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Evaluation of Methanolic Extract of Bambara Nut on Renal Indices of Wistar Rats

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Authors' contributions

This work was carried out in collaboration among all authors. Author AIA conceptualized and designed the study and also wrote the manuscript. Author AUM carried out the analyses of the study. Author OCN managed the literature searches. Author NOO managed the statistical analysis while author PNA wrote the protocol of the study. All authors read and approved the final manuscript.

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ABSTRACT

Background: Bambara nut is a common household food in Nigeria especially among the Igbos of the South Eastern part of the country.
Aim: This study is aimed at investigating the effect of Bambara nut on renal indices of Wistar rats.
Methodology: The Songkhla 1 variety (red seed coat) of Bambara nuts were locally sourced in Obinze area of Owerri, Imo State, Nigeria. The seeds were peeled and ground to a fine powder using a coffee grinder and extracted using *soxhlet apparatus and methanol as the solvent.* Twenty-four adult male Wistar rats were acclimatized for seven days during which they were fed *ad libitum* with standard feed and drinking water. They were randomly divided into four groups of six rats each. Rats in group A were administered distilled water while those in groups B, C and D were administered 100, 200 and 400 mg/kg body weight of *Bambara nut* extract 12 hourly for twenty-eight days *via* oral route of administration. At the end of the treatment, animals were sacrificed under diethyl ether as anaesthesia and blood samples were collected by cardiac puncture. Renal indices were determined using standard methods.

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Results: Investigation revealed that Bambara nut had no significant effect on the concentrations of urea, creatinine, Sodium, Potassium, Chloride and Bicarbonate at different administered doses when compared with those in control animals.
Conclusion: The results of this present study revealed that Bambara nut did not compromise the integrity of the kidney and thus not nephrotoxic.

Keywords: Bambara nut; electrolytes; nephroprotective; renal indices.

1. INTRODUCTION

Nephrotoxicity can be defined as the adverse effect of substances on renal function [1]. These substances can include molds and fungi, cancer therapeutics such as cisplatin, antibiotics such as aminoglycosides, metals such as mercury, arsenic and lead, and drugs of abuse such as cocaine. One indication of nephrotoxicity is a change in renal function as assessed by the glomerular filtration rate (GFR), blood urea nitrogen (BUN), serum creatinine (sCr), or bilirubin concentrations; however, nephrotoxicants can induce kidney damage without changing any established clinical marker of renal function. For example, studies have shown that proximal tubule necrosis in male Sprague Dawley rats exposed to gentamicin can be as high as 75% prior to any increases in BUN or sCr [2].

Bambara nut (*Vigna subterranea*) is classified under the family *Leguminosae*, sub-family *Faboidea* and genus *Vigna*. It is a seed of Africa origin used locally as a vegetable and it was first found in West Africa [3]. Bambara nut is a crop with great potential to sustain the dietary needs of both urban and rural communities [4]. Its seed consist of 49.0 to 63.5% carbohydrate, 15.0 to 25% protein, 4.5 to 7.4% fat, 5.2 to 6.4% fibre, 3.2 to 4.4% ash and 2% mineral [3].

Fig. 1. Bambara nut

It might be surprising to say that most people in Nigeria may not be conversant with the name Bambara nut as the local name is commonly used but it forms most parts of some families' daily meal. Locally, it is called 'Okpa' in Igbo, 'Epa-Roro' in Yoruba, 'Kwaruru' or 'Gurjiya' in Hausa [5]. The traditional uses of Bambara nut to treat several ailments are noteworthy, and present a gap for detailed study on the therapeutic and pharmaceutical value of the crop [6]. Jideani and Diedrick [7] reported that the medicinal role of Bambara nut is mainly based on information obtained from communities in several parts of Africa where this crop is reportedly responsible and useful for treatment of various ailments. For example as a treatment for diarrhoea, a mixture of Bambara nut and water from boiled maize are consumed; to alleviate the nausea associated with pregnancy, Bambara nut seeds are chewed and swallowed by pregnant women. Other prophylactic and therapeutic use of Bambara nut includes use against protein deficiency kwashiorkor, treatment of veneral diseases, treatment of polymenorrhea (roasted Bambara nut seeds are used); treatment for internal bruising, treatment of cataracts (mixture of water and crushed Bambara nut seeds are used [8]. Bambara nuts have been reported to possess both hypoglycemic and hypolipidemic properties in Wistar rats [9]. Recently, Megwas et al. [5] reported that Bambara nut ameliorated ethanol-induced oxidative stress in Wistar rats. Furthermore, Airaodion et al. [10] reported that Bambara nut is not hepatotoxic. This study is therefore aimed at investigating its effect on the renal indices of Wistar rats.

