academicJournals

Vol. 11(37), pp. 470-474, 8 October, 2017
DOI: 10.5897/AJPP2017.4839
Article Number: 7D2BB3466274
ISSN 1996-0816

http://www.academicjournals.org/AJPP

African Journal of Pharmacy and Pharmacology

Full Length Research Paper

Evaluation of some metabolic activities and immuno-stimulatory potential of methanolic seed extract of *Garcinia kola* (Heckel) in female albino Wistar rats

B. B. Bukar[1*], D. W. Dayom[2], O. S. Okpeke[1], L. Ior[1], K. D. Falang[1] and M. O. Uguru[1]

[1]Department of Pharmacology and Toxicology, University of Jos, P. M. B 2084, Jos, Plateau State, Nigeria.
[2]Clinical Pharmacy and Pharmacy Practice, University of Jos, P. M. B 2084, Jos, Plateau State, Nigeria.

Received 22 August, 2017: Accepted 28 September, 2017

Evaluation of some metabolic activities and immuno-stimulatory potential of the methanolic seed extract of *Garcinia kola* (Heckel) in female albino Wistar rats was carried out using standard methods. Animals in treated groups were administered 125, 250 and 500 mg/kg body weight, respectively of the extract through the oral route for 21 days. The control group received normal saline (0.5 ml/100 g body weight) for the same period during which food and water intakes were monitored. At the end of the treatment period, complete and differential blood counts were evaluated for each group. Results show that the extract at the doses of 125 and 250 mg/kg caused significant increase in body weight of treated animals compared with those of control ($p < 0.05$). Similarly, there was significant increase in food and water intake as well as faecal and urine output in groups treated with 250 and 500 mg/kg ($p < 0.05$). Results of the complete blood count showed that the extract caused significant increase in total white blood cell count (WBC) ($p < 0.05$) but a non-significant increase in haemoglobin ($p > 0.05$). There was significant increase in lymphocytes in groups that received the extract at a dose of 125 mg/kg compared with control ($p < 0.05$). These results suggest that the methanolic seed extract of *G. kola* possesses immuno-stimulatory potential that may justify the ethno-medicinal claims of the efficacy of *G. kola* in treatment of some immune disorders.

Key words: *Garcinia kola,* haematology, methanol extract, food/water, ethno-medicinal immuno-stimulatory.

INTRODUCTION

Garcinia kola (Heckel) is a tropical forest tree that grows between 15 and 17 m high (Burkill, 1985). The fruits and seeds are edible but have strong bitter taste (Ehiem and Eke, 2014). The seeds have many social and medicinal uses in traditional settings of many communities in the West and Central African sub-region (Adesuyi et al., 2012). It is traditionally used for the management of different ailments in such communities. Various scientific investigations have been carried out to confirm or refute such traditional claims. Interestingly, it has been found

*Corresponding author. E-mail: bukata33@yahoo.com. Tel: +2348036183979.

Table 1. Effect of *Garcinia kola* extract on some metabolic parameters in female albino rats.

Treatment	Food Intake (g)	Water Intake (ml)	Body Weight (g)	Faecal Output (g)	Urine Output (ml)
Control	38.5 ± 7.2	44.6 ± 5.3	121.4 ± 4.8	30.7 ± 3.1	18.9 ± 1.8
G. kola (125 mg/kg)	46.3 ± 2.3*	53.2 ± 4.8*	128.3 ± 2.5*	28.8 ± 4.7^{+}	15.3 ± 3.6^{+}
G. kola (250 mg/kg)	44.8 ± 5.1*	51.1 ± 6.2*	130.6 ± 3.1*	31.3 ± 4.2^{+}	12.6 ± 4.9*
G. *kola* (500 mg/kg)	40.6 ± 4.6^{+}	41.8 ± 3.3^{+}	122.1 ± 4.4^{+}	33.6 ± 5.1*	15.5 ± 1.6^{+}

Values are mean ± SEM; *p < 0.05; n = 5; control = normal saline; 0.5 ml/100 g.

to exhibit effects such as hepato-protective (Iwu, 1985; Usifo et al., 2012; Adaramoye et al., 2005; Udenze et al., 2012; Galam et al., 2013), aphrodisiac (Ralebona et al., 2012; Bukar et al., 2016), antioxidant (Esomonu et al., 2005), hypoglycaemic (Iwu et al., 1990), antimicrobial (Adegboye et al., 2008; Gabriel et al., 2011; Jackie et al., 2014; Abah et al., 2014), analgesic, anti-inflammatory and antipyretic (Olaleye et al., 2000) properties among others.

