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RESEARCH PAPER

HEPATIC POTENTIALS OF *XYLOPIA AETHIOPICA* LEAVES IN ADULT WISTAR RATS

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ABSTRACT

This study investigates the possible toxic effect of *Xylopia aethiopica* on liver function via the evaluation of some serum biochemical parameters. The study involved 24 adult rats with weight ranging from 150 to 300g and divided into four groups of 6 rats each (A, B, C and D). For 21 days, varying doses of 1.2g/kg, 3.0g/kg and 6.0g/kg per body weight of *Xylopia aethiopica* leaves powder were administered daily to test groups B, C and D respectively. Group A however served as control that received normal feed and water only. At the end of the experiment, the animals were sacrificed under light anesthesia to obtain blood samples for the estimation of liver enzyme activities. The results showed that there was a significant increase ($P<0.05$) in the activities of Aspartate amino transferase (AST) and Alkaline Phosphates (ALP) but a significant decrease ($P<0.05$) in the levels of total protein, albumin and globulin levels in the test groups as compared to the control. On the other hand, the activities of Gamma glutamyl transferase (GGT) and Alanine amino transferase (ALT) of the test groups were not significantly different ($P>0.05$) from that of the control. Therefore, the observed changes in the parameters accessed, signifies the hepatotoxic impact of *Xylopia aethiopica* leaves.

Keywords: ***Xylopia aethiopica, hepatoxicity, Herbs, Nigeria.***

INTRODUCTION

Since ancient times, Africans, particularly those of the south Sahara, are known for the use of plants and plant extracts in the treatment and management of diseases (Woode *et al*., 2011). It was assumed to be the outcome of widespread poverty and illiteracy, which limits their accessibility to conventional medical services. Notwithstanding however, a large number of these tropical plants and their extracts have shown beneficial therapeutic effects such as contraceptive, aphrodisiac, fertility enhancing capacities, as well as anti-oxidant, anti-inflammatory, anti-cancer, and anti-microbial potentials (Raji *et al*., 2006). Also, evidence abound that plant derivatives have therapeutic potentials against vast human, animal and plant diseases (Ogbonnia *et al*., 2008), and thus, plants have become indispensable to human and animal existence. In fact, several conventional products derived from plants exists today and interestingly, many herbal products are now been approved by health agencies and organizations.

Amongst these plants with great therapeutic potential is *Xylopia aethiopica* which is commonly referred to as "African guinea pepper" or "Ethiopian pepper". It is an angiosperm of the Annonaceae family, and grows predominantly in humid forest zones of West Africa (Puri and Talata 1978; Woode *et al*., 2011). It is found all over the low land rain forest and most fringe forest in the savannah zones of Nigeria (Sofowara, 1978). Although it is said to serve as a 'pepper substitute' in Europe and India (Sofowara, 1978), it is highly valued in other countries for its medicinal and pharmacological properties (Okeke *et al*., 2008). The seeds have been reported to contain bitter principles like alkaloids, glycosides, saponnis, tannins, sterols, carbohydrate, protein, free fatty acids, mucilage's

and acidic compounds (Burkhill, 1985); some of which might be responsible for the documented medicinal and pharmacological properties of *Xylopia aethiopica.* According to Burkhill (1985) and Okeke *et al*. (2008), the fruit serves as spice, while its aqueous decoctions are used especially after child birth due to its antiseptic properties.

Despite the fact that all foods have therapeutic, nutritional or toxic effects qualities (Chike and Adienbo; 2010; Uzodike and Onuoha, 2010), *Xylopia aethiopica* abuse and mass-consumption, has been reported in populations (Riddle, 1992; Onyeyili, 2000; Hashemi *et al*., 2008). In fact, *Xylopia aethiopica* has been implicated in liver damage (Contran *et al*., 2005).

In addition, physiologically, the liver is involved in the homeostasic regulation of living system via its biochemical pathways that are necessary for growth, immunity and in the supplies of nutrients and energy (Ward and Daly, 1999). Therefore, the maintenance of a healthy liver is essential for the overall well being of an individual. Also, the fact that the liver is the central organ of metabolism and detoxification of drugs/toxins, implies that it is at a great risk of toxic damage (Bussieres and Habra, 1995). Hence, this study was undertaken to investigate the impact of *Xylopia aethiopica* on liver function as indicated by liver enzymes activity and protein levels using adult Wistar rat as a model.

