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# Effect of Bambara Nut on Hepatic Biomarkers 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

**Aim:** This study sought to investigate the effect of Bambara nut on hepatic biomarkers 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-one days *via* oral route of administration. At the end of 28 days of treatment, animals were sacrificed under diethyl ether as anaesthesia and blood samples were collected by cardiac puncture. Hepatic biomarkers were determined using standard methods.
**Results**: Bambara nut was observed to unperturbed the activities of AST and ALT as well as the

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concentrations of total protein, albumin, globulin, and bilirubin. However, the extract was observed to significantly ($p<0.05$) increase the activity of ALP but decreased the activities of amylase and lipase when respectively compared with those in the control group.
**Conclusion:** Observations from this present study showed that Bambara nut is harmless to the liver. Thus, it is not hepatotoxic.

*Keywords: Bambara nut; health benefit; hepatic biomarker; safety.*

## 1. INTRODUCTION

Liver is the major organ which plays key roles in processing critical biochemical and physiological phenomena including metabolism and detoxification of endogenous and exogenous compounds, such as drugs and xenobiotics, homeostasis, growth, energy and nutrient supply [1]. Hepatic injury could occur by hepatotoxic agents such as drugs, alcohol, hydrocarbon and viral infections [2]. Liver diseases like jaundice, cirrhosis and fatty liver have been public health concern across the world [3]. Prevalence of chronic liver disease worldwide is 18.5% and cirrhosis is 4.5 to 9.5% while 2 million people die each year. In terms of medication, conventional or synthetic drugs are limited. Moreover they can have serious side effects [4,5]. Due to this fact, a huge number of medicinal plants have been used to figure out hepato-protective activities [6]. Approximately 160 phytochemical constituents originated from 101 plants have been reported to be potentially hepato-protective [7]. At present, medicinal plants have been a vital source of treatment of liver [8].

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 [9]. Bambara nut is a crop with great potential to sustain the dietary needs of both urban and rural communities [6]. 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 [9].

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 [10]. 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 [11]. Jideani and Diedrick [12] 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 [13]. Bambara nuts have been reported to possess both hypoglycemic and hypolipidemic properties in Wistar rats [14]. Recently, Megwas et al. [10] reported that Bambara nut ameliorated ethanol-induced oxidative stress in Wistar rats. This study therefore sought to assess its effect on the hepatic biomarkers 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. [15,16]. 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.

**Fig. 1. Bambara Nut**

## 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 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 28 days of treatment, animals were anaesthetized using diethyl ether and were sacrificed and blood samples were collected *via* cardiac puncture.

## 2.3 Determination of Hepatic Biomarkers

Aspartate Aminotransferase (AST) and Alanine Aminotransferase (ALT) activities were determined using Randox commercial Enzyme kits according to the method of Reitman and Frankel [17]. Alkaline Phosphatase (ALP) activity was determined by Phenolphthalein Monophosphate method described by Babson et al. [18]. Amylase inhibition assay was determined by the method of Bernfield [19]. Lipase activity was determined using Biorex diagnostic kit according to the methods of Lorentz [20]. Total bilirubin concentration was determined by diazo method described by Royden and Alfred [21]. Conjugated bilirubin concentration was determined by the method of Compernolle [22]. Unconjugated bilirubin was determined by subtracting conjugated bilirubin from total bilirubin.

## 2.4 Statistical Analysis

Results are expressed as mean $\pm$ 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

Bambara nut was observed to unperturbed the activities of AST and ALT as well as the concentrations of total protein, albumin, globulin, and bilirubin. However, the extract was observed to significantly ($p<0.05$) increase the activity of ALP but decreased the activities of amylase and lipase when respectively compared with those in the control group. These results are presented in Figs. 2-12.

## 4. DISCUSSION

Evaluation of hepatic biochemical parameters including enzymes (aspartate transaminase, alanine transaminase, and alkaline phosphatase) and metabolites (total proteins and albumin) are very useful in assessing the functional integrity of liver during subacute exposure of chemical substances or natural products/plant extracts [23]. The transaminase (ALT and AST) are enzymes of carbohydrate and amino acid metabolism while alkaline phosphatase is involved in hydrolysis of phosphate bonds. They are often used in assessing the functional integrity of liver, plasma membrane and endoplasmic reticulum [24].