2. MATERIALS AND METHODS

2.1 Collection and Extraction of Plant Material

Bambara nut, the Songkhla 1 variety (red seed coat) were locally sourced in Obinze area of Owerri, Imo State, Nigeria and were identified by a botanist. Immature and damaged seeds were removed. The seeds were peeled and ground to a fine powder using a coffee grinder and stored

in screw-cap bottle at -20°C. The extraction was done using soxhlet apparatus and methanol as the solvent according to the methods described by Airaodion et al. [11,12]. About 25 g of the powder was packed into the thimble of the soxhlet extractor. 250 mL of methanol was added to a round bottom flask, which was attached to the soxhlet extractor and condenser on a heating mantle. The solvent was heated using the heating mantle and began to evaporate moving through the apparatus to the condenser. The condensate dripped into the reservoir housing the thimble containing the sample. Once the level of the solvent reached the siphon, it poured back into the round bottom flask and the cycle began again. The process was allowed to run for a total of 18 hours. Once the process was completed, the methanol was evaporated in a rotary evaporate at 35^{o}C with a yield of 2.17 g which represents a percentage yield of 8.68%. The extract was preserved in the refrigerator until when needed.

2.2 Animal Treatment

Twenty-four (24) adult male Wistar rats with body weight between 140 and 160 g were used for the experiment. They were acclimatized for seven (7) days during which they were fed *ad libitum* with standard feed and drinking water and were housed in clean cages placed in well-ventilated housing conditions (under humid tropical conditions) throughout the experiment. All the animals received humane care according to the criteria outlined in the 'Guide for the Care and Use of Laboratory Animals' prepared by the National Academy of Science and published by the National Institute of Health. They were randomly divided into four (4) groups of six (6) rats each. Animals in group A were administered distilled water (this group served as the control group) while those in groups B, C and D were administered 100, 200 and 400 mg/kg body weight of *Bambara nut* extract for twenty-eight (28) days, 12 hourly via oral route of administration. At the end of the treatment, animals were anaesthetized using diethyl ether and were sacrificed and blood samples were collected *via* cardiac puncture.

2.3 Determination of Renal Indices

Creatinine concentration was determined using Jaffe reaction described by Toora and Rejagopal [13]. Urea concentration was determined using a Randox Commercial Kit based on the methods of Fesus et al. [14]. The concentrations of Sodium (Na), Chloride (Cl), Potassium (K) and Bicarbonate (HCO_3^-) were determined using Biorex diagnostic kit according to the methods of Lorentz [15].

2.4 Statistical Analysis

Results are expressed as mean ± standard deviation. The levels of homogeneity among the groups were assessed using One-way Analysis of Variance (ANOVA) followed by Tukey's test. All analyses were done using Graph Pad Prism Software Version 5.00 and P values < 0.05 were considered statistically significant.

3. RESULTS

Investigation revealed that Bambara nut had no significant effect on the concentrations of urea, creatinine, Sodium, Potassium, Chloride and Bicarbonate at different administered doses when compared with those in control animals (Figs. 2-7).

4. DISCUSSION

Studies on the tissue biomarker alterations might reflect the metabolic abnormalities and cellular injuries in some organs. The kidney has extremely important function in detoxification and excretion of metabolic wastes and xenobiotics [16]. Exposure to toxic chemicals causes alterations in some tissue enzyme activities [17]. The kidneys control the excretion of urea, creatinine, and reabsorption of electrolytes into the blood. Defeat in activities of kidney will result in accumulation of electrolytes, urea, and creatinine in the biological fluid [18]. The results of renal indices of animals sequel to treatment with Bambara nut are presented in Figs. 2-7.

