Original Research Article

Effect of Consumption of Garri Processed by Traditional and Instant Mechanical Methods on Lipid Profile of Wistar Rats

ABSTRACT

Aim: This study focuses on the effect of consumption of garri processed by traditional and instant mechanical methods on the lipid profile of Wistar rats.

Methods: Cassava samples were obtained from the International Institute of Tropical Agriculture (IITA), Ibadan and were processed using traditional and instant mechanical methods. Fifteen adult male Wistar rats were purchased from the Animal Holding Unit of the Department of Physiology, University of Ibadan, Nigeria with body weight between 100 and 120 g. They were acclimatized for seven (7) days during which they were fed *ad* libitum with standard feed and drinking water. They were randomly divided into three groups of five rats each. The rats in group A were then fed with pure standard feed, those in group B were fed with garri processed by instant mechanical method while those in group C were fed with garri processed by traditional method. After twenty-eight days of feeding, the animals were fasted overnight and anaesthetized using diethyl ether. Blood samples were collected by cardiac puncture. Lipid profile was determined using standard methods.

Results: Garri samples significantly decreased weight gained by animals when compared with those in the control group. Lipid profile was also unhinged by both garri samples.

Conclusion: This study showed that garri processed by traditional method as well as those processed by instant mechanical method perturbed lipid profile of animals after 28 days of feeding. These perturbations were more severe in animals fed with garri processed by instant mechanical method, which was attributed to its cyanide and other anti-nutritional composition. Processors of garri should be enlightened on the dangers of garri high in cyanide to human health and should be encouraged to avoid short-cut practices in the production of garri. Garri should be allowed to ferment for at least 72 hours before roasting.

Keywords: *Cyanogenic glucoside, lipid profile, instant mechanically processed garri, traditionally processed garri, fermentation*

1. INTRODUCTION

Cassava (*Manihot esculenta* Cranz) is a woody shrub native to South America of the spurge family, Euphorbiaceae [1]. It is extensively cultivated as an annual crop in tropical and subtropical regions for its edible starchy tuberous root, a major source of carbohydrates [2]. Cassava is the third largest source of food carbohydrates in the tropics, after rice and maize. It is a major staple food in the developing world, providing a basic diet for over half a billion people [3]. It is one of the most drought-tolerant

crops, capable of growing on marginal soils. In 2014, global production of cassava root was 268 million tonnes, with Nigeria as the world's largest producer of nearly 55 million tonnes or 21% of the world total [4].

Cassava is classified as either sweet (*Manihot palmata*) or bitter (*Manihot esculenta* or *Manihot utilissima*). Like other roots and tubers, both bitter and sweet varieties of cassava contain anti-nutritional factors and toxins, with the bitter varieties containing much larger amounts [5]. It must be properly prepared before consumption, as improper preparation of cassava can leave enough residual cyanide to cause acute cyanide intoxication, goiters, and even ataxia, partial paralysis, or death [6]. The more toxic varieties of cassava are a fall-back resource (a "food security crop") in times of famine or food insecurity in some places. Farmers often prefer the bitter varieties because they deter pests, animals, and thieves [7].

In Nigeria, as in most African countries, cassava is one of the most important carbohydrate sources. About 95 percent of cassava is consumed as food and less than 5 percent of it is used for industrial purposes [8]. It is usually consumed in processed forms. In recent times, several processing options have emerged from cassava such as garri, fufu, starch, flour, tapioca and chips. Irrespective of these options, garri (roasted granules) and edible starch (which is a by-product from drying the grated tubers) have maintained an important position in the food timetable of many households in Nigeria and other countries of the world, although starch consumption is most notable in the south-south region of Nigeria [9].

Toxicity of cyanide in cassava products has been reported. There are as well few reported cases of death linked to consumption of cassava meals [10]. The incidents of cassava toxicity parallel severe hunger condition associated with drought or wartime when processors adopt “shortcut” (shortened process time) in order to meet market demand. Consumption of garri has always been a trend in Nigeria and some other parts of the world. The consumption of this product has been accompanied with some side effects like food poisoning and other related effects due to inefficiency in the course of production, which inevitably leads to improperly processed product, and when this product is consumed, it results to food poisoning and its effect can be fatal [10].

