Assessment of the Effects of Microbial Fermentation on Selected Anti-Nutrients in the Products of Four Local Cassava Varieties from Niger State, Nigeria

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Abstract The effect of microbial fermentation on the anti-nutrient composition of some cassava products was evaluated by the inoculation of the pulps of four local varieties of cassava, using microorganisms (*Saccharomyces cerevisae and Lactobacillus bulgaricus*). The results of the study revealed that microbial fermentation caused significant reductions (P < 0.05) in the cyanide (86%), tannins (73%), oxalate (61%), phytate (72%) and saponins (92%) contents of the cassava products. The results of the study suggest that cassava products can be nutritionally improved with *Saccharomyces cerevisiae* and *Lactobacillus bulgaricus* fermentations.

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1. Introduction

It was estimated that the world production of cassava roots stood at 184 million tonnes in 2002. Reports indicated that the production is highest in Africa where 99.1 million tonnes were grown, 51.5 million tonnes were cultivated in Asia and 33.2 million tonnes in Latin America and the Caribbean [1]. Of the countries of the world, Nigeria is found to be the largest exporting country of dried cassava with a total of 77% of world export in 2005, seconded by Vietnam, with 13.6%.. Indonesia was next with 5.8% and Costa Rica 2.1%.. World-wide cassava production recorded an increase of 12.5% between 1900 and 1988 [1,2]. In Nigeria the crop contributes over 50% of food intake and provides substantial percentage of food energy in the daily diet. Also, it significantly serves as livestock feed and industrial raw material for production of alcohol and acetones, starches for textiles and pharmaceutical industries over the years [3].

Among the world's staple crops, cassava has the lowest protein/energy ratio (7.4 mg protein/cal compared to 26 mg protein/cal for maize) and is deficient in iron, zinc, pro-vitamin A or β-carotene and vitamin E. This is coupled with the fact that it has one major problem; the roots and leaves of poorly processed cassava plants contain a substance that, when eaten, can trigger the production of cyanide.This is a serious problem for the 500 million people who rely on cassava as their main source of calories, among them are subsistence farmers in Sub-Saharan Africa [4,5].

Proteineous foods are usually known to be expensive and beyond the reach of most of the populace. Its scarcity has greater impact on children, whose physical and mental developments require nutritionally balanced diets. Malnutrition leads to stunting and underweight [6]. The utilisation of cassava is limited by its extremely low protein content and so the consumption of its products has been implicated in malnutrition. Low protein intake in Africa has been attributed to the increasing high cost of traditional sources of animal protein [7].

In many regions of the world over 30% of the population have insufficient calories in their diet to meet their nutritional needs. By 2020, the Food and Agricultural Organisation (FAO) [8] has estimated that one billion persons will be undernourished or receive insufficient calories in their diet. Nowhere is this problem more severe than in sub-Saharan Africa. [4].

Cyanogens are found in 3 forms in cassava: cyanogenic glucoside (95% linamarin and 5% lotaustrin), cyanohydrins, and free cyanide. These glucosides, typified by linamarin [alpha-Hydroxyisobutyronitrile-beta-D-glucose] or [2-(beta-D-Glucopyranosyloxy)-2-methylpropanenitrile] (molecular formular: $C_{10}H_{17}NO_6$) and lotaustralin [2-(beta-d-glucopyranosyloxy)-2-methylbutyronitrile] or [2-(-D-Glucopyranosyloxy)-2-methylbutyronitrile] (moleular

formular: $C_{11}H_{19}NO_6$) are hydrolysed to hydrocyanic acid (HCN) by endogenous linamarase (beta-glucosidase or β-glucosidase) when cassava tissues are disrupted by cutting, grating, bruising or other mechanical means.

Different processing techniques exist to remove cyanogens and their effectiveness depend on the processing steps and the sequence utilized, and it is often time-dependent [9,10,11]. Cyanide is found to be the most toxic factor restricting the consumption of cassava roots and leaves. Indeed, cassava, particularly its bitter varieties, has a cyanide level higher than the Food and Agricultural Organisation/World Health Organisation (FAO/WHO) (1991) recommendations, which is < 10 mg cyanide equivalents/kg DM, to prevent acute toxicity in humans. Cassava leaves have a cyanide content ranging from 53 to 1,300 mg cyanide equivalents/kg of dry matter (DM), and cassava root parenchyma has a range of 10 to 500 mg cyanide equivalents/kg DM; both of these are much higher than what is recommended.