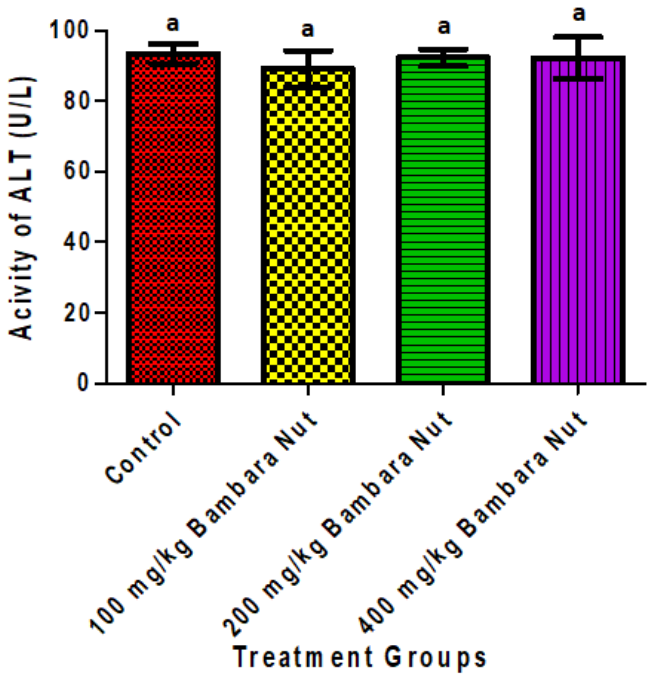


**Fig. 2. Effect of Bambara Nut on the Activity of Alanine amino transferase (ALT) of Animals after 28 days of Treatment**

*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

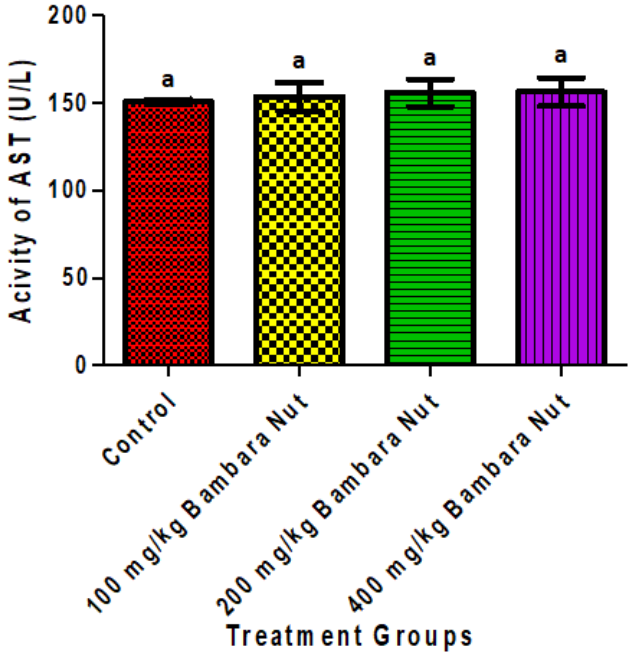


**Fig. 3. Effect of Bambara Nut on the Activity of Aspartate amino transferase (AST) of Animals after 28 days of Treatment**