On 20th March 1994, one Mrs. Loveth Osueke was reported in the National dailies to have died after eating African Salad made from cassava which she bought from Ariaria market in Aba, Abia State, Nigeria. More recently, it was also reported in the national dailies on Tuesday 1st November, 2016 that six persons (including a mother, her three children and her two neighbours) died after consumption of cassava product (‘lafun’) in Ogaminana area of Okene in Kogi State, Nigeria [11].

Fermentation is an important processing technique for cassava, especially in Africa. Three major types of fermentation of cassava roots are recognized: the grated root fermentation, fermentation of roots under water and mould fermentation of roots in heaps [12]. The grated cassava roots are allowed to ferment in sacks for 3-7 days, which encourages lactic acid fermentation. The pH after 3 days decreases from 6 to 4 and the fermentation is dominated by lactic acid bacteria [5]. Grating is important for bringing linamarin into contact with linamarase allowing its hydrolysis to glucose and cyanohydrin and then to HCN [13]. The hydrolysis continues during the fermentation process. Lactic acid fermented products are reported to have significant concentrations of cyanohydrins because pH decreases during fermentation and cyanohydrin is stable at low pH. The processes of garri production reduce cyanogen contents by more than 95 % [12]. Fermentation of cassava roots under water, followed by sun drying, is reported to be the best for cyanogens removal [14]. This type of fermentation is used more in areas where there is a sufficient supply of water such as near a river or lake, and is common in countries such as Nigeria, Democratic Republic of Congo,

Tanzania and Malawi [12]. Heap fermented cassava root products are produced in Tanzania [15], Uganda and Mozambique [16]. The process involves peeling of cassava roots, sun drying for 1 to 3 days, heaping and covering, fermentation, scraping off the molds, crushing into crumbs, sun drying, pounding and sieving into flour. During the fermentation of the roots, the temperature inside the heaps increases between 23 and 29 ℃ higher than the temperature outside the heaps (2 to 12 ℃). According to Sani and Farahni, [16], heap fermentation is dominated by the *Neurospora sitophila, Geotrichum candidum* and *Rhizopus oryzae.* Heap fermentation of cassava roots followed by sun drying is capable of reducing the cyanogen levels by 95 % [16]. Airaodion et al. [17] have reported the nutritional and anti-nutritional composition of garri processed by traditional and mechanical methods. Renal and hepatotoxic effect of garri processed by traditional and mechanical methods has also been reported [18]. This study therefore, focuses on the effect of consumption of garri processed by traditional and instant mechanical methods on the lipid profile of Wistar rats.

2. MATERIALS AND METHODS

2.1. Production of Garri

Cassava samples were obtained from the International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria and were processed using traditional and instant mechanical methods. For garri processed by instant mechanical method, cassava was grated and dewatered using hydraulic press and was roasted (fried) within 24 hours of harvest (Fig. 1). For garri processed by traditional method, the grated cassava mash was allowed to stay for 24 hours in the sack before dewatering using sticks. The dewatering process took 3 days before roasting (Fig. 2). The two methods of processing were as described by Airaodion et al. [17].

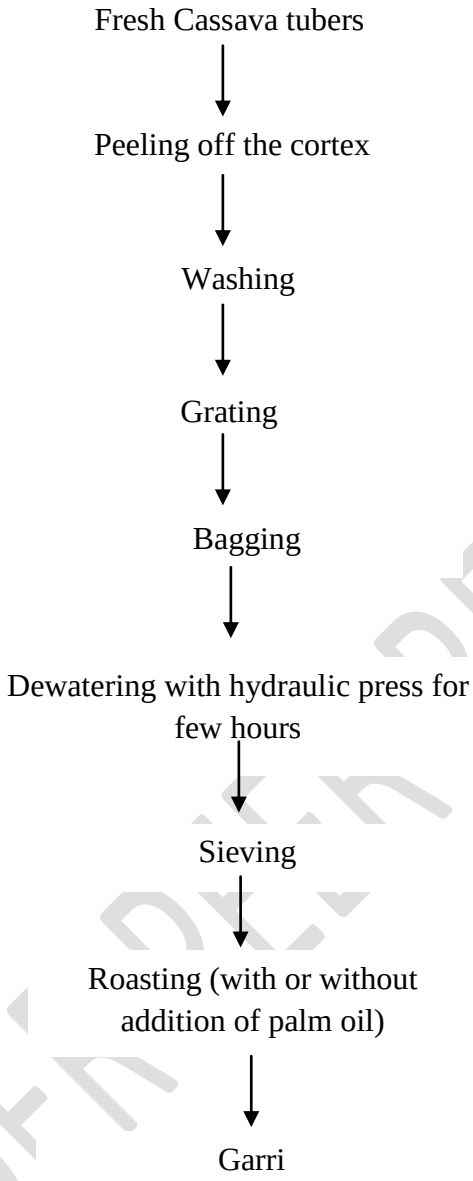

Fig. 1: Stages of processing Garri by Instant Mechanical Method [17]

