematological parameters determined (such as ALT, MDA, AST, catalase and RBC). A decrease in alkaline phosphatase activity and PCV concentrations were also recorded. These results suggest that the hepatic integrity of the rats were preserved as the parameters evaluated did not show alteration in concentrations and activity of hepatic markers. On the other hand, urea and creatinine levels increased significantly indicating possibility of kidney damage. The phytochemical screening revealed the presence of tannins, alkaloids, plant-protein, flavonoids, saponins, terpenes, glycosides and reducing sugars in the methanol extract. This study has shown the functional characteristics and effects of the methanol extracts of the stem bark of *P. biglobosa* on liver and kidney function of albino rats in short time treatment with the extract as the liver remained intact whereas the kidney reflected changes.

Key words: Parkia Biglobosa • Stem Bark • Hepatic • Hematological • Phytochemical • Liver and Kidney Functions

INTRODUCTION

Plants have the ability to synthesize a wide variety of chemical compounds that are used to perform important biological functions. Many of these phytochemicals have beneficial effect on long-term health when consumed by humans and can be used to effectively treat human diseases. At least 12,000 of such chemicals have been isolated so far; a number estimated to be less than 10% of the total [1]. Most of the claims of efficacy of the extracts derived from *P. biglobosa* stem bark have been scientifically established. However, little information on their effects on body organs (kidney and liver) are available.

P. biglobosa have been used in Nigeria and other West African rural communities to treat variety of diseases [2]. The efficacy of the various preparations of *P. biglobosa* is widely acclaimed by the Hausa communities of northern Nigeria for the treatment of such diseases as malaria, diabetes mellitus and pains. The stem

barks is boiled in water and taken as a decoction for the treatment of malaria, inflammatory diseases and infections to diarrhea [3]. The bark soaked in ethanol is also used in some communities for antidiarrhoeal properties and as an effective anti- snake venoms that protects against neurotoxic, haemotoxic and cytotoxic effects of poisonous snakes [4]. Also, the leaves, fruits and seeds of *P. biglobosa* have also been used to manage various diseases [5].

The bark is also used with lemon for wounds and ulcers. In cote d' Ivoire and Nigeria, bark infusion is used as a tonic for diarrhea [6]. Bark is used as a mouthwash, vapour inhalant for toothache, or for ear complaints. It is macerated in baths for leprosy and used for bronchitis, pneumonia, skin infections, sores, ulcers, bilharzia, washes for fever, malaria, diarrhoea, violent colic and vomiting, sterility, venereal diseases, guinea worm, oedema and rickets and as a poison antidote. The leaves are also used for burns and toothache as well as for sore eyes in Gambia [7]. Recently, the attention of researchers

has been drawn to the great potentials in *Parkia biglobosa* as a source of an antibacterial agent. [8], have all reported the presence of plant secondary metabolites which are known to exhibit antibacterial activity against a wide range of organisms. This paper presented the report of the efforts made to identify the compounds in the stem bark of *Parkia biglobosa* responsible for their bioactivity.

The origin of *Parkia biglobosa* is traced to the West African sub-region where it was first encountered by the Scottish surgeon, Mungo Park as he explored the Niger basin between 1795 -1799. He went ahead to describe this tree in his writing "Travels in the interior districts of Africa. Robert Brown described the genus *Parkia* in 1826. He named it after Mungo Park, who made 2 remarkable journeys of exploration into the interior of West Africa in 1795-1797 and 1805. *Parkia biglobosa* is a multipurpose fodder tree that belongs to the family *Mimosaceae*.

Parkia biglobosa occurs in a belt between 5°N and 15°N, from the Atlantic coast in Senegal to southern Sudan and northern Uganda. The belt is widest in West Africa (maximum 800 km) and narrows to the east. It was probably introduced to São Tomé and Príncipe. Trial plantations have been established in Tanzania and African locust bean was introduced to the Caribbean region over 200 years ago, probably as a consequence of the slave trade and later possibly to Guyana. The use of the fermented beans of African locust bean dates back many centuries and was already described in the 14th century.