Administration of Bambara nut was observed to have no significant effect in the plasma concentrations of urea and creatinine when compared with those in control animals as presented in Figs. 2 and 3. Airaodion et al. [19], reported that the relationship between high renal restitive index (RI) and cardiovascular and renal outcomes is significant and persisted after multivariate Cox regression analysis, including traditional risk factors. The serum creatinine concentration is widely interpreted as a measure of the glomerular filtration rate (GFR) and it is used as an index of renal function in clinical practice [20]. Glomerular filtration of creatinine, however, is only one of the variables that determine its concentration in serum. Alterations in renal handling and metabolism of creatinine and methodological interferences in its

measurement may have a profound impact on the serum concentration of creatinine metabolism and is constant among individuals and over time, with the creatinine production rate being equal to the renal excretion rate [21]. In the theoretical situation where both criteria are satisfied, the serum creatinine is inversely proportional to the GFR, so that each halving of the GFR results in a doubling of the serum creatinine concentration [22]. Secretion of creatinine was observed even in early studies of the clearance of exogenously administered creatinine [23]. Mandell et al. [24], reported that the exogenous creatinine clearance decreased as the concentration of creatinine in the blood was acutely increased10-fold by creatinine infusion. This decrease was thought to be due to saturation of the tubular secretory mechanism, because the inulin clearance was not affected by this exogenous increase of the creatinine concentration in the blood [20,23]. Creatinine reabsorption during low rates of urine flow is thought to result from its passive back-diffusion from the lumen to the blood [21]. Thus, when urine flow rate is very low, passive reabsorption of creatinine might result in a lower creatinine clearance and a higher concentration of serum creatinine than what one would expect solely on the basis of the Glomerular Filtration Rate (GFR) [19,20]. Dietary protein deficiency leads to negative nitrogen balance and loss of muscle mass, thereby decreasing creatinine production. Less severe alterations in the diet, however, also may have important effects on the size of the creatine pool and creatinine excretion, which are independent of nitrogen balance and muscle mass. The nonsignificant difference observed in the concentrations of urea and creatinine in animals treated with different doses of methanolic extract of Bambara nut extract when compared with those in control animals might be suggestive that the extract did not compromise renal functional capacity and thus, not toxic to the kidneys.

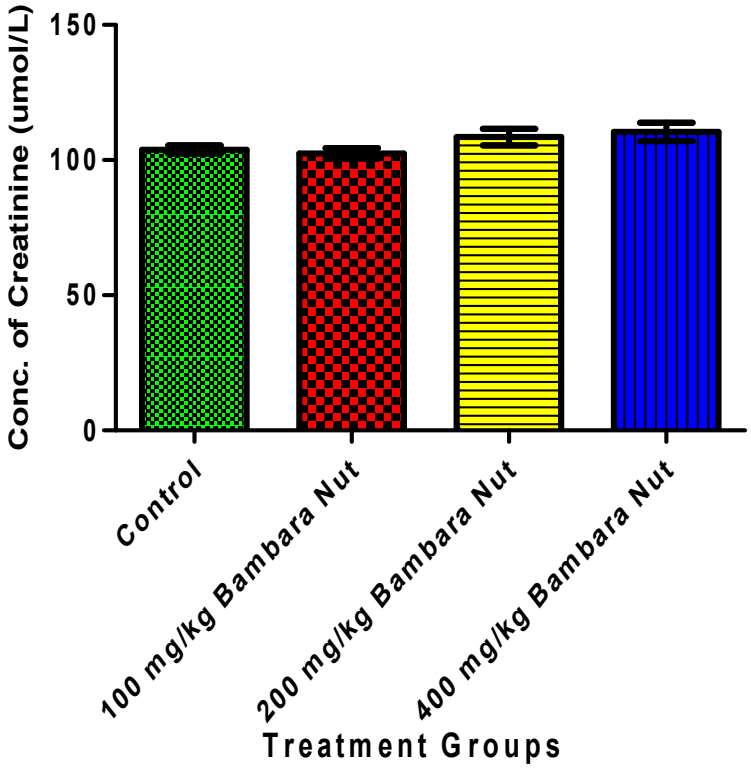

Fig. 2. Effect of Bambara nut extract on the concentration of creatinine of animals after 28 days of treatment