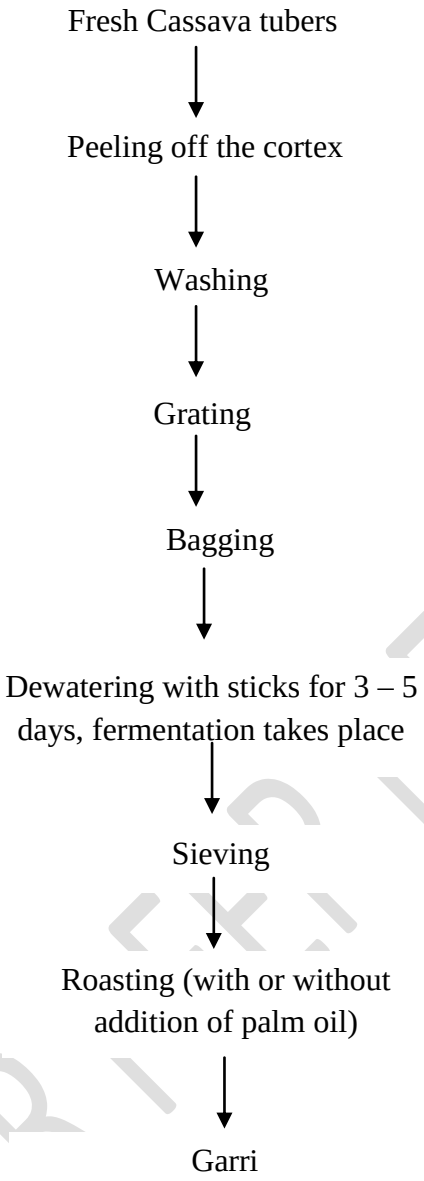

Fig. 2: Stages of processing garri by traditional method [17]

2.2 Experimental Design

Fifteen adult Wistar rats (*Rattus norvegicus*) were purchased from the Animal Holding Unit of the Department of Physiology, University of Ibadan, Nigeria with body weight between 100 and 120 g. They were housed in Imrat animal house, Ibadan. They were acclimatized for 7 days during which they were fed *ad* libitum with standard feed and drinking water. Throughout the experiment, the animals were housed in clean cages placed in well-ventilated housing conditions (under humid tropical conditions). The rats were randomly divided into three groups consisting of five rats in each group. Prior to the rats being fed with different garri samples, they were given known amount of standard feed and the remaining feed was weighed after 24 hours to ascertain the actual quantity of feed each group took. This was done prior to proper feeding and the average was computed. On the average, it was observed that each group sufficiently ate 150 g of mash feed meaning that each rat can take 30 g of meal per day. The rats in group A were then fed with 150 g of pure standard feed, rats in group B were fed with 150 g of Garri processed by instant mechanical method and the rats in group C were fed with 150 g of garri processed by traditional method. After 28 days of feeding, the animals were fasted overnight and anaesthetized using diethyl ether. Blood samples were collected by cardiac puncture. The animals were dissected through incision on the abdominal wall vertically and horizontally and the organs were harvested, washed in 10% formysaline and dried with tissue paper before weighing.

2.3 Determination of Lipids

Lipids were extracted and determined according to previously described methods [19,20].

2.4 Statistical Analysis

Data were subjected to analysis of variance using the Statistical Package for Social Sciences (SPSS), version 20.0. Results were presented as Mean ± Standard Error of the mean (SEM). 2–tailed t–test was used for comparison of the means. Differences between means was considered to be significant at $p<0.05$.