*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

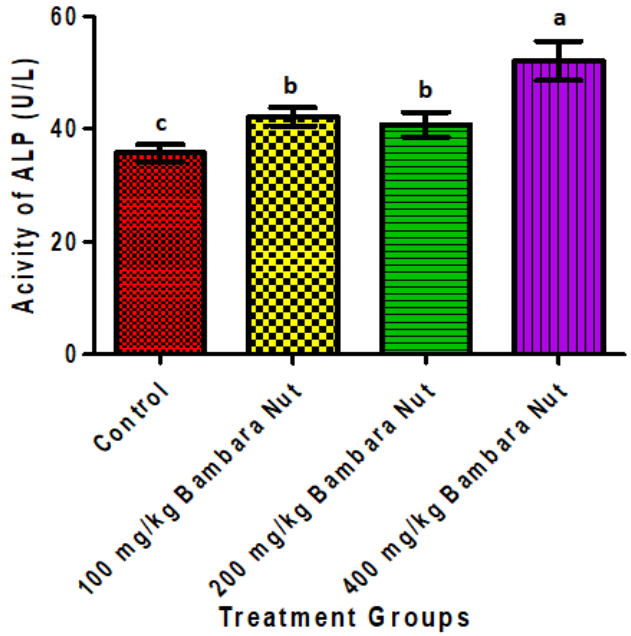


**Fig. 4. Effect of Bambara Nut on the Activity of Alkaline Phosphatase (ALP) of Animals after 28 days of Treatment**

*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

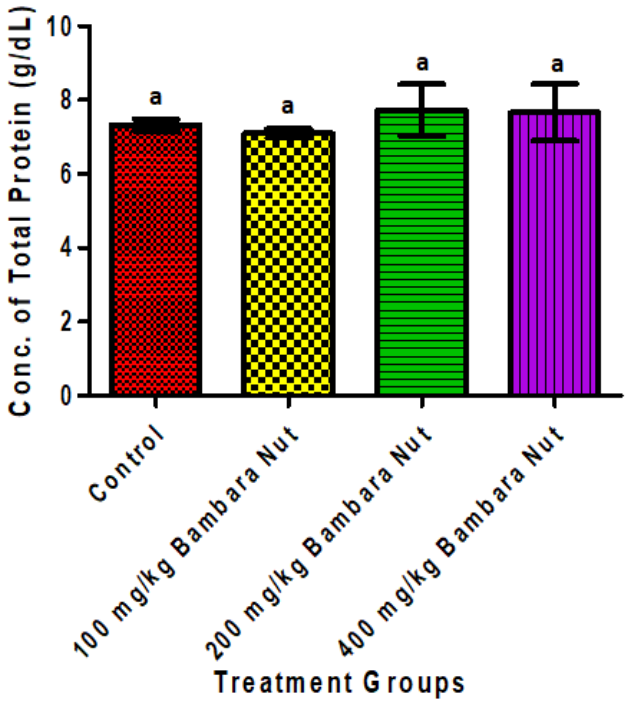


**Fig. 5. Effect of Bambara Nut on the Concentration of Total Protein of Animals after 28 days of Treatment**

*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

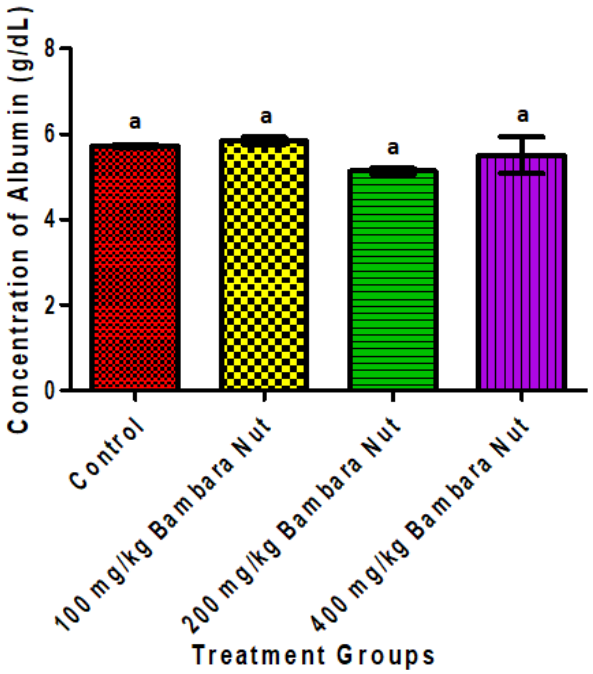


**Fig. 6. Effect of Bambara Nut on the Concentration of Albumin of Animals after 28 days of Treatment**

*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

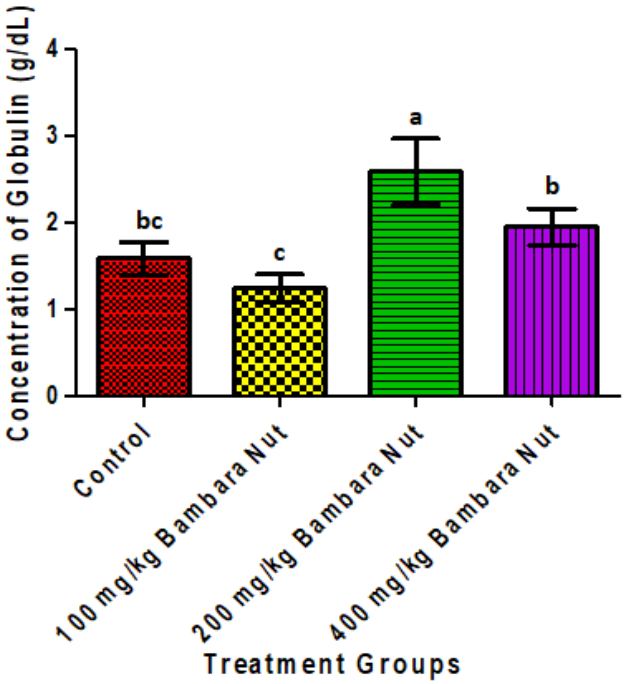


**Fig. 7. Effect of Bambara Nut on the Concentration of Globulin of Animals after 28 days of Treatment**

*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

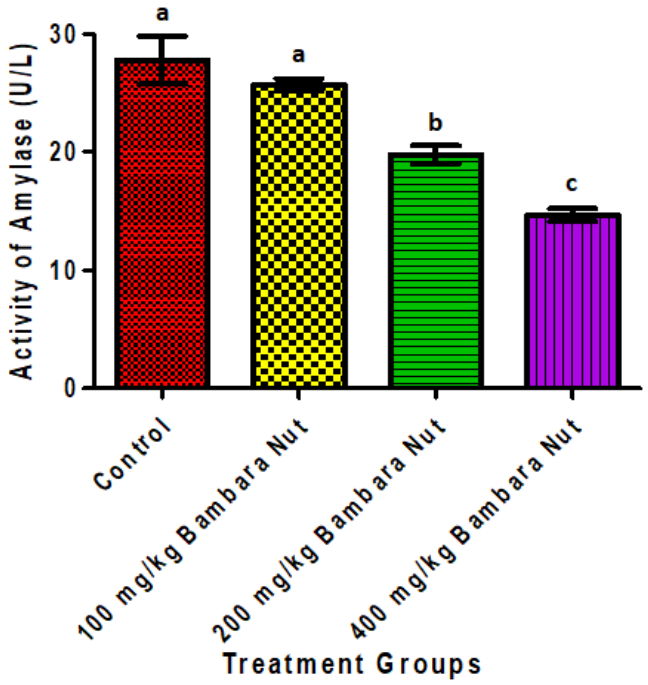


**Fig. 8. Effect of Bambara Nut on the Activity of Amylase of Animals after 28 days of Treatment**
*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