Results are presented as mean ± SD with n = 6

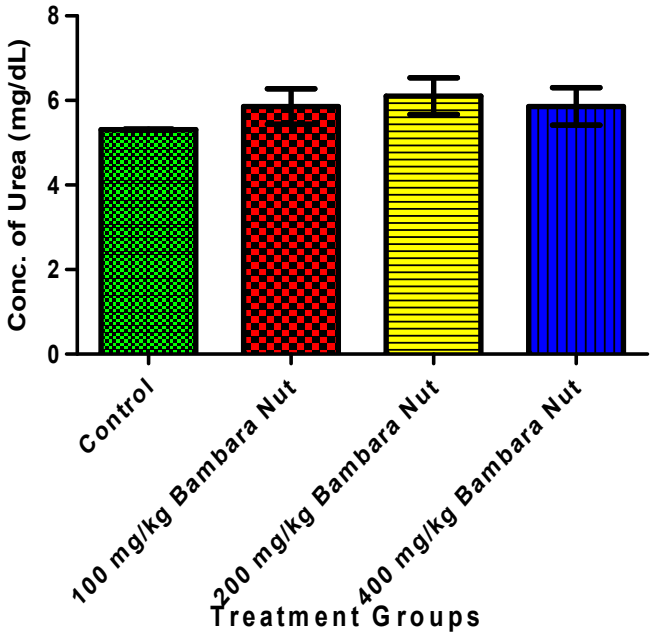

Fig. 3. Effect of Bambara nut extract on the concentration of urea of animals after 28 days of treatment

Results are presented as mean ± SD with n = 6

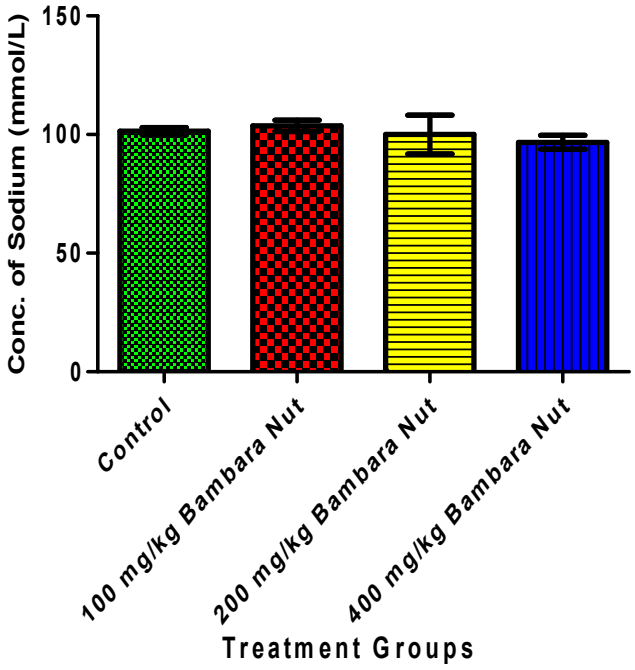

Fig. 4. Effect of Bambara Nut extract on the Concentration of Sodium of Animals after 28 days of Treatment

Results are presented as mean ± SD with n = 6

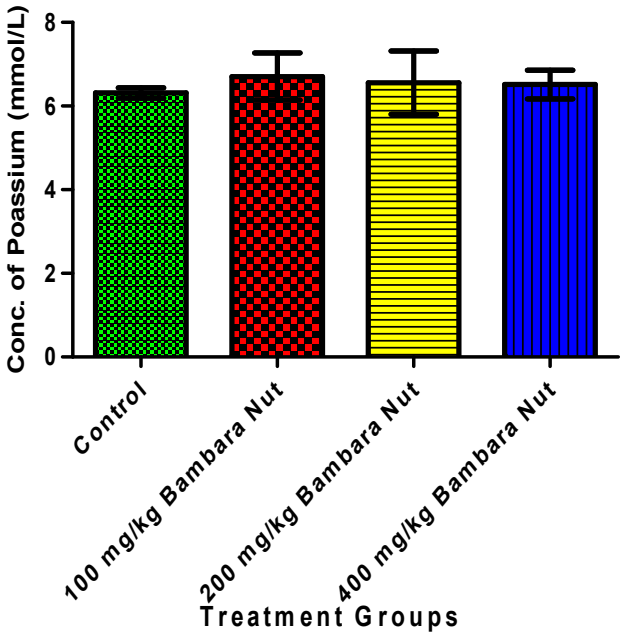

Fig. 5. Effect of Bambara nut extract on the concentration of potassium of animals after 28 days of treatment

