Original Article

The effect of different cassava (*Manihot utilissima*) components on liver function of male Wistar rats

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Abstract

Introduction: Liver is the main organ in vertebrates that is responsible for metabolism of substances ingested. Cassava is a rich source of carbohydrate that provides calories for many Nigerians. It is grossly deficient in protein, fat, some minerals and vitamins. The effect of consumption of the different cassava components on the liver function of Wistar rats was studied. **Material and Method:** Male rats (n=28) with weights between 165g-260g were randomly selected and assigned into three experimental groups and one control group of n=7 per group. The rats in the experimental groups were fed with normal rat chow with inclusion of 50% different cassava components thoroughly mixed with the feeds on a daily basis for eight weeks. The control group received equal amount of normal rat chow daily without the inclusion of any cassava components for the same period. All the rats had access to water ad libitum. The rats were sacrificed after 8 weeks of the experiment. Blood samples were collected for estimation of liver function. **Result and Discussion:** Average values of aspartate aminotransferase (AST), alanine amino transferase (ALT), alkaline phosphatise (ALP), total protein (TP), albumin (ALB), conjugated bilirubin (CB) and total bilirubin (TB) were recorded for each group and compared among the groups. It was observed that there were no statistically significant differences in the parameters investigated except the level of AST which increased in the experimental group fed with inclusion of 50% popo gari and starch in their diet but decreased in the other experimental group, conjugated bilirubin decreased in the experimental groups and the total bilirubin level decreased statistically in the experimental groups fed normal chow with inclusion of 50% popo gari and gari in their diet but no statistically significant difference in the group with inclusion of 50% starch in their diet when compared with the control group. **Conclusion:** This percentage used may not have adverse effect on liver function.

Key words: *Gari*, liver function, *popo gari*, starch

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INTRODUCTION

Cassava is a major food crop in Nigeria (FAO, 2001) which is strategically valued for its role in food security, poverty alleviation and as a source of raw materials for agro-allied industries in Nigeria with huge potential for the export market (Egesi, 2007). The presence of cyanogenic glycosides constitutes a major limitation to the use of cassava in both human and animal foods (Tonukari, 2004; Sanguanpong, 2003). Cassava tubers are traditionally processed by a wide range of methods which reduce their toxicity, improve pal ability and convert the perishable fresh root into stable products. This toxic agent present in cassava is a factor that can affect the proper functioning of the body systems depending on the percentage concentration (Gonzalez-Reimers *et al.,* 1994). Cyanide of dietary origin has been implicated in the etiology of various disease conditions (Tylleskar *et al.,* 1992; Banae, *et al.,* 1992; Rosling, 1987). Some abnormalities that border on pathological changes as well as metabolic integrity of the organism under repeated intakes of cassava with high cyanide content does occur. Known cases of acute cyanide poisoning from the consumption of cassava are rare, probably because of preparation process of cassava for consumption can destroy the linamarase and remove much of the free acid however, the possibility of chronic toxicity has not been eliminated (Osuntokun, 1970; Jaffiol *et al.,* 1991; Tylleskar *et al.,* 1992).

Liver is the main body organ responsible for metabolism of the substance ingested by vertebrates and contains enzymes capable of hydrolyzing absorbed linamarin to a metabolite which can dissociate to release cyanide ions (Maduagwu and Umoh, 1986). This subjects the liver to a variety of diseases and disorders. Detection of liver damage is associated with the presence of certain liver enzymes in the body such as aspartate aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase (ALP) which reside in the body and when the liver is injured, these enzymes spilt into the blood stream, raising the enzyme levels in blood and signaling liver damage. Therefore, increased serum enzyme level is proportional to the extent of tissue damage.

About 20% of cassava component, *gari* (combination of starch and fibers) fed to albino rats has been reported to cause glucose effect by lowering enzymes activity through reducing cyclic alkaline monophosphate concentration but an increase in mean enzymes concentrations and a decrease mean protein concentration were observed at 80% and 100% of the same cassava component fed to experimental animals (Braide *et al.,* 2011). Overall reduction in the activity of the succinate dehydrogenase and cytochrome C oxidase was observed in albino mice fed on maize (control) and *gari* based diet for 5 weeks (Ezeji *et al.,* 2009).

The aim of this study was to determine the effect of 50% inclusion of cassava components in rats' diet on the liver function.