3. RESULTS

A significant decrease was observed in the weight gained by animals fed with garri samples when compared with those fed with standard animal feed (control group). However, no significant difference was observed in the weight gained by animals fed with garri processed by instant mechanical method when compared with those fed with garri processed by traditional method (Table 1). No significant difference was observed in the relative organ weight of animals fed with garri samples when compared with those fed with standard animal feed (control animals) as shown in table 2. Garri samples perturbed the lipid profile of animals (table 3). Animals fed with garri samples had significantly lower total cholesterol and high density lipoprotein content when compared with those fed with standard animal feed but only high density lipoprotein content was significantly different when animals fed with garri processed by instant mechanical method were compared with those fed with garri processed by traditional method. Conversely, a significant increase was observed in low density lipoprotein and triacylglycerol when animals fed with garri samples were compared with those fed with standard animal feed at $p<0.05$, and the difference was also significant when animals fed with garri processed by instant mechanical method were compared with those fed with garri processed by traditional methods. No significant difference was observed when the phospholipid content of animals fed with garri samples were compared with those fed with standard animal feed (control group) at $p<0.05$.

Table 1: The Effect of Garri Samples on Animal Body Weight after 28 days of Feeding

Group		Weight (g)	Weight Gain (g)
Control	Initial	111.21±7.26	86.20±2.05[a]
	Final	197.46±7.13	
Instant Mechanical Garri	Initial	112.22±8.26	67.64±5.59[b]
	Final	179.80±3.70	
Traditional Garri	Initial	112.62±6.29	65.00±6.04[b]
	Final	177.61±7.23	

Values are presented as Mean±SEM with n = 5.

Values with different superscripts along the same column are significantly different at $p<0.05$

Table 2: Effect of Garri Samples on Relative Organ Weight of Animals after 28 days of Feeding

Organs	Control	Instant Mechanical Garri	Traditional Garri
Heart (%)	0.36 ± 0.02^{a}	0.37 ± 0.02^{a}	0.36 ± 0.02^{a}
Liver (%)	5.12 ± 0.11^{a}	4.89 ± 0.14^{a}	5.07 ± 0.08^{a}
Spleen (%)	0.24 ± 0.02^{a}	0.23 ± 0.05^{a}	0.23 ± 0.04^{a}
Lung (%)	0.43 ± 0.04^{a}	0.39 ± 0.06^{a}	0.40 ± 0.05^{a}
Kidneys (%)	0.78 ± 0.06^{a}	0.81 ± 0.03^{a}	0.81 ± 0.04^{a}
Testes (%)	0.77 ± 0.05^{a}	0.80 ± 0.03^{a}	0.81 ± 0.02^{a}

Values are presented as Mean$\pm$SEM with n = 5.

Values with different superscripts along the same row are significantly different at $p<0.05$

Table 3: Effect of Garri Samples on the Lipid Profile of Animals after 28 days of Feeding

Lipids	Control	Instant Mechanical Garri	Traditional Garri
TC (mg/dL)	110.03 ± 6.82^{a}	103.90 ± 7.01^{b}	102.30 ± 3.37^{b}
HDL (mg/dL)	57.41 ± 6.73^{a}	30.91 ± 2.88^{b}	42.84 ± 3.81^{c}
LDL (mg/dL)	42.83 ± 3.26^{a}	60.98 ± 4.79^{b}	51.49 ± 3.72^{c}
HDL/LDL	1.34 ± 0.04^{a}	0.51 ± 0.01^{b}	0.83 ± 0.02^{c}
TC/HDL	1.92 ± 0.03^{a}	3.36 ± 0.02^{b}	2.39 ± 0.03^{c}
TC/LDL	2.57 ± 0.02^{a}	1.70 ± 0.00^{b}	1.99 ± 0.02^{b}
TAG (mg/dL)	91.49 ± 3.60^{a}	106.35 ± 6.72^{b}	116.11 ± 5.11^{c}
VLDL (mg/dL)	18.30 ± 0.91^{a}	21.27 ± 0.83^{ab}	23.22 ± 1.02^{b}

Values are presented as Mean$\pm$SEM with n = 5.