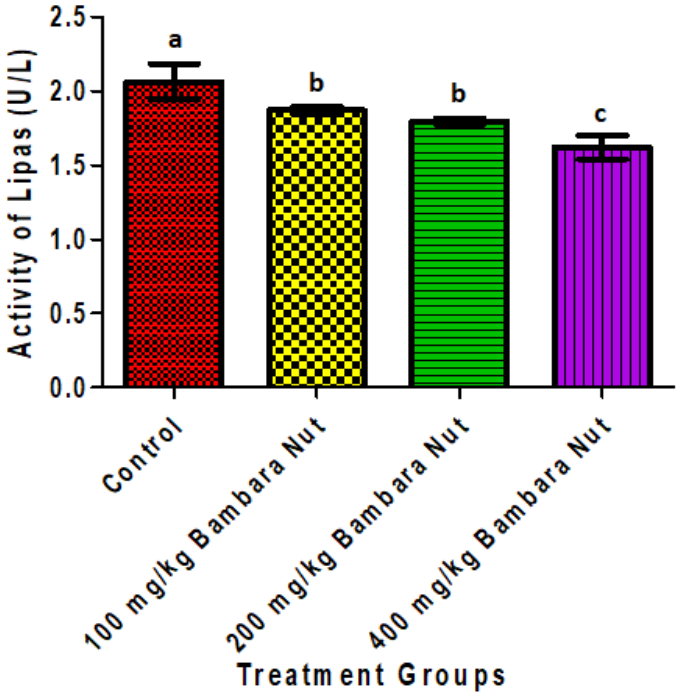


**Fig. 9. Effect of Bambara Nut on the Activity of Lipase of Animals after 28 days of Treatment**
*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