MATERIALS AND METHODS

The rats were obtained and maintained in the animal house of the Department of Anatomy, School of Basic Medical Sciences, University of Benin, Benin City, Edo state, Nigeria. The animals were fed with grower's mash obtained from Edo feeds and Flour Mill Limited, Edo State, Nigeria.

A total of 28 adult male Wistar rats with weights ranging between 160 g and 265 g were randomly selected for the study and assigned into three experimental groups and a control group. There were seven rats in a group. The cassava components used were cassava fiber (*popo gari*), cassava starch and grated, roasted cassava tuber (*gari*) which were obtained from Ekiosa Market Benin-city, Nigeria. The rats in the experimental groups were given the normal rat chow with inclusion of 50% cassava components, respectively. The different cassava components were thoroughly mixed with the feeds and given on a daily basis for 8 weeks. The control group received equal amount of feeds daily without any cassava components added for the same period. All the rats were given water liberally. The rats were sacrificed; blood was drawn out through the aorta after dissecting the rat so as to get enough blood and collected in plain bottles for liver function analysis.

The estimation of total and conjugated bilirubin (CB) was done using Evelyn and Malloy method (Magos, 1960), AST and ALT estimation was done by monitoring the concentration of pyruvate hydrozone formed from 2, 4-dinitrophenylhydrazine (Babson *et al.,* 1966), ALP estimation was done using phenolphthalein monophosphate method (Henry *et al.,* 1974), total protein (TP) estimation was done using biuret method (Tietz, 1995) while estimation of albumin (ALB) was done using Bromocresol green method (Doumas *et al.,* 1971).

Statistical Analysis

Statistical analysis was done using analysis of variance, $P = 0.05$ or less were taken as significant and mean differences were compared using a multiple comparison test designed by Duncan in 1955. The mean and standard error of the mean for each of the groups of animals were calculated.

RESULTS

Table 1 and Figure 1 show the total bilirubin (TB) and the CB level in the control and experimental groups. The TB in the control group was 0.225 ± 0.13 mg/dl while that of the experimental groups with inclusion of 50% *popo gari*, *gari* and starch in their daily diets were 0.125 ± 0.03, 0.150 ± 0.03, 0.225 ± 0.06 mg/dl, respectively. It was observed that there was no statistically significant reduction at $P < 0.05$ in the experimental groups with the inclusion of 50% starch in their diet. However, there was a statistically significant decrease in TB in experimental groups with the inclusion of 50% *popo gari* and *gari,* respectively, in their diet when compared with that of the control group.

The CB level in the control group was 0.19 ± 0.04 mg/dL while that of the experimental groups with inclusion of 50% *popo gari*, *gari* and starch in their daily diets were 0.188 ± 0.04, 0.075 ± 0.01, 0.100 ± 0.04, 0.138 ± 0.04 mg/dL, respectively. The CB in the experimental groups decreased when compared with that of the control group which was statistically significant at $P < 0.05$.

Table 1 and Figure 2 show the AST, ALT, and ALP in the control and experimental groups.

Aspartate aminotransferase level in the control group was 131.25 ± 32.22 IU/L and in the experimental groups with inclusion of 50% *popo gari, gari* and starch in their diet were 153.75 ± 14.75 IU/L, 92.00 ± 19.14 IU/L and 107.75 ± 11.45 IU/L, respectively. Statistically significant increase was observed in the experimental groups with inclusion of 50% *popo gari*, while in the experimental group with inclusion of 50% *gari* and starch in their diet showed a statistically significant decrease when compared to the control group.

Alkaline phosphatase level in the control group was 14.00 ± 1.83 IU/L and in the experimental group, it was 12.50 ± 1.55 IU/L, 16.00 ± 2.55 IU/L and 14.20 ± 1.65 IU/L. There was no statistically significant difference between the level of ALP in the experimental groups and the control groups.

Alanine aminotransferase in the control group was 41.25 ± 4.85 IU/L and in the experimental groups with inclusion of 50% *popo gari, gari* and starch in their daily diet were 43.75 ± 3.07 IU/L, 37.75 ± 3.57 IU/L, and 29.25 ± 3.68 IU/L, respectively. There was no statistically significant difference between the control and the experimental groups when compared to the control group.

Table 1 and Figure 3 show the TP and ALB level in the control and experimental groups.

The TP level in the control group was 5.20 ± 0.07 g/dL and in the experimental groups with inclusion of 50% *popo gari*, *gari* and starch in their diet were 5.27 ± 0.05 g/dL, 4.53 ± 0.24 g/dL and 4.98 ± 0.05 g/dL; respectively. The group with the inclusion of 50% *gari* in their diet showed a statistically significant decrease while there was no statistically significant difference observed in the other group when compared to the control group.