African locust bean is a multipurpose tree that is as highly valued as Shea butter tree. Fermented seeds ('soumbala', 'dawadawa', 'netetu') serve primarily as a condiment for seasoning sauces and soups. Roasted seeds are used as a coffee substitute known as 'Sudan coffee' or 'café nègre'. The mealy pulp from the fruits is eaten or is mixed with water to make a sweet and refreshing drink rich in carbohydrates. Boiled pods are used to dye pottery black; the ash is applied as a mordant. The bark is rich in tannins and may be used for tanning hides, but the resulting leather is often of moderate quality especially with regard to colour, which is often reddish, uneven and darkens when exposed to light. The leaves are sometimes eaten as a vegetable, usually after boiling and then mixed with other foods such as cereal flour. Young flower buds are added to mixed salads. In West Africa the bark, roots, leaves, flowers, fruits and seeds are commonly used in traditional medicine to treat a wide diversity of complaints, both internally and externally, sometimes in combination with other medicinal



Source: [10].

Fig 1: Bark, flower pod, leaves, seeds of *Parkia biglobosa*

plants. The bark is the most important for medicinal uses, followed by the leaves [9]. Medicinal applications include the treatment of parasitic infections, circulatory system disorders, such as arterial hypertension and disorders of the respiratory system, digestive system and skin. In veterinary medicine, a root decoction is used to treat coccidiosis in poultry. Green pods are crushed and added to rivers to kill fish. The nutritional value of the fish is not adversely affected so long as they are cooked or dried. The fruit pulp is used as an ingredient of feed for pigs and dogs. The seeds are added to poultry feed after treatment to remove their anti-nutritional properties. The leaves are a useful, but not very palatable fodder.

Aim of the Study: The aim of the present study was to extract and analyse the various phytochemicals in *P. biglobosa* and check their effects on organ functions of albino rats.

Objectives of the Study: To achieve the above aim, the following were done:

- Qualitative determination of phytochemicals and investigation of its effect on organ function.

MATERIALS AND METHODS

Plant Material: The stem bark of *P. biglobosa* was collected in the month of November, 2012 at the University of Nigeria. The authentication was done by Pharm. Ezea of Pharmacy Department, University of Nigeria, Nsukka. The plant material was cleaned and dried

under shade to avoid destruction of active compounds. The dried material was ground with an electric grinder into powder. This was stored in an air-tight container ready for extraction. Pulverized leaf sample of *P.biglobosa* (500g) was weighed out into a beaker; using Harvard trip. balance of 2Kg- 5lb (5lb was converted to 200g). Using measuring cylinder, 1000ml of methanol was measured and added into the bottle and was allowed to stand for 48h. After 48h, it was filtered using whatman filter paper into a conical flask (500ml×3). The filtrate was poured into a disc plate to enable evaporation at room temperature for 5 days, leaving the extract of the *P.biglobosa* leaf in the bowl. Using spatula the extract was collected and stored in a sample collection container at room temperature.

Determination of Percentage Yield:

$$\% \text{ YIELD} = \frac{\text{Weight of extract}}{\text{Weight of pulverized stem bark}} \times 100$$

The phytochemicals were carried out based on the procedures outlined by [11].

Animal Studies: The animals used in this work were Wistar albino rats of about 12-18 weeks. The animals were acclimatized for one week under standard environmental condition, with 12 hours light/dark cycle maintained on a regular vital feed and water.