Antinutrients are natural or synthetic compounds that interfere with the absorption of nutrients They reduce the body's ability to absorb or use essential nutrients like vitamins and minerals, and are found in all plant foods, although the types and amounts vary tremendously from food to food. Examples of antinutrients include phytic acid (or phytates), lignans, saponins, phytoestrogens, oxalates, phenolic compounds (tanins), protease inhibitors. Lipase inhibitors, amylase inhibitors and trypsin inhibitors

Phytate (inositol hexakisphosphate) is a compound found in high abundance in cassava, with approximately 624 mg/100 g in roots. Phytic acid is able to bind cations such as magnesium, calcium, iron, zinc, and molybdenum and can, therefore, interfere with mineral absorption and utilization which may affect requirements. It may also bind proteins preventing their complete enzymatic digestion [12,13].

The polyphenol content (tannins) in cassava leaves increases with the maturity of the plant. Polyphenols are able to form insoluble complexes with divalent ions such as iron, zinc, and copper. They can also inactivate thiamin, bind certain salivary and digestive enzymes, and enhance secretion of endogenous protein. Consequently, they inhibit nonheme-Fe absorption, reduce thiamin absorption and the digestibility of starch, protein, and lipids, and also interfere with protein digestibility [14].

Oxalic acid and oxalates are found in many plants, particularly in members of the spinach family Oxalate is the conjugate base of oxalic acid which is capable of binding to metal ions such as Ca^{2+} and Mg^{2+} to form precipitates in the body and prevent their absorption. Oxalates are among the ant-nutrients affect the utilization of protein by forming complexes with proteins, which inhibit peptic digestion. Oxalate content ranges from 1.35 to 2.88 g/100 g DM for cassava leaf meal. The negative effect of oxalates on humans depends on the level of both oxalate and calcium in the cassava leaves [15,16].

Saponins have been found to be the bioactive component of ginseng responsible for its metabolic and potential health effects. Ginseng has been known for treatment of a number of different ailments, such as providing energy, anti-inflammatory effects and preventing fatigue. It also helps to improve cognitive function and erectile dysfunction of men. Saponins can also act on the central nervous system of humans with

potential therapeutic effects. Cassava leaf meal has a steroidal saponin content ranging from 1.74 to 4.73 g/100 g DM, which compares to those found for soybeans (0.07 to 5.1 g/100 g DM), but is lower than those observed in alfalfa (5.6 g/100 g DM) and beet leaves (5.8 g/100 g DM). Saponin content increases in cassava leaf meal with plant maturity [9].

Of the series of available cassava processing methods, fermentation is the first major step in the production of products such as 'gari', 'fufu' and 'lafun'. Some of the objectives behind the fermentation include biological enrichment of the substrate in terms of protein, vitamins, essential amino acids and essential fatty acids. Others purposes for which fermentation is carried out are detoxification of food anti-nutrients, impartation of good aroma, flavour and texture as well as preservation of the fermented products [17]. It is one of the less expensive means of increasing the protein quality of cassava [17,18].

The modern processing techniques involve enhanced fermentation of cassava pulp by the addition of non-pathogenic microorganisms. The use of microorganisms to convert carbohydrates, lignocelluloses and other industrial wastes into foodstuffs rich in protein is possible due to some inherent characteristics [17].

Firstly, microorganisms have a very fast growth rate with high protein content (35 to 60%) [19]. Microorganisms produce an amount of essential nutrient(s) sufficient to correct or prevent deficiency in the consumer. Also, the nutrient(s) produced by microorganisms do not impart undesirable characteristics to food (changes in colour, taste, smell, texture, or cooking properties) and do not unduly shorten the shelf life.

Cyanide can poison a person by either inhalation or ingestion [20]. The normal range of cyanogens content of cassava tubers falls between 15 and 400 mg HCN/kg fresh weight. It was reported by [6] that cyanide levels were found in the range 6 to 370 mg/kg depending on the particular cultivar, growing conditions, (i.e. soil type, humidity, temperature) and the age of the plant [8,21].