Albumin level in the control group was 3.10 ± 0.11 g/dL and in the experimental groups with inclusion of 50% *popo gari*, *gari* and starch in their daily diet was 2.93 ± 0.10, 2.93 ± 0.09 and 2.90 ± 0.07 g/dL, respectively. This showed no statistically significant difference when compared to the control group.

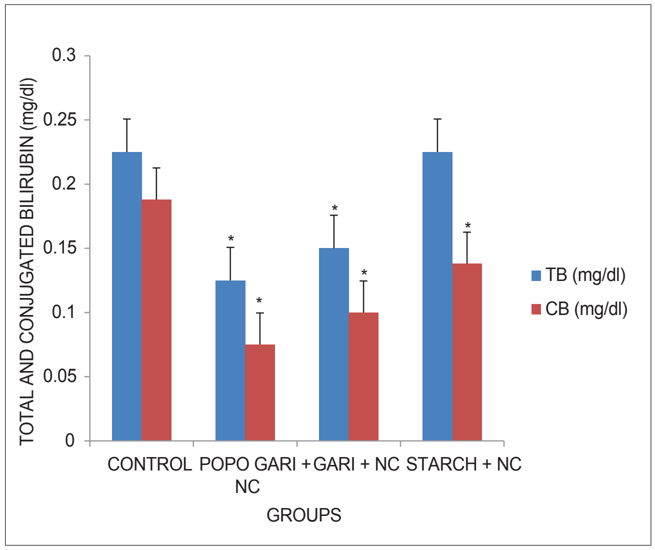

Figure 1: The value of total bilirubin and conjugated bilirubin levels in the control and experimental animals; *means statistically significant at $P < 0.05$

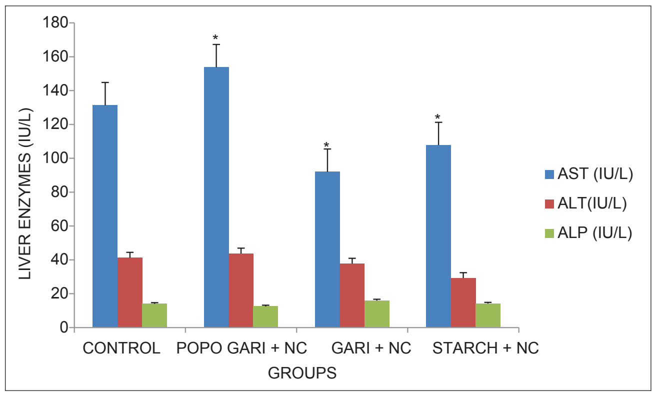

Figure 2: The value of liver enzymes levels in the control and experimental animals; *means statistically significant at $P < 0.05$

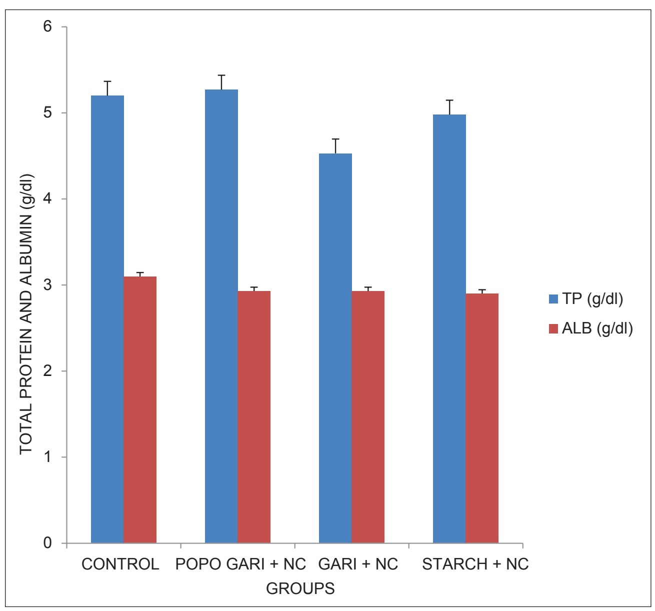

Figure 3: The value of total protein and albumin levels in the control and experimental animals; *means statistically significant at $P < 0.05$

Table 1: The values of TB, CB, liver enzymes, TP and ALB in both the control and experimental animals recorded in mean±SEM