This study evaluated the immuno-stimulatory potential of the methanolic seed extract of *G. kola* in female albino rats. This is premised on some recent ethno-medicinal claims that the seed alone or in combination with other herbs may be beneficial in the management of some disorders of the immune system such as those arising from human immune deficiency virus (HIV) and Ebola.

MATERIALS AND METHODS

Animals

Twenty five female albino rats weighing between 115 and 123 g were purchased from the Animal House Unit of the Department of Pharmacology, University of Jos following approval by the Committee on use of experimental animals. The animals were handled under ethical conditions and guidelines for the use and care of laboratory animals (NIH, 2011). They were housed in metal cages padded with sawdust placed on metal shelves in a well-ventilated room at controlled ambient temperature. The animals had access to standard solid nutritional pellets and water *ad libitum.*

Preparation of extract

G. kola seeds were purchased from Forestry Research Institute of Nigeria (FRIN), Ibadan, Oyo state, Nigeria and re-authenticated at the Federal College of Forestry, Jos in Plateau State. A herbarium voucher specimen (number FHJ 202) was prepared. The seeds were washed, de-husked and carefully cut into small pieces with a clean sharp knife to enhance drying, then dried under shade in the laboratory.

Thereafter, they were ground to powder with mortar and pestle and extracted as described by Adaramoye (2010). In the procedure, 150 g of the powdered seed was extracted continuously with methanol (99.5%) in a Soxhlet extractor for 72 h. The extract was evaporated to dryness in a vacuum evaporator maintained at 40°C until a constant yield of 73.7 g (49.13%) following repeated weighing was obtained. The brownish colored extract was stored in an airtight container in the refrigerator and reconstituted in distilled water for the purpose of the experiment.

Sub-chronic administration of *Garcinia kola*

The rats were divided into 4 groups of 5 rats each. Group I (control) was administered normal saline (0.5 ml/100 g body weight) while groups II, III and IV were administered the extract in daily oral doses of 125, 250 and 500 mg/kg body weight, respectively for 21 days. Concentrations of the extracts were prepared such that final volumes used for each administration of the extract were not more than 1 ml. On day 22, animals were sacrificed by exsanguination and the blood collected into EDTA bottles for haematological evaluations.

Food and water intake

The quantity of food and water intake per day together with the amount of faecal and urine outputs were measured throughout the 21 days of extract administration. The body weights of the rats were also determined weekly. The mean food/water intake and faecal/urine output were each calculated at the end of the treatment.

Complete blood count

At the end of the three weeks the rats were anaesthetized with phenobarbitone (60 mg/kg) and their blood collected from the femoral vein into EDTA bottles for determination of haemoglobin (Hb), packed cell volume (PCV), total white blood cell (TWBC) and differential count of the white blood cells using Haematology Auto-Analyzer (Mindray BC 3200, Shenzen, China). Differential cell counts were done on a thin slide, prepared with a smearing blood sample using Wright-Giemsa's stain. The absolute number of each white cell type was calculated by multiplying the number of each cell counted (expressed as a decimal fraction) by the total white blood cell (TWBC) count.

RESULTS

Effect of *Garcinia kola* seed extract on some metabolic parameters

The effect of the extract on some metabolic parameters in female albino Wistar rats is shown in Table 1. The extract at the doses of 125 and 250 mg/kg caused significant increase in body weights of treated animals compared with those of control ($p < 0.05$). However, at the dose of 500 mg/kg, the increase in body weight of the treated animals was not significantly different from those of the control ($p > 0.05$). There was a significant increase in both food and water intakes in treated animals at

Table 2. Effect of *Garcinia kola* seed extract on complete blood in female albino rats.

Treatment	Hb (g/dl)	PCV (%)	TWBC $\times 10^9$/L	Platelets ($\times 10^3$/µl)
Control	16.20 ± 0.73	43.00 ± 1.26	11.06 ± 0.24	443.00 ± 46.74
G. kola (125 mg/kg)	16.80 ± 0.37$^+$	46.80 ± 1.01$^+$	16.88 ± 1.90*	577.60 ± 29.92*
G. kola (250 mg/kg)	16.60 ± 0.51$^+$	48.10 ± 1.53*	16.76 ± 1.92*	451.20 ± 95.27$^+$
G. *kola* (500 mg/kg)	16.50 ± 0.40$^+$	47.40 ± 0.68*	11.68 ± 0.65$^+$	417.80 ± 81.61$^+$

Values are Mean ± SEM; *$p < 0.05$; $^+p > 0.05$; n = 5; control = normal saline; 0.5 ml/100 g. Hb = Haemoglobin; PCV = packed cell volume; TWBC = total white blood cell.