Values with different superscripts along the same row are significantly different at $p<0.05$

TC = Total Cholesterol, HDL = High Density Lipoprotein, LDL = Low Density Lipoprotein, TAG = Triacylglycerol, VLDL = Very Low Density Lipoprotein

4. DISCUSSION

Cassava (*Manihot esculenta Crantz)* and its products have been widely studied and reported to have beneficial effect, and sometimes with toxic effects [18]. Most of these effects are due to the presence of some non-nutrient factors in them. Cassava is a staple food in human diets in over 80 countries [17]. Starch, a product of cassava tubers is one of the most popular staple foods of the people of the rain forest belt of West Africa and contains mainly starch-20% amylase and 70% amylopectin having lost the soluble carbohydrates (i.e. glucose and sugar) during processing. Cassava can be processed into different products such as garri, tapioca, Africa salad, 'lafun' etc., the most common being garri. Cassava products are used as a primary staple food; careful processing to remove these toxic constituents is required to avoid chronic cyanide intoxication [18]. Airaodion et al. [17] has previously reported that garri processed instant mechanical method contained higher cyanogenic glucoside and other antinutrients when compared with garri processed by traditional method. This study therefore, focuses on the effect of consumption of garri processed by traditional and instant mechanical methods on the lipid profile of Wistar rats.

Result from this study revealed that there was a significant decrease in weight gain in the experimental animals when compared with those of control animals at $p<0.05$. The possible mechanism of action for this decrease in weight gain is not clearly understood, but it could be as a result of the presence of some anti-nutritional constituents that is present in the garri samples

thus inhibiting neurotransmitters that are involved in appetite which then cause a reduction in growth rate. This contradicts the work done by Eze et al. [21], in which there was no significant difference in the mean body weight, when animals were fed with methanolic extracts of garri. This indicates that the oral doses administered had no effect on the growth of rats as significant changes in body weights have been used as an indicator of adverse effects of drugs and chemicals. Nevertheless, the growth of an organism comprises many factors including physiological, biological and cellular processes [22].

Result from this study also revealed that there was no significant difference in the relative organ weight (heart, liver, spleen, lung, kidneys and testes) of experimental groups (groups fed with garri processed by tradition and instant mechanical methods) when compared with those of the control group respectively at $p<0.05$. This corresponds with the work done by Okafor et al. [23] who investigated the histological effect of cassava starch on the liver of adult Wistar rats. This is also similar to earlier report of Eze et al. [21], in which there was no significant difference in the mean organ weight (heart, liver, spleen, kidneys and testis), when fed with chloroform and methanol extracts of garri.

In this study, a significant decrease was observed in the concentration of total cholesterol in animals fed with garri processed by traditional and instant mechanical methods when compared with that of the control group at $p<0.05$. The decrease in cholesterol level is in agreement with what was reported by Oladunjoye et al. [24] where a low cholesterol level was observed in lye treated cassava peal meal. Cassava peel contains high hydrocyanic acid and the presence of hydrocyanic acid in cassava can also exert hypochlesteronic influence as glucosides have ability to interfere with the intestinal absorption of the dietary cholesterol and lipid [25]. Studies in the past have linked cassava dietary intake with a decrease in blood cholesterol level and this was said to be due to the presence of high dietary fibre in cassava [26]. The hypolipidemic effects of dietary fibre are mediated by the action of soluble fibre in binding bile acids, thereby increasing the faecal excretion and interrupting the enterohepatic circulation of bile salts [27]. The increased dietary fibre often results in reduction in availability of cholesterol for incorporation into lipoproteins [28]. High dietary fibre has been reported to increase excretion of cholesterol and thereby lowering blood cholesterol level [26].

In this study, it was observed that the concentration of triglycerides in garri processed by traditional method was significantly higher than those found in the groups treated by garri processed by mechanical method and the control group. This was similar to an increase in the level of triglyceride observed by Dhas et al. [29], in a study that investigated the effects of hydrogen cyanide exposure in cassava workers. Triglycerides may also increase in impaired thyroid function since hydrogen cyanide increases thyroid stimulating hormone (TSH) and decreases thyroxine (T_4) and triiodothyronine (T_3) resulting in abnormal thyroid function. Thus, the increase in triglyceride can also be due to impaired thyroid function [30,31].

5. CONCLUSION

This study showed that garri processed by traditional method as well as those processed by instant mechanical method perturbed lipid profile of animals after 28 days of feeding. These perturbations were more severe in animals fed with garri processed by instant mechanical method, which was attributed to its cyanide and other anti-nutritional composition. Processors of garri should be enlightened on the dangers of garri high in cyanide to human health and should be encouraged to avoid short-cut practices in the production of garri. Garri should be allowed to ferment for at least 72 hours before roasting.

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