Results are presented as mean ± SD with n = 6

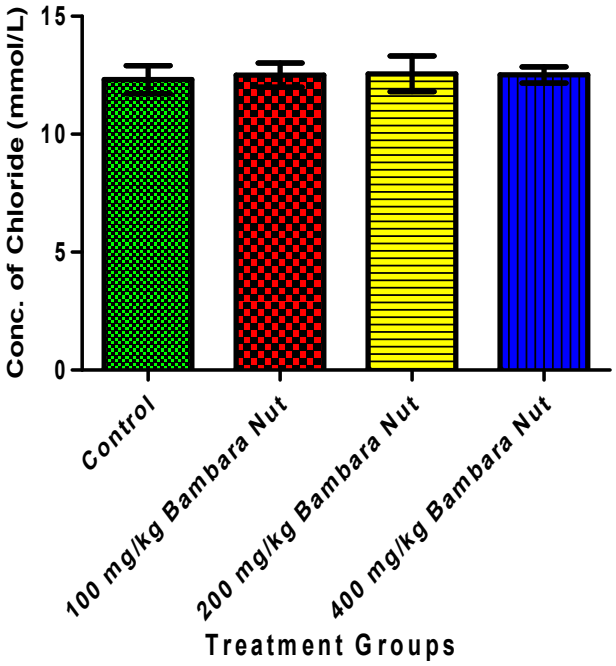

Fig. 6. Effect of Bambara nut extract on the concentration of chloride of animals after 28 days of treatment

Results are presented as mean ± SD with n = 6

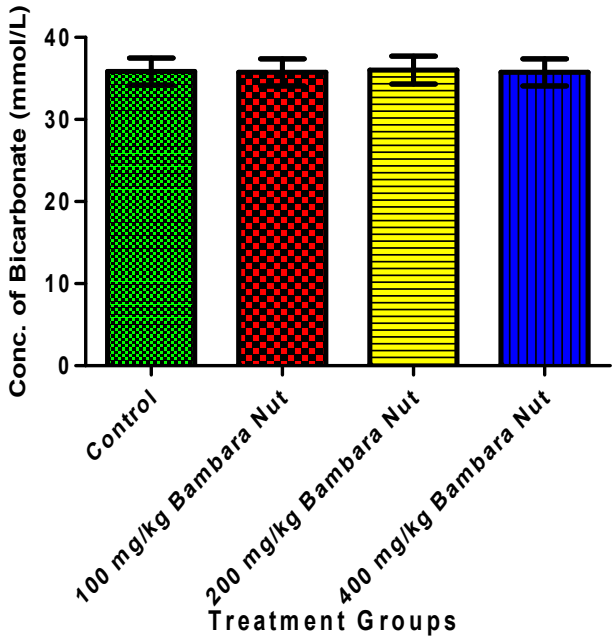

Fig. 7. Effect of Bambara nut extract on the concentration of bicarbonate of animals after 28 days of treatment

Results are presented as mean ± SD with n = 6

In this study, the concentrations of sodium (Na^+), potassium (K^+), chloride (Cl^-) and bicarbonate (HCO_3^-) were not significantly perturbed in animals exposed to different doses of Bambara nut for 28 days when compared with those in control animals (Figs. 4-7). Inorganic electrolytes occur in large quantities in both extracellular and intracellular fluids. Due to their ability to dissociate readily into their constituent ions or radicals, they comprise the single most important factor in the transfer and movement of water and electrolyte between three divisions of extracellular and intramuscular components [25]. Increase in serum Na^+ concentration has been reported to occur due to excessive loss of heat from the body fluid or increased production of aldosterone to other mineral corticoids which will in turns increase the reabsorption of Na^+ concentration. Aldosterone can achieve this since its action on the membrane aldosterone receptors has been linked to stimulating Na^+/H^+ exchanger [26]. Serum chloride is an electrolyte that can be used to assess renal functions. Therefore, the result of this study showed that methanolic extract of Bambara nut did not interfere with the normal functioning of the kidneys.

5. CONCLUSION

The results of this present study revealed that methanolic extract of Bambara nut did not compromise the integrity of the kidney and thus not nephron-toxic.

CONSENT

It is not applicable.

ETHICAL APPROVAL

Animal ethic Committee approval has been collected and preserved by the author.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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