Table 3. Effect of *Garcinia kola* seed extract on differential WBC counts in female albino rats.

Treatment	Lymphocytes (%)	Neutrophils (%)	Monocytes (%)	Eosinophils (%)
Control	73.80 ± 0.92	19.30 ± 1.01	5.00 ± 0.00	1.40 ± 0.24
G. kola (125 mg/kg)	76.90 ± 0.58*	16.40 ± 0.51*	5.00 ± 0.00	1.80 ± 0.20$^+$
G. kola (250 mg/kg)	71.40 ± 1.17$^+$	22.00 ± 0.95$^+$	5.00 ± 0.00	1.60 ± 0.24$^+$
G. *kola* (500 mg/kg)	71.20 ± 1.62$^+$	21.60 ± 1.21$^+$	5.60 ± 0.40	1.60 ± 0.42$^+$

Values are Mean ± SEM; *$p < 0.05$; $^+p > 0.05$; n = 5; control = normal saline, 0.5 ml/100 g.

doses of 250 and 500 mg/kg ($p < 0.05$). However, while there was a non-significant increase ($p > 0.05$) in food intake at the dose of 500 mg/kg, there was non-significant decrease ($p > 0.05$) in water intake with the same dose.

The extract caused increase in faecal output at doses of 250 and 500 mg/kg. However, while the increase was significant at 500 mg/kg ($p < 0.05$), it was not so at 250 mg/kg ($p > 0.05$). On the other hand, the extract at the dose of 125 mg/kg caused decrease in faecal output but this was not significantly different compared with control ($p > 0.05$). The extract generally caused decrease in urine output. The decrease in urine output was significant at the dose of 250 mg/kg ($p < 0.05$) but not significant at 125 and 500 mg/kg ($p > 0.05$) compared with control.

Effect of *Garcinia kola* seed extract on full blood count

The results indicate that the extract increased haemoglobin level of treated animals at all the dose levels (Table 2). However, this was found not to be significantly different compared with the control ($p > 0.05$). The result of the effect of the extract on packed cell volume showed that the extract caused increase in PCV of animals in all treated groups. However, while this was observed to be significant with animals in the groups that received 250 and 500 mg/kg ($p < 0.05$), it was not significant with those in the group that received 125 mg/kg ($p > 0.05$). There was significant increase in total white blood cell count (Table 2) of animals in groups treated with 125 and 250 mg/kg of the extract ($p < 0.05$) while in the group treated with 500 mg/kg, the increase was not significant compared with the +control ($p > 0.05$). The results showed that the extract caused increase in platelet counts at all the dose levels. The increase was particularly significant at the dose of 125 mg/kg ($p < 0.05$) while at higher doses of 250 and 500 mg/kg, the increase was not significantly different compared with control ($p > 0.05$).

The extract significantly ($p < 0.05$) increased lymphocytes count at the dose of 125 mg/kg compared with control (Table 3). However, at the higher doses of 250 and 500 mg/kg, the extract caused non-significant ($p > 0.05$) decreases in lymphocyte counts compared with the control. The result for the neutrophil count showed that the extract caused significant decrease in count in the group treated with 125 mg/kg compared with control ($p < 0.05$). However, at the higher doses of 250 and 500 mg/kg the extract caused non-significant increase in count compared with control ($p > 0.05$). The extract increased eosinophils count of animals at all doses used in this experiment. The results for the effect of the extract on monocyte count indicated that the extract had no effect on the count, except for the non-increase at the highest dose.

DISCUSSION

The methanolic seed extract of *G. kola* caused increase in body weight of treated animals. This observation is consistent with the increase in food intake in treated groups. Similarly, the extract increased water intake at the dose of 125 and 250 mg/kg but a non-significant decrease at the dose of 500 mg/kg. There was increased faecal output groups treated with 250 and 500 mg/kg but

a decrease output at the lower dose of 125 mg/kg. Urine output was observed to decrease in all groups of rats treated with the extract.

