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RESEARCH PAPER

ASSESSMENT OF SOME HEPATIC ENZYME ACTIVITIES IN ADULT RABBITS CHRONICALLY FED CRUDE *GARCINIA KOLA*

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ABSTRACT

Therapeutic potentials of *Garcinia kola* (*G. kola*) have been extensively documented and several researches have asserted its protective uniqueness against liver disorders/diseases. It is the aim of this study to assess the level of some enzyme involved in liver cellular integrity in rabbits chronically fed *G. kola*. To achieve this objective, twenty-four rabbits of comparable weights were randomly divided into four groups; consisting of a control (group A) and test (group B: BT1, BT2 and BT3). For 6 weeks, the control rabbits were fed standard animal feed with water given *ad libitum*, while doses of reconstituted *G. kola* powder (1200, 1500 and 1800mg *G. kola per* kg body mass) were administered daily by gavage to the respective test groups B aside from feed and water. At the end of the experiment, a dose depended significant increased in serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) activities was observed. On the other hand, alkaline phosphatase (ALP) activity was significantly reduced compared to control. These results suggest that G. *kola* may have some biochemical influences on the liver with chronic usage and as such a need for regulation of it ingestion.

***Key words*: Garcinia kola, Chronic, Crude, Liver, Male, Rabbits.**

INTRODUCTION

It is a known fact that plants serve as sources of novel drug compounds and as such, medicines derived from plants have made large contributions to human health and well being (Iwu *et al.*, 1999). This was affirmed by Dada and Ikuerowo (2009) who stated that attention has shifted from synthetic drugs to natural plant products as the use of plant extracts for enhancing organ systems and body performance in man are now on the increase. According to WHO (1976), medicinal plants contain substances that can be used for therapeutic purposes, and added that medicinal plants have also served as precursors for the synthesis of useful drugs (WHO 1976). One of such plants is *Garcinia kola* (*G. kola*); commonly known as bitter kola in local parlance, but 'Adu' in Esan, 'Miji-goro' in Hausa, 'Akilu or Ugolo' by the Igbos and 'Orogbo' in Yoruba ethnic groups of Nigeria respectively.

Indeed, a number of experimental models have been documented on *G. kola* as an anti-inflammatory and hepatoprotective agent (Akintonwa and Essien, 1990; Olaleye et al., 2000; Farombi et al., 2000; Farombi et al., 2001). Antioxidant and scavenging properties *in vitro* and *in vivo* have been attributed to *G. kola* as well (Farombi et al, 2002a; Farombi et al., 2002b). Its hypoglycemic effects (Iwu et al., 1990b; Adaramoye and Adeyemi, 2006) and

antilipoperoxidative effect (Adaramoye *et al*., 2005; Ahumibe and Braide, 2009; Omage et al., 2011) have also been documented. In fact, Omage et al. (2011) reported its positive effect on superoxide dismutase activity. Along this line of thought, Mathew *et al*. (2007) suggests that it might possess natural antioxidants necessary for the protection against carbon tetrachloride induced free radical damage, since marked decrease in the levels of lipid peroxides was recorded in pretreated rats. In addition, several studies have reported satisfactory outcome on its antihepatotoxicity potentials (Akintonwa and Essien, 1990; Iwu, 1985, Braide, 1991; Adegoke *et al.*, 1998; Adaramoye and Akinloye, 2000; Farombi, 2000; Farombi *et al*, 2000; Muragesh et al., 2005) and reproductive significance (Braide et al., 2003; Akpantah et al., 2005).

Recall that the liver plays a major role in metabolism, storage, synthesis and detoxification (Anthea et al., 1993; Guyton and Hall, 2006). According to Fernandez-Checa and Kaplowitz, (2005), every drug is known to be associated with hepatotoxicity almost certainly due to its ability to generate free radicals and to cause disturbance in hepatocyte biochemistry. Hence, *G. kola* like other drugs is metabolized by the liver. Based on the reported hepato-protective potentials couple with reproductive power of *G. kola*, there is this assumption that it might be consumed in excess. If that be the case, Garcinia kola may expose the liver to a variety of diseases and disorders. The present study therefore, assesses the serum levels of hepatic emzymes with liver integrity significant in chronic ingestion of *G. kola*.

MATERIALS AND METHODS

Plant of study: The seeds of *G. kola* were obtained from a local market in Ireukpen, Ekpoma, Edo-Nigeria. The coat of the seeds were removed and subsequently cut into pieces to increase its surface area for sun-drying. Grinding of the dried pieces into fine powder followed this procedure and finally the resultant *G. kola* powder was measured using Electric Balance (Denver Company USA -200398. 1REV.CXP-3000). The measurement was done separately, and each weighed sample was packed in a marked plastic pack and stored in a dry glass bottle.