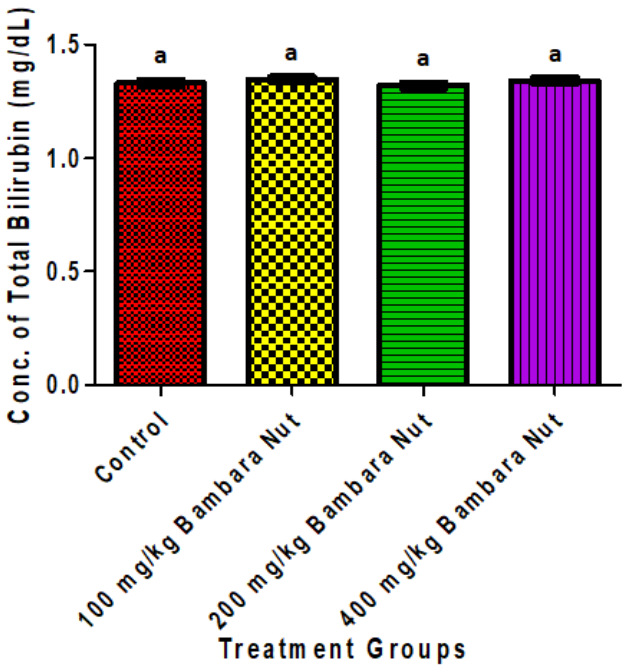


**Fig. 10. Effect of Bambara Nut on the Concentration of Total Bilirubin of Animals after 28 days of Treatment**

*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

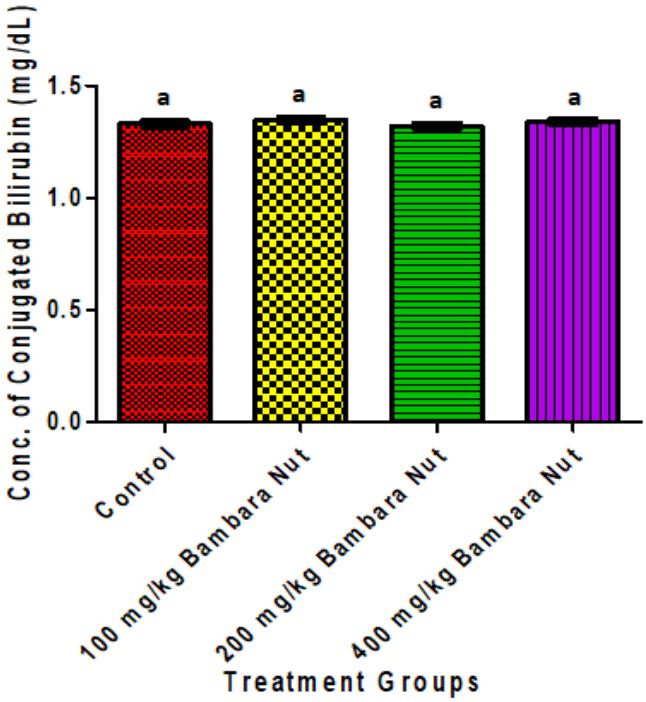


**Fig. 11. Effect of Bambara Nut on the Concentration of Conjugated Bilirubin of Animals after 28 days of Treatment**

*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

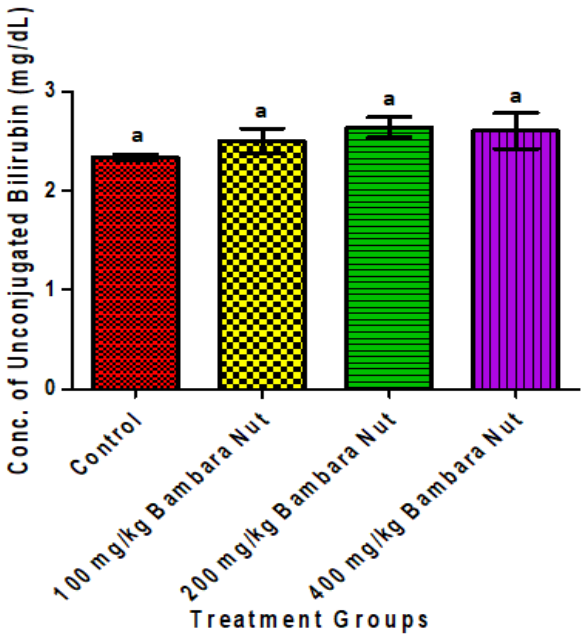


**Fig. 12. Effect of Bambara Nut on the Concentration of Unconjugated Bilirubin of Animals after 28 days of Treatment**

*Results are presented as mean ± SD with n = 6. Bars with different letters are significantly different at P<0.05*

In this study, no significant difference was observed in the activities of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) in animals treated with extracts of Bambara nut when compared with those of the control animals at P<0.05. It has been reported that an increase in the enzymatic activities of ALT and AST in the serum directly reflects hepatocellular damage [25]. Results of this study therefore suggest that extract of Bambara nut is not hepatotoxic at administered doses. This might be suggestive that treatment of animals with extracts of Bambara nut did not perturb the transcription of the genes involved in glucose uptake, glycolysis and lipogenesis [26]. Glucose represses the induction of inducible operons by inhibiting the synthesis of cyclic Adenosine monophosphate (cAMP) a nucleotide that is required for the initiation of transcription of a large number of inducible enzyme systems including the Lac operon. Cyclic AMP (cAMP) is required to activate an allosteric protein called catabolite activator protein (CAP) which binds to the promoter CAP site and stimulates the binding of ribonucleic acid (RNA) polymerase to the promoter for the initiation of transcription, but cAMP must be available to bind to CAP which binds to deoxyribonucleic acid (DNA) to facilitate transcription. In the presence of glucose, adenylase cyclase (AC) activity is blocked. AC is required to synthesize cAMP from Adenosine Triphosphate (ATP) [27]. Therefore, if cAMP levels are low, CAP is inactive and transcription does not occur. Thus, the effect of glucose in suppressing these inducible enzymes is by lowering cyclic AMP level. The extracts of Bambara nut had no effect on cAMP in treated rats, thus the nonsignificant effect in these inducible enzymes. ALT is considered most reliable marker of hepatocellular injury because it is solely confined to the liver, unlike AST which is also abundantly present in other body organs such as the kidneys, brain, and hearts [28,29].

Alkaline phosphatase (ALP) is involved in the hydrolysis of a wide range of phosphomonoester substrates. In this study, a significant (p<0.05) increase in the activity of ALP was observed in animals treated with 400 mg/kg body weight of Bambara nut when compared with those in the control group. ALP is a marker enzyme for the plasma membrane and endoplasmic reticulum of the tissues [23]. It is often employed to assess the integrity of the plasma membrane, since it is localized predominantly in the microvilli in the bile canaliculli, located in the plasma membrane, thus, its significant effect in this study might not be primarily related to hepatotoxicity. Since ALP

hydrolyses phosphate monoesters, its significant increase in animals exposed to Bambara nut could constitute a threat to the life of the cells that are dependent on a variety of phosphate esters for their vital process as it may lead to indiscriminate hydrolysis of phosphate ester metabolite of the liver [30]. Consequently this may adversely affect the facilitation of the transfer of metabolites across the cell membrane of animals exposed to high doses Bambara nut. This effect might be due to the high content of tannins in Bambara nut.