The World Health Organisation (WHO) has set the safe level of cyanogens in cassava flour at 10 ppm and the acceptable limit in Indonesia is 40 ppm [22]. Phytates should be lowered as much as possible, ideally to 25 mg or less per 100 g or to about 0.03 percent of the phytate-containing food eaten. At this level, micronutrient losses are minimized [23]. Because retention of phosphorus decreases when phytate in food is 30 – 40% or more of total phosphorus.

The ability of tanins to form complexes with numerous types of molecules such as carbohydrates, proteins, polysaccharides, bacterial cell membranes, enzymes involved in protein and carbohydrates digestion accounted for their adverse effects on animal nutrition. Not only do they form complexes with proteins but also precipitate them. Tannin-protein complexes involve both hydrogen-bonding and hydrophobic interactions. Some tannins were found to produce toxic and anti-nutritional effects in monogastric and ruminant animals and cause reduced intake, lower nutrient digestibility and protein availability. Tannins are commonly found in fruits (grapes, persimmon, blueberry, etc.), tea, chocolate, legume forages (trefoil, etc.), legume trees (Acacia spp., Sesbania spp., etc.), and grasses. They contribute to many aspects of our daily lives as they are responsible for the astringent taste being

experienced when consuming wine or unripe fruits, and for the enchanting colours seen in flowers and in autumn leaves [24,25,26].

This study examines the effect of microbial fermentation (using *Saccharomyces cerevisae and Lactobacillus bulgaricus*) on reducing the anti-nutrient compositions of the cassava products obtained by the inoculation of the pulps of four local varieties of cassava. This is geared towards the provision of enriched cassava products to the populace.

2. Materials And Methods

2.1. Materials

All chemicals used were of analytical grade. The inoculants used for the study were *S cerevisiae* and *L. bulgaricus*. They were obtained from the culture bank of the Department of Microbiology, Federal University of Technology, Minna, Nigeria.

Tuberous roots of four local varieties ('Baba Iya', 'Wahabi', 'Dan Warri' and 'Kpace Bokun') of cassava were obtained from the sampling farm cultivated at Wushishi in Kontagora Senatorial Zone of Niger State, Nigeria. The tubers were each processed into three different products- 'fufu', 'gari' and 'lafun'. These varieties were identified by a Research Fellow and, an agronomist, at the Niger State Agricultural Development Projects, Minna, Niger State, Nigeria.

2.2. Method of Processing the 'Gari'

The fresh roots were peeled and grated, after which the grated pulp was put in sacks (polypropylene) and the sacks placed under heavy stones for 4 days to expel excess liquid from the pulp while it was fermenting. The dewatered and fermented lumps of pulp were then crumbled by hand, and most of the fibrous matter removed. The remaining mass was sieved with traditional sieves of iron mesh. After being sieved, the fine pulp was then roasted in an iron pan over fire.This resulted in a finished white lumpy product called 'gari'. For the production of microbial treated 'gari', the pulp was seperately inoculated with the microbes prior to fermentation.

2.3. Method of Procssing the 'Lafun'

'Lafun' in Nigeria, 'cossettes' in Zaire and Rwanda, 'kanyanga' and 'mapanga' in Malawi and 'makopa' in Tanzania was processed by peeling the fresh root of cassava, followed by grinding and then draining the extracts of the pulp under heavy weight. Sun-drying of the pulp for 5 days until dried yielded 'lafun' The dried crumbs was then milled into flour. Meanwhle, cassava mash to be made into 'lafun' products were neither fermented nor inoculated to agree with the method used locally for preparationas as the fermentation process is usually skipped. In essence, therefore, lafun products provide room for comparison between the fermented products in one hand and the microbially fermented products on the other.

2.4. Method of Processing the 'Fufu'

The freshly peeled roots were soaked in water for 3 days so as to absorb water and get fermented and soft. After fermentation, the water was drained off, most of the fibre were removed and the roots pounded in a wooden mortar, until a soft mash was formed. Excess water was extracted from the mash by placing heavy objects such as rocks on top of the sacks containing the 'fufu'. In order to obtain a microbial fermented 'fufu', 1500 cfu each of *S cerevisiae* and *L. bulgaricus* per 1kg of cassava pulp were seperately introduced prior to fermentation.