Animals: Twenty-four male adult rabbits of comparable weight purchased from Aduwawa cattle market, Benin City, were used for the study. The rabbits were randomly divided into four groups, namely, group A (control; n = 6), and B (test groups) [BT1, BT2 and BT3 (n =6 each)]. They were housed in separately labeled wooden cages and allowed acclimatization for a period of 10 days. Through out the duration of study, they had *ad libitum* access to water and standard laboratory animal feed from Bendel Feed and Flour Mill, Ewu, Edo State, Nigeria and grass supplementation. The cages were swept clean every morning and the animal's feet and head examined regularly for evidences of infection such as sore feet, sore mouth and discharges from their eyes and nose.

Treatment with plant material*:* The difference in feed composition between the control and test groups is that the test group was given the test material (*G. kola*). Test groups received by gavage graded concentrations of *G. kola* powder (suspended in distilled water) daily for Forty-two days. The *G. Kola* was reconstituted with distilled water to obtain suspensions of appropriate concentrations for oral administration. Three doses; 1200mg/kg, 1500mg/kg and 1800mg/kg of the reconstituted powdered *G. kola* were administered to the test groups T1, T2 and T3 respectively. These values are chosen based on comparable information from previous work (Ahumibe and Braide, 2009).

Sample collection and analysis: 24 hours after the last administration of the *G.kola*, blood samples was collected from each of the rabbits by means of a cardiac puncture using 5ml hypodermic syringe and needle under chloroform anaesthesia. Serum aspartate amino transferase (AST) and alanine amino transferase (ALT) activities for cellular liver integrity were estimated with the Randox reagent kit using 2, 4-dinitrophenylhydrazine substrate as described by Reitman and Frankel (1957). Alkaline phosphatase (ALP) activity for billary tract integrity was determined with the Randox reagent kit using the p-nitrophenylphosphate substrate as described by Bassey et al. (1946).

Data analysis: The mean ± standard deviation was determined and the one-way ANOVA statistical test was performed using SPSS version 17 soft ware. The value $P < 0.05$ was considered statistically significant.

RESULTS

The mean ± SD of the enzymes with liver cellular integrity significant are presented in table 1. Compared to the control (10.97±1.87iu/L), there was a dose depended increase in the level of serum alanine amino transferase (ALT) with ingestion of *G. kola*. Similarly, a dose depended increased were observed for the level of serum aspartate amino transferase (AST) in the test groups compared to the control (14.10±1.55iu/L). The observed increased in serum ALT and AST were significantly different ($p < 0.05$) as compared to the level for the control group. On the other

hand, alkaline phosphatase (ALP) presented a steady decrease in serum level with increased ingestion of *G. Kola*. The observed decrease in the test groups were statistically significant (P<0.05) compared to the control (26.17±1.77iu/L). See Fig 1 for summarized bar chart of the observed changes in serum enzymes level on ingestion of Garcinia kola.

Table 1: Serum levels of ALT, AST, and ALP in rabbits fed Garcinia kola compared to control

Parameter	Control group (A)	Test group (B)		
		T1	T2	T3
ALT (IU/L)	10.97±1.87[a]	14.23±1.28[b]	17.08±0.23[c]	16.85±1.68[c]
AST (IU/L)	14.10±1.55[a]	20.20±2.41[b]	23.67±1.48[c]	25.45±1.32[d]
ALP (IU/L)	26.17±1.77[a]	22.47±1.65[b]	20.50±1.48[c]	19.67±1.36[c]

Values are Mean ±SD; Mean in a row with different superscripts indicates significantly different (p≤0.05).