MATERIALS AND METHODS

Experimental animals: Twenty four (24) Adult Albino Wistar rats with weights between 150 and 300g were procured from the animal farm of Anthonio Services Nigeria, Ekpoma, Edo State, and transferred to the experimental Laboratory of Anthonio Research Center, Ekpoma, Edo State, where they were allowed two (2) weeks acclimatization. They were kept in wire mesh cages with a tripod that separates the animal from urine and faeces in order to prevent contamination. During the period of acclimatization, the rats were fed with growers' mash and water was provided *ad libitum*. The animals were maintained and utilized in accordance with the standard guide for the care and use of Laboratory animals.

Animal grouping: The animals were assigned into four groups of 6 rats each (A, B, C and D). Group A served as the control, while groups B, C and D served as the test groups treated with graded doses of the *Xylopia aethiopica* leaves powder.

Substance Preparation: Fresh leaves of *Xylopia aethiopica* were collected from a natural habitat at Eke Village of Udi Local Government Area of Enugu State, Nigeria. It was identified and authenticated by a Botanist in the Department of Botany, Faculty of Natural Sciences, Ambrose Alli University, Ekpoma, Nigeria. The leaves were then spread on a dry table in a ventilated room with total absence of direct sunlight to air dry.

The 'dried leaves' was blended into fine powder using an electric blender (made in China) and the resultant fine powder was measured using an Electric Balance (Denver Company, USA, 200398. IREV.CXP-3000) and packaged in small plastic envelopes for storage pending usage.

For the purpose of this study, pastes were prepared by adding measured quantity of *Xylopia aethiopica* powder to feed (grower mesh) and mixed with sprinkles of water as described by Nwaopara *et al.,* (2011).

Experimental Procedure: The experimental groups B, C and D were administered 1.2g/kg; 3.0g/kg and 6.0g/kg of the measured *Xylopia aethiopica* leaves respectively. The respective *Xylopia aethiopica* doses were prepared by mixing with feed to add up to 100g. Group A received the normal feed (100g) and water only. The experiment proper (substance ingestion) lasted for a period of 21 days.

Sample Collection: At the end of the experiment, animals were sacrificed under chloroform anesthesia and blood samples were collected through the jugular vein using a hypodermic syringe (2ml) and transferred to an anticoagulant bottle (Lithium Heparin). The blood samples were separated at 3000 rpm using a centrifuge (10 minutes) to obtain plasma for the estimation of the biochemical parameters.

Estimation of Biochemical parameters: Estimation of the activities of ALT, AST, ALP and GGT was done using Randox Laboratory test kit (Antrim, UK). Specifically, ALT and AST activities were estimated using the method described by Reitman and Frankel (1957), while ALP and GGT were done using the methods described by Deutshe (1972) and Szasz (1969) respectively.

The estimation of Total protein and Albumin was done using Cromatest Laboratory test kit (Spain). Plasma Albumin (ALB) was determined by the method described by Doumas *et al*., (1971), while Plasma total protein (TP) was determined by the method described by Gomall *et al*., (1949). Globulin was then calculated using the formula described by Ochie and Kolhatkar (2000) as Serum Globulin = Total protein – Serum albumin (TP-ALB).

Statistical Analysis: Data obtained from biochemical estimations were expressed as mean ± SEM. Statistical significance was determined using one way analysis of variance (ANOVA, LSD) while values of $P<0.05$ was considered to be statistically significant.

RESULTS

Table 1 presents the impact of *Xylopia aethiopica* on liver enzyme activities and protein levels in adult Wistar rats. *Xylopia aethiopica* leaves ingestion was observed to bring about a dose dependent increase in liver enzyme activities. These increases were observed to be significant ($p<0.05$) in group C (35.12±2.42 IU/L) and D (59.62±1.76 IU/L) for AST and in group D (146.72±13.17 IU/L) for ALP. However, no significant ($p>0.05$) different was observed in the test groups for ALT and GGT as compared to control.

For liver proteins, it was observed that a 21 day ingestion of *Xylopia aethiopica* leaves induced a dose dependent reduction in Total Protein, Albumin and Globulin. Over all, significant increases ($p<0.05$) were observed in group C and D for Total Protein (16.00±0.68 g/dl and 11.15±0.43 g/dl respectively), Albumin (5.44±0.37g/dl and 3.30±0.33g/dl respectively) and Globulin (10.57±0.46g/dl and 7.85±0.67g/dl respectively) when compared to the values of the control.