2.5. Chemical Analysis of the Cassava Products

Triplicate samples of each cassava product from each of the four varieties were collected (n = 28) and the amount of cyanide in the samples was determined using the Alkaline Picrate Method described by [27]. The saponin content was estimated gravimetrically using a soxhlet extractor involving two different solvents (acetone and methanol) [28]. Determination of oxalate in the samples was carried out in accordance with the methods described by [29]. Phytate was determined in the samples using the method of anion-exchange [30].

2.6. Statistical Analysis

Data was reported as averages of triplicate determinations and analysed using Analysis of Variance (ANOVA). Duncan multiple test at 5% level of significance was used to determine the significant differences among the samples. The statistical package used was IBM SPSS Statistics version 20 (IBM Corp. Amonk. NY).

3. Results and Discussion

3.1. Effect of Inoculation on the Cyanide Content of the Products

From the results in Table 1 and Table 2, the cyanide contents of non-fermented 'lafun' products were significantly higher compared to the 'gari' and 'fufu' products that were subjected to natural and microbial fermentations. However, there were no significant differences ($p < 0.05$) between the values obtained in the natural and microbial fermented products except in the products obtained for 'Baba Iya' variety as shown in Table 1, which has been acclaimed by the farmers in the community to be the most toxic. For example, 'gari' and 'fufu' produced from 'Baba Iya' through natural fermentation had the values reduced from 5.92 ± 0.00 and 4.20 ± 0.08 mg/100 to 2.22 ± 0.04 and 1.84 ± 0.21 mg/100g respectively when treated with *L. bulgaricus*, and were reduced to 1.75 ± 0.05 and 1.10 ± 0.21 mg/100g respectively for 'gari' and 'fufu' products subjected to *S. cerevisae* fermentation.

These values obtained from this research work can be compared with the ones reported by [37], which indicates that the unfermented low-cyanide cassava flour (lafun) recorded 1.09 mg/100 g cyanide content and the *Rhizopus oryzae* and *Saccharomyces crevisae* fermented of the low-cyanide cassava flour were found to be 0.34mg/100g and 0.37 mg/100 g cyanide respectively. Similarly, the

unfermented medium-cyanide cassava flour recorded 2.45 mg/100 g cyanide composition while the *Rhizopus oryzae* and *Saccharomyces crevisae* fermented of the medium-cyanide cassava flour contained 0.85 mg/100 g and 0.84 mg/100 g cyanide respectively. In another report, reference [38] shows that cyanide contents of cassava flour (lafun), garri and fufu were 6 ± 0.12 ppm, 10 ± 0.13 ppm and 5 ± 0.10 ppm respectively.

Table 1. Selected Anti-nutrient compositions of natural, microbial and non fermented cassava products of KB and BI varieties

Variety	product	Parameters				
		Cyanide µg/100g	Oxalate mg/100g	Phytate (mg/100g)	Tannins g/100g	Saponnins (%)
KB	Gari	2.77±0.15[c]	9.44±1.21[e]	297.50±0.45[b]	7.53±2.40[f]	5.06±0.00[e]
	Lafun	3.48±0.03[c]	9.71±0.25[e]	267.50±8.05[b]	8.62±1.00[g]	4.49±0.01[d]
	Fufu	1.05±0.08[a]	8.29±0.50[d]	252.41±7.41[b]	6.91±0.09[e]	2.25±0.10[b]
LIKB	Gari	1.7±0.05a[b]	9.03±1.23[e]	201.00±4.70[b]	6.42±1.14[e]	4.21±0.02[d]
	Fufu	0.98±0.13[a]	8.20±0.28[d]	214.22±3.32[b]	5.89±2.11[d]	1.89±0.00[a]
SIKB	Gari	2.70±0.23[c]	9.23±0.25[e]	242.00±6.70[b]	4.98±106[c]	3.91±0.02[d]
	Fufu	0.72±0.21[a]	8.23±0.11[d]	237..50.±8.8[b]	5.23±0.82[d]	2.21±0.12[b]
BI	Gari	5.92±0.00[e]	10.11±0.00[f]	248.04±6.90[b]	6.19±0.03[e]	3.89±0.00[c]
	Fufu	4.20±0.08[d]	11.27±1.01[g]	204.50±0.45[b]	5.86±0.55[d]	2.19±0.00[b]
	Lafun	6.25±0.08[f]	12.93±0.08[h]	267.54±3.94[b]	6.00±0.92[e]	4.27±0.00[d]
LIBI	Gari	2.22±0.04[b]	6.23±0.25[b]	177.00±4.70[a]	4.13±1.34[c]	2.45±0.00[b]
	Fufu	1.84±0.21[b]	6.01±0.15[b]	196.50±7.80[a]	4.81±2.40[c]	3.48±0.00[c]
SIBI	Gari	1.75±0.05[b]	5.83±0.02[a]	201.50.±8.8[b]	5.00±0.84[d]	4.44±0.00[d]
	Fufu	1.10±0.21[a]	5.11±1.00[a]	164.54±3.84[a]	5.11±0.61[d]	4.22±0.01[d]