The sixteen rats were weighed and grouped into three groups A, B and Control according to their body weight. Each of the rats in the cage was marked with picric acid to differentiate them. The rats received methanol extract of *P. biglobosa* stem-bark extract orally for a period of 14 days except the control that received only feed and water. The rats in cage-A received 200mg/kg of the extract, cage-B received 400mg/kg of the extract and cage C -control received only feed and water. The dose was administered to the rats according to their body weight as shown below.

$$\text{VOL. AMOUNT} = \frac{\text{mg/Kg b.w} \times \text{Wt. of animal in}}{1000 \text{ Conc. in mg/ml of extract}}$$

STOCK CONC = 1g = 20ml of normal saline

X = 1000 mg, X = 50mg/ml., b.w = body weight

- Haematological parameters of packed cell volume (PCV) and red blood cell (RBC) were determined using [12] methods.
- *In-vitro* determination of ALT activity in serum by the [13] colorimetric method was carried out using Quimica Clinica Applicada (QCA) test kit, Spain.

- *In-vitro* determination of AST activity in serum by the [13] colorimetric method was carried out using Quimica Clinica Applicada (QCA) test kit, Spain.
- Determination of ALP activity in serum was carried out by the [14] Quimica Clinica Applicada (QCA) test kit method (QCA, Spain).
- Malondialdehyde (MDA) was assayed using thiobarbituric acid reacting substances by the method of [15].
- Catalase was assayed using [15] methods.
- The concentration of serum urea was determined using the method of [16] as outlined in Randox kits, UK.
- The concentration of serum creatinine was determined using the method of [16] as outlined in Randox kits, UK.

RESULTS

Qualitative Phytochemical Analysis: The phytochemical test was used to analyze the bio-active compounds (phytochemicals) present in the plant extract and the degree of occurrence. The more the plus (+) signs, the more level of occurrence. The minus (-) sign represents no occurrence.

The result in Figure 2 shows that at $P > 0.05$, there was significant decrease in the level of Alkaline phosphatase with respect to the control at concentrations 200mg/kg and 400mg/kg

The result in Figure 3 shows that at $P > 0.05$, there was no significant difference in the level of Alanine Amino Transferase with respect to the control at concentrations 200mg/kg and 400mg/kg

The result in Figure 4 shows that at $P > 0.05$, there was no significant difference in the level of Aspartate amino transferase with respect to the control at concentrations 200mg/kg and 400mg/kg

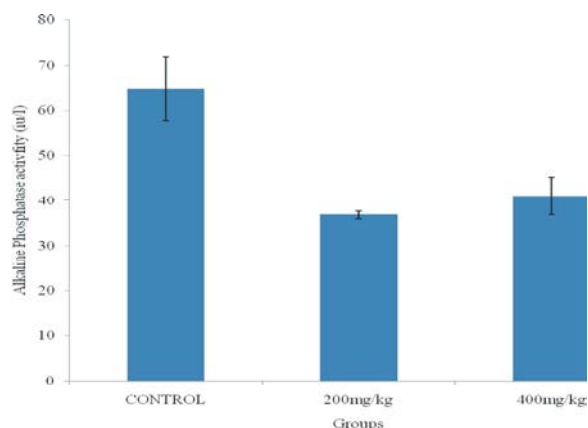
The result in Figure 5 shows that at $P > 0.05$, there was significant increase in the level of Urea with respect to the control at concentrations 200mg/kg and 400mg/kg

The result in Figure 6 shows that at $P > 0.05$, there was a significant increase in the level of serum Creatinine with respect to the control at concentrations 200mg/kg and 400mg/kg.

The result in Figure 7 shows that at $P > 0.05$, there was no significant difference in the level of serum malondialdehyde with respect to the control at extract concentrations of 200mg/kg and 400mg/kg.