In this study, no significant difference was observed in the concentrations of total protein and albumin in animals treated with extracts of Bambara nut when compared with those of control animals. This might suggest that Bambara nut did not affect the synthetic ability of protein by the liver. It is also an indication that Bambara nut did not distort the functional activity of the liver by interfering with the equilibrium in the rate of synthesis and destruction, removal or clearance of total protein and albumin from the system of the animals [31]. Increase in total protein has been reported to lead to dehydration which is detrimental to cellular homeostasis [32] which negatively affect the metabolic activities of the liver and consequently the health of the animals. Albumin binds and transports metal ions, bilirubin, and drugs. Its level is used to assess the synthetic function of the liver. Serum protein levels are regulated via synthesis in the liver and its levels thus reflect the synthetic ability of the liver. Therefore, the result of this study is an indication that Bambara nut did not compromise the integrity of the liver.

Administration of Bambara nut in this study was observed to have inhibited the activity of amylase. Amylase is a key enzyme involved in starch breakdown. In humans, the diabetogenic process may be caused by immune destruction of the β-cells in the Islets of Langerhans in the pancreas and this is apparently mediated by white blood cell production of Reactive Oxygen Species (ROS) [33]. It is believed that inhibition of the enzymes involved in the digestion and uptake of carbohydrates can significantly decrease the postprandial increase of blood glucose level after a mixed carbohydrate diet and therefore can be an important strategy in the management of hyperglycemia linked to type 2 diabetes [34,35]. The inhibition of amylase by extract of Bambara nut collaborate the significant reduction in blood glucose level reported by Megwas et al. [10]. This effect could be attributed to the presence of biologically active phytochemicals such as phenolic and some non-phenolic constituents of the extract [36].

In consonance to the inhibition of amylase, lipase activity was also observed to be significantly inhibited by the extract of Bambara nut in a dose-dependent manner. Lipase is the enzyme responsible for digestion and absorption of triglycerides. Its inhibition is one of the widest studied methods used to determine the potential activity of natural products to inhibit dietary fat absorption [37]. Decrease in energy intake from dietary fat through inhibition of this enzyme may be an excellent strategy to prevent and treat obesity [38]. The inhibition of lipase by extract of Bambara nut collaborate the significant reduction in triglyceride level observed by Megwas et al. [10].

Bilirubin is the breakdown product of heme moiety of hemeoglobin; other hemeoproteins include cytochromes, catalase, peroxidase, tryptophan pyrrolase and a small pool of free heme [39]. Increase in concentration of direct reacting bilirubin in blood causes hyperbilirubinaemia, which is toxic under certain conditions inducing jaundice, hyperbilirubinemia-induced auditory dysfunction and neurotoxicity resulting in brain damage [40]. On the other hand, mild unconjugated hyperbilirubinaemia behaves as mild antioxidant and might offer protection against cardiovascular diseases and tumour development [41]. Recent research survey has reported that low concentration of direct reacting bilirubin induces stroke in body and sometimes causes cardiac problems too. Serum bilirubin levels are often enhanced under a variety of clinical conditions. In the circulation of blood, bilirubin is bound to serum albumin, which prevents its potential toxicity thought to be caused by free bilirubin [42]**.** Despite its high affinity of binding to albumin, bilirubin is rapidly and selectively taken up by the liver, biotransformed upon conjugation with glucuronate, and secreted into bile [41]. Thus bilirubin is converted into bilirubin glucuronic acid in the liver and excreted along with bile. The nonsignificant difference observed in the concentrations of bilirubin in this study suggests that the extract did not cause liver damage.

## 5. CONCLUSION

Observations from this present study showed that Bambara nut is harmless to the liver. Thus, it is not hepatotoxic.