Means with the same superscript within a column are not significantly different (p > 0.05)
BI = Baba Iya, LIBI = L. *bulgaricus* fermented, SIBI = *S. cerevisea* fermented, KB = Kpace Bokun, LIKB = L. *bulgaricus* fermented, SIKB = *S. cerevisea* fermented

Table 2. Selected anti-nutrient compositions of natural, microbial and non fermented cassava products of DW and W varieties

Variety	Product	Parameters				
		Cyanide µg/100g	Oxalate mg/100g	Phytate (mg/100g)	Tannins g/100g	Saponins (%)
W	Fufu	0.89±0.15[a]	6.23±0.45[b]	312.09±6.41[c]	2.99±0.07[a]	3.54±0.01[c]
	Gari	2.31±2.99[b]	9.01±0.07[d]	310.50±8.30[c]	3.89±1.00[b]	2.05±0.00[b]
	Lafun	6.34±0.05[f]	12.93±0.08[h]	278.50±4.45[b]	4.89±0.58[c]	5.69±0.01[e]
SIW	Fufu	0.82±0.10[a]	5.74±0.20[a]	271.27±5.37[b]	2.97±1.26[a]	2.57±0.00[b]
	Gari	0.78±0.02[a]	8.48±1.11[c]	291.90±8.00[b]	2.71±0.02[a]	2.21±0.02[b]
LIW	Gari	1.92±0.04[b]	8.70±0.25[c]	304.58±3.90[c]	2.03±1.04[a]	2.00±0.00[b]
	Fufu	0.86±0.21[a]	6.14±0.00[b]	244.10±6.44[b]	2.44±0.00[a]	2.48±0.00[b]
DW	Gari	1.69±0.39[b]	9.23±0.81[e]	287.50.±8.8[b]	7.42±0.72[e]	4.07±0.00[d]
	Fufu	0.99±0.06[a]	5.28±1.81[a]	298.04±6.90[b]	7.18±0.58[c]	3.02±0.01[c]
	Lafun	4.22±1.44[d]	9.57±0.25[e]	248.50±0.45[b]	5.32±1.07[c]	3.24±0.01[c]
LIDW	Gari	1.51±0.26[a]	9.03±0.04[e]	177.54±3.94[a]	6.03±0.57[d]	2.35±0.00[b]
	Fufu	0.87±0.06[a]	5.18±0.29[a]	217.00±4.70[b]	3.89±0.62[a]	3.01±0.01[c]
SIDW	Gari	1.51±0.05[a]	9.40±0.01[e]	226.70±7.80[b]	4.22±1.81[b]	2.11±0.00[b]
	Fufu	1.18±0.10[a]	5.21±0.65[a]	201.50±8.81[b]	5.86±0.94[e]	2.98±0.02[a]

Means with the same superscript within a column are not significantly different (p > 0.05) DW = Dan Warri, LIDW = L. *bulgaricus* fermented, SIDW = *S. cerevisea* fermented W = Wahabi, SIW = *S. cerevisea* fermented, LIW = L. *bulgaricus* fermented.

It was evident from the results that the level of reduction in cyanide level by the action of *L. bulgaricus*-aided fermentation was not significantly different (P < 0.05) from the ones brought about by *S. cerevisae*-facilitated fermentation (Duncan multiple test). However, the ability of the two organisms to bring about reduction of cyanide in 'fufu' and 'gari' products was found to be similar as presented in Figure 1, which contained the results expressed as percentage decrease in the cyanide content. The percentage decrease in cyanide content was highest in Baba Iya species that was *S. cerevisae*-treated 'gari' (70.43%) and *L. bulgaricus*-fermented 'gari' from 'Kpace Bokun' had 38.6% decrease in cyanide level. On the other hand, gari from Wahabi species that was 'inoculated with *S. cerevisae* recorded 43.2% decrease while *L. bulgaricus*-treated 'fufu' from Baba Iya also recorded a 56.2% decrease.