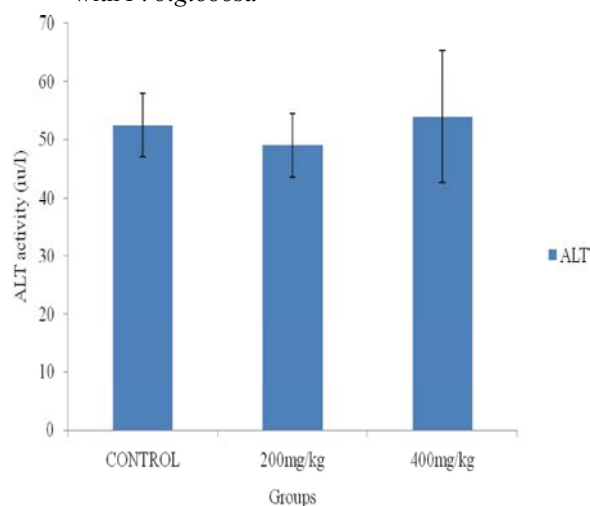
Table1: Phytochemistry table

Chemical compounds	Methanol extract
Alkaloids	+++
Flavonoids	+++
Glycosides	+++
Acidic compounds	-
Saponins	+++
Tannins	+++
Reducing sugar	++
Plant proteins	+++
Fats and oil	-
Resins	-
Carbohydrate	+++
Steroids	+
Terpenoids	+



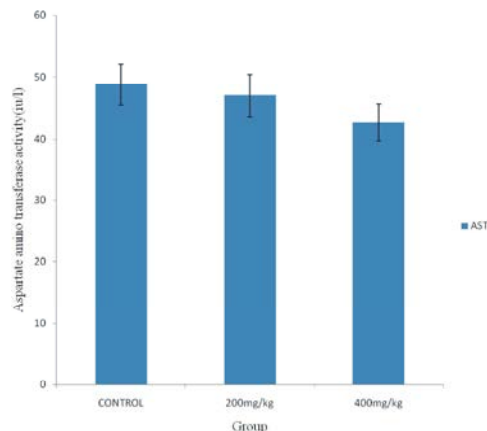
Determination of Alanine Amino Transferase (ALT) in rats:

Fig. 2: Alkaline Phosphatase Activity in the rats treated with *P. biglobosa*



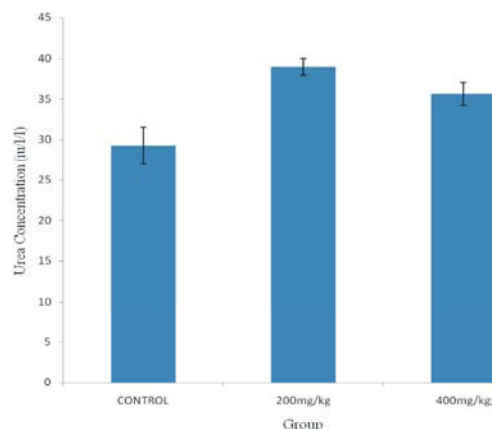
Determination of Alanine Amino Transferase (ALT) in rats:

Fig. 3: Alanine Amino Transferase Graph activity in rats treated with *P. biglobosa*



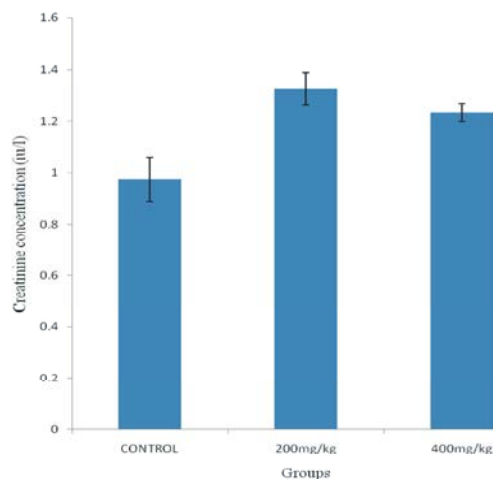
Determination of Aspartate Transaminase (AST)

