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EFFECT OF FERMENTING CASSAVA WITH *RHIZOPUS ORYZAE* ON THE CHEMICAL COMPOSITION OF ITS FLOUR AND GARI PRODUCTS

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Abstract

Cassava (*Manihot esculenta* Crantz) pulp was fermented with pure strain of *Rhizopus oryzae*. The product was subsequently processed into gari and flour, the form in which it is popularly consumed in Nigeria. The nutritional evaluation of the flour and the gari shows that there was an increase in the protein content of the flour (97%) and the gari (53%) determined by the Kjeldahl method, while there was a general decrease in the carbohydrate level, [flour (5.0%), gari (10.4%)]. There was no considerable increase in the fat, ash and lipid. The mineral contents (Zn, Mg, Fe, Ca, Na and K) were also low except in the case of gari where some of the elements were considerably low.

The antinutrients, that is, tannin [flour 0.16mgTA/100g), gari (0.08mg/100g)] and total cyanide [pulp (17.21mg/kg), gari (14.85mg/Kg)] except phytate [flour(1497.18mg/100g), gari (912.26mg/100g)] contents of the fermented cassava products were considerably low. This implies that *Rhizopus oryzae*, a cheap and non-pathogenic fungi, can be used to increase the protein level and at the same time, decrease the tannin and cyanide content of cassava products.

INTRODUCTION

Cassava (*Manihot esculenta* Crantz) is of great economic importance in the economy of several tropical countries of Africa where its consumption (in terms of carbohydrate content) exceeds those of other root crops [1]. Though an important carbohydrate source with very low protein content, cassava can be processed into many food products such as gari, fufu and flour [2]. The major advantages it has over other root crops include ease of propagation, high yields, pest and drought resistance [3, 4]. However certain varieties contain large amounts of cyanogenic glucosides (linamarin and lotaustralin) which are hydrolysed to hydrocyanic acid (HCN) by the endogenous enzyme linamarase when the plant tissue is damaged during harvesting, processing or other mechanical processes [5].

There are a number of roles that microorganisms can play in cassava processing, either positive or negative. The positive effects are generally regarded as part of the fermentation process namely, product preservation, flavour development, cyanide elimination and changes in functional properties while the negative effects include spoilage of cassava products and contamination by pathogenic microorganisms. A recent report has shown that cassava fermentation with certain fungi increases the level of protein in cassava. Aspergillus niger fermentation of cassava increases its protein content from 2-3% to 18 - 20% [6] while Rhizopus oryzae fermentation of cassava flour increases its protein content up to 11% [2].

Gari (the form in which cassava is popularly consumed in Nigeria), a dehydrated storage cupboard staple food, consumed raw or cooked and obtained from roots of cassava, could be processed at the small, medium or industrial scale [7] . Since quality of the product is inextricably tied up with pre-processing, processing and post-processing factors, it becomes needful to evaluate the effect of fermenting cassava with Rhizopus oryzae (starter culture) on the nutrients, antinutrients (tannin, phytate and cyanide) and some nutritionally valuable metals (Zn, Fe, Na, K, Ca and Mg).

MATERIALS AND METHODS

Sample preparation

Cassava tubers were collected from the Research farm in the Federal University of Technology, Akure, Nigeria, and they were peeled and crushed. Thereafter, they were pressed using hydraulic press to reduce the cyanide level. Glass distilled water was used.

Fermentation

The Rhizopus oryzae used was isolated and cultivated using potato dextrose agar and broth, respectively . It was then inoculated into 1kg of the mash, as the starter culture and 730mL nutrient solution (Urea, 80g; $MgSO_4$. $2H_2O$, 7g; KH_2PO_4, 13g; Citric acid, 20g). It was then allowed to ferment for 3 days [2]. The product obtained was subsequently sieved and fried into gari.

Compositional analysis

The nutritional composition (ash, fat, crude fibre and carbohydrate) of the fermented and unfermented flour and gari were evaluated using the standard AOAC [8] method. The protein was determined using the micro-kjeldahl method (N x 6.25) while the nutritionally essential elements (Na, Mg, K, Fe, Zn and Ca) were determined using Atomic Absorption Spectro-photometer (AAS). The antinutrient contents of both the fermented and unfermented products were estimated. Phytate was determined by the method of Young and Greaves [9], being based on the ability of standard ferric chloride to precipitate phytate present in the samples. The tannin content was determined using the Makker *et*. *al.*[10] method. This is based on the ability of tannin-like compounds to reduce phosphotungstomolybdic acid in alkaline solution to produce a highly coloured blue solution, the intensity of which is proportional to the amount of tannins. The intensity is measured in a spectrophotometer at 700nm. The cyanide content of the pulp and gari was determined by silver nitrate titration procedure described by De Bruijn [11].

RESULTS AND DISCUSSION

In Africa, the successful use of biotechnology for plant propagation and breeding could dramatically raise crop productivity and overall food production . Tissue culture techniques are already being used to produce more drought and disease resistant varieties of different crops including cassava [12]. Better fermentation techniques in solid media such as protein-enriched cassava flour can improve the nutritional value of crops [13]. Rhizopus oryzae, a non-pathogenic and cheap fungi, was used to ferment cassava pulp and processed to gari, with the aim of increasing the protein content and at the same time detoxifying the products (flour and gari).