Table 1: Liver Profile of rats fed graded doses of *Xylopia aethiopica* for 21 days

Parameters	Group A (Control)	Group B	Group C	Group D
AST (IU/L)	20.23±1.83[a]	26.33±5.30[a]	35.12±2.42 [b]	59.62±1.76[b]
ALP (IU/L)	84.68±18.29[a]	98.43±16.60[a]	133.92±18.51[a]	146.72±13.17[b]
ALT (IU/L)	107.08±8.50	118.13±14.81	173.53±26.59	185.90±36.52
GGT (IU/L)	3.35±0.45	3.45±0.31	3.63±0.46	4.60±0.45
Total Protein (g/dl)	24.91±1.53[a]	21.17±1.39[a]	16.00±0.68[b]	11.15±0.43[b]
Albumin (g/dl)	7.55±0.37[a]	6.32±0.33[a]	5.44±0.37[b]	3.30±0.33[b]
Globulin (g/dl)	17.36±1.49[a]	14.86±1.61[a]	10.57±0.46[b]	7.85±0.67[b]

The results are mean of six rats in each group ± SEM. Values in a row with a different superscript are significantly different from control at $p<0.05$.

DISCUSSION

Liver Enzymes are well known biomarkers for the prediction of liver toxicity (Gray and Howorth, 1982; Rahman *et al*., 2001) and as such, have been used in scientific reports. Available evidence show that damage to liver cells results in elevations of these enzymes in the serum (Wolf *et al*., 1972) and the measurement of enzyme activities is of clinical and toxicological significance in determining liver damage by toxicants or in diseased conditions (Singh *et al*., 2001).

Thus, the observed increase in the activities of AST, ALP, ALT, and GGT in the *Xylopia aethiopica* treated groups indicates that *Xylopia aethiopica* leaves have capacity to induced liver damage, while the observed dosage dependent alterations in enzyme activity imply that the toxic effect of *Xylopia aethiopica* increases with increase in dose.

Interestingly, the findings from this study are in line with the reports by Singh *et al*., (2001) and Navaro *et al*., (1993) that the administration of plant extract increases the activities of ALT and AST. Does it mean therefore, that all plants are toxic? On the contrary however, Aquaisua et al. (2011) reported that crude extracts of *Blighia unijugate* for example, have no toxic effects on both the kidney and liver of rats. On the other hand, Abdulrahman *et al.,* (2007) and Ogunka-Nnoko *et al.,* (2012) reported that extracts of *vitex doniana* and *Sorghum bicolor* to have toxic effects on both the kidney and liver of rats.

Although elevated levels of alkaline phophatase (ALP) have been associated with bone diseases, it is also an indicator for obstructive jaundice and intra-hepatic cholestasis (Adebayo *et al.*, 2010). Hence, the observed higher activities of the enzymes relative to control, suggests that *Xylopia aethiopica* can induce hepatic cell damage and/or other diseases like osteotoxicity.

Our findings are also in line with previous reports that Xylopic acid content of *Xylopia aethiopica* induces significant reduction in serum Total protein and Albumin (Abass, 2012) but disagrees with the report by Woode *et al*. (2011) that *Xylopia aethiopica* is hepatoprotective to several hepatotoxins. Of interest is the fact that the report by Woode *et al*., (2011) on the hepatoprotective effect of *Xylopia aethiopica*, were based on lower doses of 30 mg/kg, 100 mg/kg and 300mg/kg as against the relatively higher doses used in this study. The dissimilarities between the results of this study and that of Woode *et al*. (2011) may therefore hinge on the differences in dosage as dosage determines toxicity.

On the other hand, since albumin is known to serve as the binding and transport proteins for metal ions, bilirubin, and drugs (Guyton and Hall, 2006), its level therefore, reflects 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 (Rothschild *et al*., 1972). Thus, the significant reduction in serum proteins with the ingestion of *Xylopia aethiopica* leaves is suggestive of the fact that the leaves may have inhibited the synthetic function of the liver and as such hepatic toxicity. Moreover, the fact that globulin is produced in the liver and contributes to immune system (Merleb *et al*., 2007), a significant decrease in globulin concentration indicates therefore, that if the liver is exposed to the active contents by *Xylopia aethiopica* leaves, the immune system may also be compromised. In fact, the cytotoxic activity of xylopic acid has been reported in animal models and *in vitro* cell (Abass, 2012), while Adaramoye *et al*. (2011), reported that it increases the level of p21 and p53 gene transcripts.

Based on the findings of this study, therefore we opine that excessive and indiscriminate ingestion of *Xylopia aethiopica* leaf is toxic to the liver; hence, the need for caution.

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AUTHORS' CONTRIBUTIONS

Obodo B.N., Iweka F.K. and Obhakhan J.O. performed this study with assistance from Dada F.L., Festus O.O., Onoyovwi A.O., Maduagwuna G.N. and Okoye C.F. All authors were actively involved in the preparation and correction of this manuscript.