Fig. 4: Aspartate amino transferase activity in rats treated with *P. biglobosa*



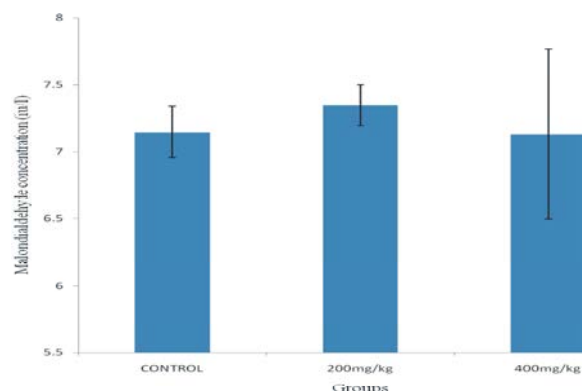
Determination of Urea in rats

Fig. 5: Urea concentration in blood serum of rats treated with *P. biglobosa*

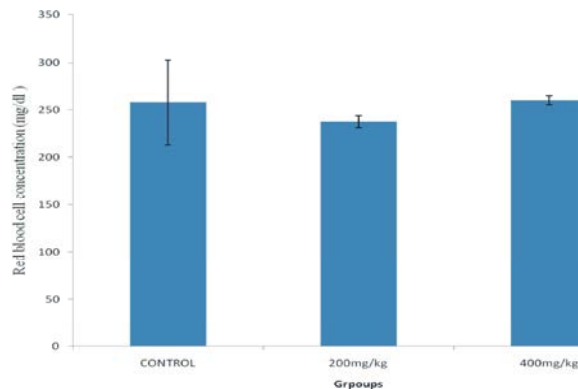


Determination of Creatinine in rats.

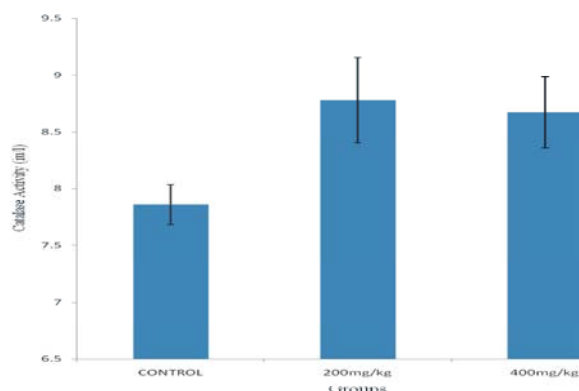
Fig. 6: Creatinine concentration in rats treated with *P. biglobosa*



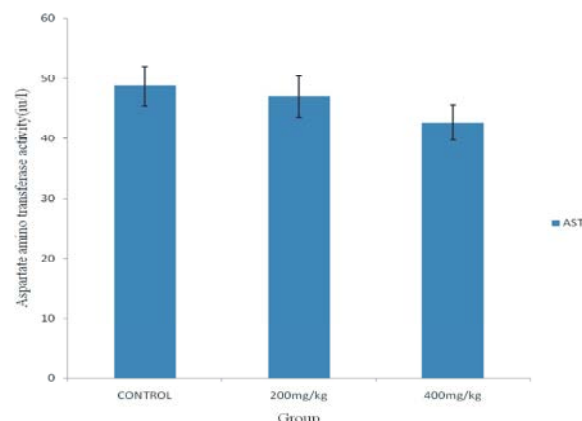
Determination of Malondialdehyde (MDA) in Rats:
Fig. 7: Malondialdehyde concentration in rats treated with *P. biglobosa*



Determination of Red Blood Cells in rats.
Fig 10: Red Blood Count Graph concentration in rats treated with *P. biglobosa*



Determination of catalase activity in rats.
Fig. 8: Catalase activity in rats treated with *P. biglobosa*



Determination of Packed Cell Volume in rats.
Fig 9: Packed Cell Volume Test Graph

The result in figure 8 shows that at $P>0.05$, there was no significant difference in the level of catalase with respect to the control at concentrations 200mg/kg and 400mg/kg

The result in Figure 9 shows that at $P>0.05$, there was no significant difference in the level of Packed cell volume with respect to the control at concentrations 200mg/kg and 400mg/kg

The result in Figure 10 shows that at $P>0.05$, there was no significant difference in the level of Red blood cells with respect to the control at concentrations 200mg/kg and 400mg/kg.