The proximate composition of the fermented and unfermented flour and gari in Table I reveals that *Rhizopus oryzae* fermention of cassava increases the protein content of both the flour (8.66%) and gari (5.60%) compared to the unfermented cassava products (flour, 4.43%; gari, 3.64%). The increase in the protein content of both the flour and gari may be due to the fact that *Rhizopus oryzae* which degrades cassava products readily [2], may have secreted extracellular enzymes [6] in the cassava pulp, which consequently increases the protein content of the flour and the gari, as well as the microbial biomass.

However, the protein content of the flour was much higher than that of the gari. The higher protein content of the flour compared to that of the gari may be due to the effect of processing the pulp into gari, which entails pressing, sieving and frying the pulp. Some of the protein may have leached off during pressing and burnt off during frying . It is documented that pre-processing, processing and post-processing methods of preparation of cassava products determines the quality of cassava products [7].

There was no considerable difference in the crude fibre and fat, while there was an increase in ash content of the flour (4.21) unlike the gari (2.28) which had almost the same content with unfermented flour (2.29). There was a general decrease in the carbohydrate content of both the flour (81.61) and the gari (84.23). This may be as a result of the proportionate increase in the protein content. Moreover, some of the carbohydrates might have been used up as carbon source by the organism and converted to microbial biomass.

As shown in Table II, there was no obvious trend in the change in the levels of the elements (Zn, Mg, Fe, Ca, Na and K) of the products except that the fermented gari recorded a general increase. This may probably be due to the fact that a metallic frying pan was used for processing the pressed and sieved pulp into gari and some of those metals may have leached into the gari [7].

The levels of some allelochemicals (tannin, phytate and total cyanide) which the plant probably uses for defense [14], were also determined in the products. Tannins affect nutritive value of food products by forming a complex with protein (both substrate and enzyme) thereby inhibiting digestion and absorption [15]. They also bind Fe, making it unavailable [16] and recent evidence suggests that condensed tannins may cleave DNA in the presence of copper ions [17]. The tannin content of the flour (0.15%) and the gari (0.08%), when compared with the unfermented cassava flour (0.19%), did not give any appreciable decrease in the flour unlike the gari. This probably means that the highest proportion of the decrease in the tannin content may be due to garrification [18] and not fermentation.

Contrary to expectation, the phytate (which is capable of chelating divalent cationic minerals like Ca, Fe, Mg, and Zn, thereby inducing dietary deficiency) content of both the flour (1497.18mg/100g) and the gari (912.26mg/100g) were higher than those of the unfermented products [(flour, 874.37mg/100g and gari, 662.84mg/100g)] although both values were considerably low. In fact, the mechanism of this increase in the phytate level of the fermented product could not be ascertained but it may be due to the fact that some of the plant metabolites or the content of the nutrient solution may have been converted to phytate / phytate-like products.

The total cyanide content as shown in Table III reveals that Rhizopus oryzae fermentation of cassava reduces the cyanide content of both the pulp (17.2mg/kg) and the gari (13.5mg/kg). This low cyanide content may be as a result of the ability of the microorganisms to utilize cyanogenic glucosides and their breakdown products. Moreover cassava processing which involves grating, fermentation and roasting can lead to a reduction in the total cyanide in fresh peeled roots [19]. Akinrele et .al. [20] considered 30mgHCN/kg to be acceptable in gari. Since the value obtained for both flour and gari are far below this critical value, the products can therefore be considered safe as regards cyanide poisoning.

The present study, therefore, reveals that Rhizopus oryzae greatly influences the chemical composition of cassava products positively, by increasing the protein level of cassava products and at the same time reducing the level of some antinutrients, specifically total cyanide and tannin.

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Table I. Proximate composition (%) of *Rhizopus oryzae*-fermented cassava products (flour and gari) (mean ± SD)

	Flour		Gari	
	Unfermented	Fermented	Unfermented	Fermented
Protein	4.4±0.1	8.7±0.1	3.6±0.1	5.6±0.2
Fat	3.8±0.1	2.0±0.1	3.6±0.1	4.2±0.2
Ash	2.3±0.2	4.2±0.2	1.9±0.2	2.3±0.1
Crude fibre	3.8±0.1	3.5±0.4	3.7±0.2	3.7±0.1
CHO	85.7±0.1	81.6±0.3	87.2±0.2	84.23±0.1

Table II. Mineral composition (mg/100g) of *Rhizopus oryzae*-fermented cassava products (flour and gari).

	Flour		Gari	
	Unfermented	Fermented	Unfermented	Fermented
Zn	130.6	37.7	57.5	79.5
Mg	433.8	433.9	277.5	760.1
Fe	259.9	26.0	22.5	42.5
Ca	615.7	203.8	167.1	417.8
Na	437.5	517.4	513.7	1095.7
K	498.2	489.9	555.5	764.6

Table III. Levels (mg/kg) of some antinutrients of *Rhizopus oryzae*-fermented cassava products (flour and gari) .

	Flour		Gari	
Antinutrient	Unfermented	Fermented	Unfermented	Fermented
Tannin[a]	0.2	0.2	0.1	0.1
Cyanide	21.3	17.2	14.9	13.5
Phytate	874.4	1497.2	662.8	12.3

[a]mgTA/kg