## 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.

## REFERENCES


1. Mahmood DN, Mamat SS, Kamisan HF , Yahya F, Kamarolzaman FFM, Nasir N , Mohtarrudin N, Tohid, and Zakaria AZ . Amelioration of paracetamol-induced hepatotoxicity in rat by the administration of methanol extract of *Muntingia calabura* L. leaves. BioMed Research International. 2014;1-10.
2. Airaodion AI, Ogbuagu U, Ekenjoku JA, Ogbuagu EO, Airaodion EO, Okoroukwu VN. Hepato-protective efficiency of ethanol leaf extract of *Moringa oleifera* against hydrocarbon exposure. International Journal of advances in Herbal and Alternative Medicine. 2019;03(01):32-41.
3. Airaodion AI, Ogbuagu EO. Effect of *Cyperus esculentus* L. (Tiger nut) milk on hepatic and renal indices of Wistar rat. Asian Journal of Research in Nephrology. 2020;3(2):10-16.
4. Airaodion AI, Megwas AU, Ekenjoku JA, Ngwogu KO, Ngwogu AC. Nephro- and hepato-toxicity of common household insecticides used in Nigeria. International Research Journal of Gastroenterology and Hepatology. 2020;3(2):21-28.
5. Airaodion AI, Alabi OJ, Ogbuagu EO, Atiba FA, Olawoyin DS. Nephro and hepatotoxic effect of air-freshener in Wistar rats. Asian Journal of Research in Nephrology. 2020;3(2):1-9.
6. Airaodion AI, Ogbuagu EO, Ekenjoku JA, Ogbuagu U, Airaodion EO. Therapeutic effect of methanolic extract of *Telfairia occidentalis* leaves against acute ethanol-induced oxidative stress in Wistar rats. International Journal of Bio-Science and Bio-Technology. 2019;11(7):179-189.
7. Chatterjee TK. Medicinal plants with hepatoprotective properties, in Herbal Opinions, third ed. Books & Allied (P) Ltd., Calcutta. 2000;135.
8. Airaodion AI, Akinmolayan JD, Ogbuagu EO, Esonu CE, Ogbuagu U. Preventive and therapeutic activities of methanolic extract of *Talinum triangulare* leaves against ethanol-induced oxidative stress in Wistar rats. International Journal of Bio-Science and Bio-Technology. 2019;11(7):85-96.
9. Bamshaiye OM, Adegbola JA, Bamishaiye EI. Bambara groundnut: an under-utilized nut in Africa. Advances in Agricultural Biotechnology. 2011;1(1):60-72.
10. Megwas AU, Njoku OC, Akunne PN, Oladosu NO, Airaodion AI. Ameliorative potential of Bambara nuts against acute ethanol-induced oxidative stress in Wistar rats. International Journal of Health, Safety and Environment. 2021;07(2):1-12. Archive.org/megwas_et_al
11. Harris T, Jideani V, Le Roes-Hill M. Flavonoids and Tannins composition of Bambara groundnut (*Vigna subterenea*) of Mpumalanga, South Africa. Heliyon. 2018;4(9):83-88.
12. Jideani VA, Diedricks CF. Nutritional, therapeutic and prophylactic properties of *Vigna subterranean*. In: Antioxidant-antidiabetic agents and Human health. IntechOpen. 2014;187-207.
13. Koné M, Paice AG, Touré, Y. Bambara groundnut [*Vigna subterranea* (L.) Verdc. (Fabaceae)] usage in human health. In: *Nuts and Seeds in Health and Disease Prevention* (edited by V.R. Preedy, R.R. Watson and V.B. Patel). UK: Elsevier Inc. 2011;192-194.
14. Megwas AU, Akunne PN, Oladosu NO, Alabi OJ, Njoku OC, Airaodion AI. Effect of Bambara nut consumption on blood glucose level and lipid profile of Wistar rats. International Journal of Research and Reports in Hematology. 2021;3(5):1-12.
15. Airaodion AI, Adeniji AR, Ogbuagu EO, Ogbuagu U, Agunbiade AP. Hypoglycemic and hypolipidaemic activities of methanolic extract of *Talinum triangulare* leaves in Wistar rats. International Journal of Bio-Science and Bio-Technology. 2019;11(5):1-13.
16. Airaodion AI, Ekenjoku JA, Ogbuagu EO, Okoroukwu VN, Ogbuagu U. *Carica papaya* leaves might cause miscarriage. Asian Research Journal of Gynaecology and Obstetrics. 2019;2(2): 1-9.