Groups	TB (mg/dL)	CB (mg/dL)	AST (IU/L)	ALT (IU/L)	ALP (IU/L)	TP (g/dL)	ALB (g/dL)
Control	0.225±0.13	0.188±0.14	131.25±32.22	41.25±4.85	14.00±1.83	5.20±0.07	3.10±0.11
Popo gari+NC	0.125±0.03*	0.075±0.01*	153.75±14.75*	43.75±3.07	12.50±1.55	5.27±0.05	2.93±0.10
Gari+NC	0.150±0.03*	0.100±0.04*	92.00±19.14*	37.75±3.57	16.00±2.55	4.53±0.24	2.93±0.09
Starch+NC	0.225±0.06	0.138±0.04*	107.75±11.45*	29.25±3.68	14.20±1.65	4.98±−0.05	2.90±0.07

NC means Normal rat chow; *means statistically significant at $P<0.05$. SEM - Standard error of mean, TB - Total bilirubin, CB - Conjugated bilirubin, AST - Aspartate aminotransferase, ALT - Alanine aminotransferase, ALP - Alkaline phosphatase, TP - Total protein, ALB - Albumin

DISCUSSION

This study was designed primarily to investigate the effect of ingestion of various cassava components on the liver function. The effect of this diet on the TB, CB, AST, ALT, ALP, TP, and ALB serum level has been investigated. Scientists have reported that dietary composition could have marked effect upon cytochrome P450 mixed function oxidases and conjugation enzymes in animals (Bidlack *et al.,* 1986; Yang *et al.,* 1992). Glucose feeding in both man and microorganism causes profound changes in metabolism including inhibition of induction of several enzymes, stimulation of others, and blockage of glucocorticoids (Melvin and Goldberg, 1975).

Total bilirubin has been reported to be a potent physiologic antioxidant that may provide important protection against atherosclerosis, coronary artery, and inflammation (Yamaguchi *et al.,* 1996; Siow *et al.,* 1999; Stocker *et al.,* 1987), total serum bilirubin level concentrations is directly proportional to the protective factor high-density lipoprotein-cholesterol (Schwertner, *et al.,* 1994; Madhavan *et al.,* 1997). Decrease in TB and CB level in the experimental groups showed that there was no organ damage.

Increased mean enzymes concentrations and decreased mean proteins concentration at 80% and 100% *gari* diet fed to Albino rats (Braide *et al.,* 2011) which was not in consonance with the finding in this study. The difference in result may be due to the different percentage of cassava component used in the previous study which was 80% and 100% of the diet, while in this study, 50% was used. The level of TP in this study decreased, but was not statistically significant in the experimental group fed *gari* when compared to the control group. The percentage used has no effect on the liver enzymes except AST as reported by Porikos and Van Itallie in 1983 that diet has the effect upon hepatic enzymes both in animals and in healthy humans. ALP was lowered by reduced food intake (Tuba and Madsen, 1952; Jackson, 1952) and raised by dietary fat (Taylor *et al.,* 1952).

The decrease in AST level may be attributed to the fact that slowly digested carbohydrate diet gives a less rapid flow of glucose into the circulating system and a significantly lowered glucose tolerance curves with a bread diet than with a high sucrose diet (Cohen *et al.,* 2006). ALT is considered most reliable 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 (Johnson, 1995).

Increase in serum enzymes indicates cell membrane damage and some organ damage like liver (Joan *et al.,* 1987; Bogusz, 1975). It could either be by the cyanide ion or free radicals generated in the course of its metabolism or metabolism of chemical species within the system. Antioxidant status is affected as in the case of high cassava food intake and low protein food intake, the body tries to detoxify the ingested cyanide using sulfur containing amino acids of the body, protein synthesis is compromised (Okafor *et al.,* 2008). Therefore, increased AST level in the group with inclusion of 50% *popo gari* in their diet may be due to the increased level of cyanide present when compared to the other cassava components used caused by the difference in fermentation process during preparation.

SUMMARY AND CONCLUSION

Total bilirubin, CB, and AST showed statistically significant difference, but ALT, ALP, TP, and ALB showed no statistically significant difference in the experimental groups when compared to the control group.

This result is based on the 50% cassava components included in their diet and therefore can be concluded that the inclusion of this percentage of cassava component may not have an adverse effect on the liver function since most of the parameters showed no statistically significant difference. However, further study in this area is recommended using different percentage of cassava components.

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