DISCUSSION

The plant kingdom represents an enormous reservoir of biologically active compounds with various chemical structures disease preventive properties. Plants contain secondary metabolites (phytochemicals), which are present in various concentrations. These phytochemicals include the alkaloids, steroids, flavonoids, terpenoids, tannins and many others. The active properties of many drugs found in plants are secondary metabolites [12]. Therefore, basic phytochemical investigation of *P. biglobosa* extracts for their major phytoconstituents is also vital. The hematological, liver, kidney and phytochemical analysis of *P. biglobosa* were investigated to determine any functional effect that may arise as a result of a short time animal exposure to the extracts within 24h period. The results of the study showed decrease in serum alkaline phosphatase level with respect to control and this was synonymous with the result of [5], who investigated the toxicity of the stem bark of *P. biglobosa* and found alkaline phosphatase level to be lower than that of the control at different doses of the extract administered. A rise in alkaline phosphatase level is usually a characteristics found in cholestatic liver disease [13]. As such, the significant reduction in ALP

levels by the methanol extract of *P. biglobosa* shows that no possible cholestasis occurred at the dose levels tested [13]. There was also significant decrease in the plasma PCV of the rats at the end of the study. This could be as a result of feeding factor. On the other hand, there was an increase in serum creatinine and urea concentration relative to the control. However, the result was in contrast with the result of [5] that recorded no change in levels of urea and creatinine. It is worthy of note that at 400mg/kg dose, two out of four rats died making 400mg/kg the LD₅₀ (lethal dose-dose that killed 50% of the animals) and this also contrasted work of [5], who at 5000mg/kg dose recorded no casualty. The serum concentration of AST, ALT, MDA, catalase as well as the concentration of RBC did not show any significant difference with the concentration of same parameters in the control.

Furthermore, the phytochemical analysis gave hint on the potentials of the stem bark of *P. biglobosa* and the possible explanation to results of the parameters above. The result showed an appreciable amount of flavonoid in the methanol extract of *P. biglobosa* and this corresponds with the result of [14], who recorded the same high concentration of flavonoids. A number of investigators have shown that coumarin, flavonoid, terpenoid and a host of other secondary plant metabolites including arginine and glutamic acids possess hypoglycemic effects in various experimental animals model [15]. However, this hypothesis stipulates that plant which contain terpenoid and/or flavonoids possess hypoglycemic activities in diabetic and normal mammal. Therefore the hypoglycemic activity of the methanol extract of stem bark of *P. biglobosa* may be due to the present of terpenoid, which was suggested to stimulate β -cells of the pancreas and the subsequent secretion of preformed insulin. One or more of the other chemical constituents of the plant especially flavonoid is also likely to have played a crucial role in the hypoglycemic action of the plant extract [14]. Saponin, plant protein and tannins were also present in appreciable amount. Study conducted by [16], showed that a large intake of tannins may cause kidney and liver damage.

CONCLUSION

This study has shown the effects of methanol extract of stem bark of *Parkia biglobosa* on not only hepatic and renal functions, but also on the general well being of the body. It can be said that methanol extract of stem bark of *P. biglobosa* due to presence of flavonoids and no

change in malondialdehyde levels could possess good anti-oxidant properties. The extract seemed to be liver friendly as none of the liver markers were altered. However, renal function seem to have been implicated which reflected in rise in urea and creatinine concentrations.

Recommendation: At the 400mg/kg dose, two out of four rats died (which supposes an LD₅₀), however toxicological study was not carried out in this study to find the cause of the deaths. On this ground, We recommend that major toxicological studies should be done on the methanol extract of stem bark of *P. biglobosa*. Apart from the casualties recorded, methanol stem bark extract of *P. biglobosa* seem to be a good raw material for drug making as there were appreciable amounts of favourable phytochemicals present and the hepatic markers of the rats were preserved at the end of the study.

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