17. Reitman S, Frankel S. A colorimetric method for determination of serum glutamate oxaloacetate and glutamic pyruvate transaminase. American Journal of Clinical Pathology. 1957;28:56-58.
18. Babson AL, Grecley SJ, Coleman CM, Phillips GE. The use of phenolphthalein in monophosphate as a substrate for serum alkaline phosphatase. Clinical Chemistry. 1966;12:482.
19. Bernfield, P. Enzymes of starch degradation and synthesis. Adv Enzymol. 1951;12:379–428.
20. Lorentz K. Lipase. In: Thomas L, editor. Clinical laboratory diagnostics. 1st ed. Frankfurt: TH-Books Verlagsgesellschaft.1998;95-97.
21. Royden NR, Alfred P. A New Diazo Method for the Determination of Bilirubin. *Clinical Chemistry.* 1962;8(6):570-578.
22. Compernolle F. Bilirubin conjugates: isolation, structure analysis and synthesis. Bilirubin, Vol 1: Chemistry. Boca Raton, FL:CRC Press. 1982;2:59-74.
23. Airaodion AI, Ogbuagu EO, Ewa O, Ogbuagu U, Awosanya OO, Adekale OA. Ameliorative efficacy of methanolic extract of *Corchorus olitorius* leaves against acute ethanol-induced oxidative stress in Wistar rats. Asian Journal of Biochemistry, Genetics and Molecular Biology. 2019;7(6):1-9.
24. Airaodion AI, Ogbuagu EO, Ogbuagu U, Adeniji AR, Agunbiade AP, Airaodion EO. Hepatoprotective effect of *Parkia biglobosa* on acute ethanol-induced oxidative stress in Wistar rats. International Research Journal of Gastroenterology and Hepatology. 2019;2(1):1-11.
25. Ogbuagu EO, Airaodion AI, Ogbuagu U, Airaodion EO. Prophylactic propensity of methanolic extract of *Vernonia amygdalina* leaves against acute ethanol-induced oxidative stress in Wistar rats. International Journal of Bio-Science and Bio-Technology. 2019;11(7):37-46.
26. Airaodion AI, Ogbuagu EO, Ekenjoku JA, Ogbuagu U, Airaodion EO. Therapeutic effect of methanolic extract of *telfairia occidentalis* leaves against acute ethanol-induced oxidative stress in Wistar rats. International Journal of Bio-Science and Bio-Technology. 2019;11(7):179-189.
27. Airaodion AI, Ene AC, Ogbuagu EO, Okoroukwu VN, Ekenjoku JA, Ogbuagu U. Biochemical changes associated with consumption (by rats) of “garri” processed by traditional and instant mechanical methods. Asian Journal of Biochemistry, Genetics and Molecular Biology. 2019; 2(4):1-11.
28. Ogbuagu EO, Airaodion AI, Okoroukwu VN, Ogbuagu U, Ekenjoku JA. Effect of Monosodium Glutamate on Body Weight and Alanine Aminotransferase Activity in Wistar Rats. International Research Journal of Gastroenterology and Hepatology. 2019c;2(2):1-8.
29. Airaodion AI, Ngwogu AC, Ekenjoku JA, Ngwogu KO. Hepatoprotective potency of ethanolic extract of *Garcinia kola* (heckel) seed against acute ethanol induced oxidative stress in wistar rats. International Research Journal of Gastroenterology and Hepatology. 2020;3(2):1-10
30. Akanji MA, Olagoke OA, Oloyede OB. Effect of chronic consumption of metabisulphite on the integrity of the rat kidney cellular system. Toxicology. 1993;81:173-179.
31. Bashir L, Shittu OK, Busari MB, Sani S, Aisha MI. Safety evaluation of giant African land snails (*Archachatina maginata*) haemolymph on hematological and biochemical parameters of Albino rats.J Adv Med Pharm Sci. 2015;3(3):122-130.
32. Shittu OK, Lawal B, Alozieuwa BU, Haruna GM, Abubakar AN, Berinyuy EB. Alteration in biochemical indices following chronic administration of methanolic extract of Nigeria bee *Propolis* in Wistar rats. Asian Pac J Trop Dis. 2015;5(8):654-657.
33. Oberley LW. Free radicals and diabetes. Free Radic Biol Med. 1998;5:113–124.
34. Pinto MS, Ranilla LG, Apostolidis E, Lajolo FM, Genovese MI, Shetty K. Evaluation of antihyperglycemia and antihypertension potential of native Peruvian fruits using *In vitro* models. J Med Food.2009;12:278–291.
35. Shim YJ, Doo HK, Ahn SY, Kim YS, Seong JK, Park IS, Min BH. Inhibitory effect of aqueous extract from the gall of *Rhus chinensis* on α-glucosidase activity and postprandial blood glucose. Journal of Ethnopharmacology. 2003;85, 283–287.
36. Nwadi OMM, Uchegbu N, Okonkwo TM. Effect of processing methods on the anti-nutrient reduction of Bambara groundnut: A review. Sky Journal of Food Science. 2019;7(3):037–041
37. Ogbuagu EO, Unekwe PC, Airaodion AI, Nweke IN, Ogbuagu U. Hepatotoxic effect

of *Xylopia aethiopica* fruit in Wistar rats. International Research Journal of Gastroenterology and Hepatology. 2021;4(1):1-16.
38. Sosnowska D, Podsędek A, Redzynia M, Zyzelewicz D. Effects of Fruit Extracts on Pancreatic Lipase Activity in Lipid Emulsions. Plant Foods Hum Nutr. 2015;70(3):344-550.
39. Ogbuagu EO, Airaodion AI, Ogbuagu U, Nweke IN, Unekwe PC. Nephrotoxicity of ethanol extract of *Xylopia aethiopica* fruit in Wistar rats. International Journal of Advances in Nephrology Research. 2021;4(1):1-16.
40. Shapiro SM. Bilirubin toxicity in the developing nervous system. Pediatr neurol. 2003;29:410-421.
41. Airaodion AI, Ngwogu KO, Ngwogu AC, Megwas AU, Ekenjoku JA. Nephrotoxicity of monosodium glutamate (MSG) in Wistar rats. International Journal of Advances in Nephrology Research. 2020;3(1):1-10.
42. Perlstein TS, Pande RL, Creager MA, Weuve J, Beckman JA. Serum total bilirubin level, prevalent stroke, and stroke outcomes: NHANES 1999-2004. American Journal of Medicine. 2008; 121:781–788.



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