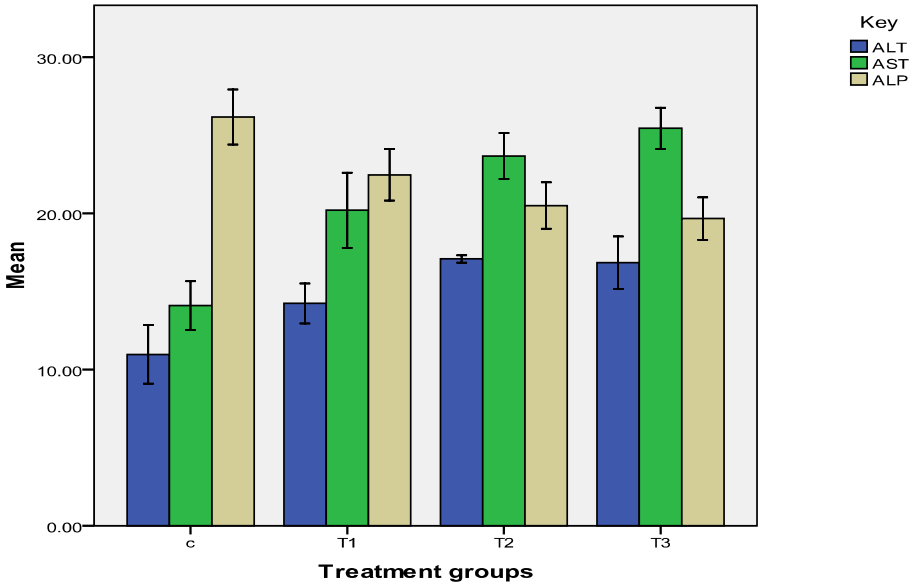

Fig 1: Summarized bar chart of enzymes of liver and biliary tract integrity.
Key: C= control, T = treatment, ALT = Alanine aminotransferase, AST = Aspartate aminotransferase, ALP = Alkaline phosphatase.

DISCUSSION

Judging by the results of this study, one can assert that in a dose dependent manner, chronic ingestion of *G. Kola* can induce alterations in the cellular integrity of the liver as well as the billary tract. Ironically however, the observed significant increase in ALT and AST levels and the corresponding decrease in the level of ALP, contradicts previous reports that *G. Kola* is hepato- protective (Akintonwa and Essien, 1990; Iwu, 1985, Braide, 1991; Adegoke *et al.*, 1998; Adaramoye and Akinloye, 2000; Farombi, 2000; Farombi *et al*, 2000).

On the contrary, our findings are in line with earlier reports by Braide and Grill (1990) that *G. kola* containing diets, induced alterations in the liver of rats fed with 10% *G. kola*. Similarly, Uko et al (2001) reported a dose-dependent decrease in rat's liver mass following *G. Kola* ingestion. Most recently, widespread vacuolar degeneration of hepatocytes, Kupffer cell proliferation, mild periportal fibrosis, multifocal centrilobular hepatocellular necrosis, mononuclear cell aggregations, cellular infiltration including neutrophils, lymphocytes and macrophages, were reported in the livers of dogs fed ethanolic extract of *G. Kola* (Nottidge et al., 2008).

Recall that in the assessment of liver damage, the serum concentrations of hepatic enzymes are important (Oze et al., 2010) and that these markers leak into the general circulation when there is necrosis or damage to the hepatic, neuronal or skeleton muscle cells (Murray et al., 2000); consequently their serum concentrations rise above normal values. Moreover, ALT and AST concentrations are normally known to be high in the liver and in a wide variety of tissues like the muscles, AST concentration is also high (Kaneko, 1980; Bush, 1991; Dial, 1995); though these markers are considered non-functional with negligible systemic concentrations (Murray et al., 2000).

Thus, the observed significant changes as shown by the result of this study, suggests that chronic ingestion of *G.kola* can induce alterations in the serum concentrations of ALT and AST and such elevations in experimental animals may reflect liver, heart and muscular toxicity. However, since ALT is known to be more specific for the liver tissue (Kaneko, 1980; Bush, 1991; Dial, 1995), the present findings suggest liver cells' toxicity with chronic doses of *G. kola*, while the increase in AST activity could be ascribed to cellular degeneration of myocardial, neuronal and liver cells.

Indeed, our results on the hepatotoxic nature of *G. kola*, appears to have challenged the hepato-protective status of *G. kola* as reported by Iwu (1985), Akintowa and Essien (1990) and Muragesh et al. (2005). Nevertheless, dosage as well as the experimental design may account for this sharp contrast as non-toxic-doses of *G. kola* were administered in the studies under comparison. Moreover, several studies have credited the potential of *G. Kola* to its constituents (Osifo et al., 2011) especially flavonoids (Madubuyi, 1995; Iwu et al., 1990a; Farombi et al., 2002a, b) which is known for its pharmacological and toxicological potentials (Elliott et al., 2000). It has also been documented that flavonoids induces morphological transformation of hamster embryo cells (Umezawa et al., 1977), inhibition of cellular glucose uptake and depletion cellular ATP content (Post and Varma, 1992), and several other effects on mammalian cells and organs (Elliott et al., 2000).

Therefore, excessive ingestion of G.kola may affect the cellular integrity of the liver and billary system in a dosage dependent manner. The mechanism of action is not known but may be attributed to the complex constituents of *G. kola* particularly flavoniods. In addition, the cells of the heart muscles and nerve are not excluded from this effect considering the significant changes in enzyme markers. Thus, to prevent avoidable consequences, moderate consumption of *G. kola* is recommended.

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AUTHORS' CONTRIBUTIONS

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