Various reports have been established to show the fact that food and water consumption in the rat is coordinated by the hypothalamus (Strominger, 1947; Bruce and Kennedy, 1951). Nonetheless, there is significant interplay between neural and chemical processes in regulation of food and water intake. This is because neural mechanisms are apparently mediated by chemical regulators, especially orexin, the stimulatory hormone for appetite (Norton et al., 1993; Ida et al., 1999; Baird et al., 2000). Nutrition has been noted as playing an important role in stimulating and modulating the immune functions. This is because the immune system needs adequate supply of nutrients to function properly. A variety of mechanisms have been highlighted on the interaction between the immune system and some foods and food components in terms of modulation of the immune functions (El-Gamal et al., 2011). The effect of the extract on the metabolic behavior of treated rats perhaps suggests its immuno-stimulatory potential in the rat model which could justify its traditional use in some immune disorders.

There was a non-significant increase in haemoglobin levels in all treated groups but a significant increase in packed cell volume at 250 and 500 mg/kg. Atsukwei et al. (2015) reported a similar finding in ethanolic seed extract of *G. kola* in male wistar rats. This supports the relationship between haemoglobin and packed cell volume and suggests that the extract may improve erythropoiesis through mechanisms yet to be determined (Smith and Bidlack, 1980; Naidu, 2003). The extract increased WBC, lymphocytes, neutrophils and platelet counts which were particularly significant ($p < 0.05$) at the dose of 125 mg/kg. Given the roles of these cellular components in immune function, the results suggest an immuno-stimulatory potential of the extract in the rat model. This observation agrees with the findings of Nworu et al. (2007, 2008) who used the mice model. Findings from this evaluation suggest that the methanolic seed extract of *G. kola* possesses immuno-stimulatory potential that authenticates its claimed ethno-medicinal use in management of some immune disorders.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENTS

We are greatly indebted to Messrs. Luka Wazoh, the Unit Head, and Solomon Tsaku, Senior Technical Assistant, Animal House, University of Jos for their assistance in taking care of the animals during the daily administration of the extract. We are also grateful to Mr. Luka Dabum of Physiology Department, University of Jos, for some of the haematological analysis and to Topsy Computers, Murtala Mohammed Way, Jos, for their clerical assistance.

REFERENCES

Abah AA, Abeglusi GA, Odukoya OA, Aya-Badejo PO, Adefuye-Ositelu AO, Adebisi KE (2014). *Garcinia kola* as antifungal therapy for oral candida infection-a comparative study. IOSR J. Dental. Med. Sci. 13:20-24.

Adaramoye AD (2010). Protective effect of *kolavirone,* a biflavonoid from *Garcinia kola* seeds in brainof wistar albino rats exposed to γ-radiation. Biol. Pharm. Bull. 33(2):260-266.

Adaramoye DA, Nwaneni VD, Anyanwo KC, Faromi EO, Emrok GO (2005). Possible antiatherogenic effect of kolavirone (a *Garcinia kola* seed extract) in hypercholesterolemic rats. Clin. Exp. Pharmacol. Physiol. 32:40-46.

Adegboye MF, Akinpelu DA, Okoh A (2008). The bioactive and phytochemical properties of *Garcinia kola* seed extract on some pathogens. Afr. J. Biotechnol. 7(21):3934-3938.

Adesuyi AO, Elum IK, Adaramola FB, Nwokocha AGM (2012). Nutritional and phytochemical screening of *Garcinia kola*. Adv. J. Food. Sci. Technol. 4(1):9-12.

Atsukwei D, Daniel EE, Odeh OS, Adams DM, Malgwi SI, Olih CP (2015). Efficacy of *Garcinia kola* seed ethanolic extract on haematological parameters in male wistar rats. J. Adv. Med. Pharm. Sci. 3(1):10-23.

Baird JP, Choe A, Loveland JL, Beck J, Mahoney CE, Lord JS, Grigg LA (2009). Orexin-a hyperphagia: Hindbrain participation in consummatory feeding responses. Endocrinology 150(3):1202-1206.

Bruce HM, Kennedy GC (1951). The central nervous control of food and water intake. Proc. Roy. Soc. B Biol. Sci. 138 (893):528-544.

Bukar BB, Uguru MO, Daniel J, Wannang NN, Dayom DW, Omale S, Okwuasaba FK (2016). Penile erectile properties and elemental analyses of the methanolic seed extract of *Garcinia kola* in some experimental animals. IOSR J. Pharm. 6(2):63-71.

Burkill HM (1985). *The useful plants of West-Tropical Africa*. 2nd Edition. Kew, London: Royal Botanical Gardens. pp. 505-615.

Ehiem JC, Eke AB (2014). Determination of drying characteristics and kinetics of bitter kola (*Garcinia kola*) using Page's model. Agric. Eng. Int. J. 16(4):278-284.

El-Gamal YM, Elmasry OA, El-Ghoneimy DH, Soliman IM (2011). Immunomodulatory effects of food. Egypt. J. Paediatr. Allergy. Immunol. 9(1):3-13.

Esomonu UG, El-Taalu AB, Anuka JA, Ndodo ND, Salim MA, Atiku MK (2005). Effect of ingestion of ethanol extract of *Garcinia kola* seed on erythrocytes in Wistar rats. Nigerian J. Physiolog. Sci. 20(1):30-32.

Gabriel FI, Emmanuel OO (2011). Pharmacological evaluation of *Garcinia kola* nut for antimicrobial activity. Int. J. Pharm. Biosci. 2(2):264-269.

Galam ZN, Gambo MI, Habeeb AA, Shugaba IA (2013). The effect of *Garcinia kola* seed on the liver histology. J. Nat. Sci. Res. 3(1):81-87.

Ida T, Nakahera K, Katayama T, Murakami N, Nakazato M (1999). Effect of lateral cerebroventricular injection of the appetite-stimulating neuropeptides, orexin and neuropeptide Y on the various behavioural activities of rats. Brain Res. 821(2):526-529.

Iwu MM (1985). Antihepatotoxic constituents of *Garcinia kola* seeds. Experimentia 42:699-700.

Iwu MM, Igboko OA, Okunji CO, Tempesta MS (1990). Antidiabetic and aldose-reductase activity of biflavone of *Garcinia kola.* J. Pharm. Pharmacol. 42:290-292.

Jackie O, Swamy AT, Matuku CN (2014). Preliminary phytochemical and in-vitro control of selected pathogenic organisms by ethanolic extract of *Garcinia kola*. Int. J. Curr. Appl Sci. 3(4):183-196.

Naidu KA (2003). Vitamin C in human health and disease is still a mystery? An overview. Nutr. J. 2:7.

National Institute of Health (NIH) (2011). Guide for care and use of

laboratory animals, 8th edition. Washington DC, USA: The National Academic Press.
Norton P, Falsiglia G, Gist D (1993). Physiologic control of food intake by neural and chemical mechanisms. J. Am. Diet Ass. 93(4):450-454.
Nworu CS, Akah PA, Esimone CO, Okoli CO, Okoye FBC (2008). Immunomodulatory activities of kolavirone, a mixture of three related bioflavonoids of *Garcinia kola* (Heckel). Immunopharmacol. Immunotoxicol. 30(2):317-332.
Nworu CS, Akah PA, Okoli CO, Esimone CO, Okoye FBC (2007). The effect of methanolic seed extract of *Garcinia kola* on some specific and non-specific immune responses in mice. Int. J. Pharmacol. 3(4):347-351.
Olaleye SB, Farombi EO, Adewoye EA, Owoyele BV, Onasanwo SA, Elegbe RO (2000). Analgesic and anti-inflammatory effect of kolavirone (a *Garcinia kola* extract). Afr. J. Biomed. Res. 3(3):171-174.
Ralebona N, Sewani-Rusike CR, Nkeh-Chunyag BN (2012). Effect of ethanolic extract of *Garcinia kola* on sexual behavior and sperm parameters in male wistar rats. Afr. J. Pharm. Pharmacol. 6(14):1077-1082.
Smith CH, Bidlack WR (1980). Inter-relationship of dietary ascorbic acid and iron on the tissue distribution of ascorbic acid, iron and copper in female guinea pigs. J. Nutr. 110:1398-1408.
Strominger JL (1947). The relationship between water intake and food intake in normal rats with hypothalamic hyperphagia. Yale J. Biol. Med. 9:279-288.
Udenze CC, Braide BV, Okwesilieze NC, Akuodor CG, Odey MO (2012). The effects of gavage treatment with *Garcinia kola* seeds on biochemical markers of liver functionality in diabetic rats. Ann. Biol. Res. 3(9):4601-4608.
Usifo UC, Uwaifo U, Charles II, William AA, Kerry EA (2012). The effect of chronic ingestion of crude *Garcinia kola* on the histology of the liver. Eur. J. Exp. Biol. 2(2):404-409.