The reduction in cyanide level in the products is attributed to several factors including:

(i) Changes in the texture in the plant tissues which make it possible for vacuole-bound cyanogenic glycosides to diffuse and come in contact with membrane-bound linamarase.

(ii) Increase in β-glycosidase activity in cassava tissue, and

(iii) Utilisation of cyanogenic glycosides and the breakdown of products by fermentation microorganisms. When cyanogenic glycoside is exposed to linamarase, it is cleaved to produce hydrogen cyanide which evaporates or pressed out during cassava processing [31] and [32].

Cassava processing which involves grating, fermentation and roasting can lead to a reduction in the total cyanide more in 'gari' [33]. The results indicated that there was a significant decrease (P < 0.05) in cyanide content of the fermented products as a result of inoculation. The results of this investigation corroborate with the findings of other researchers [32], [34], [35] and [36], though the authors used different microorganisms for the fermentation.
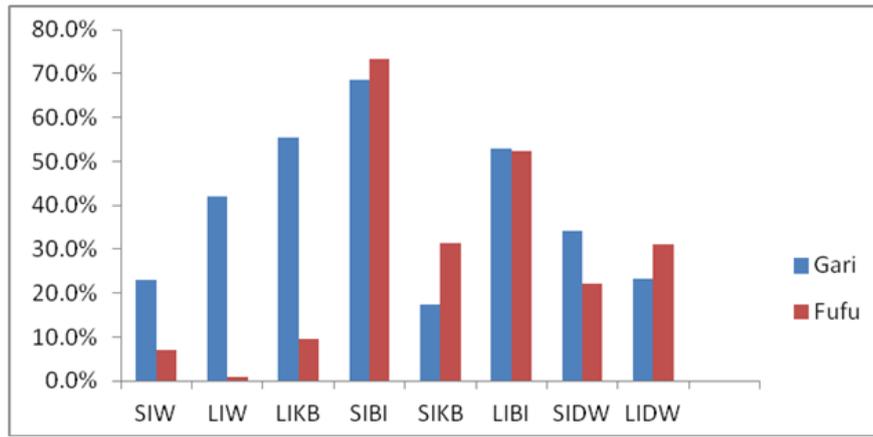
Figure 1. Percentage decrease of cyanide in 'garl' and 'fufu' samples due to inoculation

3.2. Effect of Inoculation on the Oxalate Levelof the Products

The decrease in oxalate content of the products was only significant amongst the natural and microbial fermented products produced from 'Baba Iya' variety (Table 1 and Table 2). Expectedly, the levels of oxalate in the uninoculated 'lafun' products were higher compared to the corresponding values of oxalate contents in both the natural and microbial fermented products. The results in the Table 1 and Table 2 indicated that oxalate content had values ranging from 11.27 ± 1.01 mg/100g for the naturally fermented 'fufu' from 'Baba Iya' to 5.11 ± 1.00 mg/100g dry sample for 'fufu' treated with *S. cerevisae* and from the same variety. Though, result data on the oxalate content of cassava products, is scanty, the available data showed that attempt made by [34] to investigate the effect of boiling and natural fermentation on the anti-nutritional factors of two cassava varieties (Qulle and Kello) collected from Southern Ethiopia.the

results revealed that the level of oxalate was found to be 24.93 ± 0.08 and 5.04 ± 0.02, 86.18 ± 0.10 mg/100g in raw Qulle and Kello samples respectively.

From the results of this research, reduction in oxalate was more pronounced in 'gari' products. This was attributed to processing method of 'gari' which involves grating, fermentation and roasting thereby leading to a reduction in oxalate.

In Figure 2, the oxalate level was presented as percentage derease in the products, the results indicated that reduction of oxalate was more in the 'gari' products. This was also linked to the processing method of 'gari' which involves grating, fermentation and roasting thereby leading to reductions in oxalate. The result further pointed out that the level of oxalate reduction by the action of *L. bulgaricus*-aided fermentation was not significantly different from the ones brought about by *S. cerevisae*-facilitated fermentation. There were also significant differences ($p < 0.05$) between the values of oxalate in the natural and microbial fermented products.
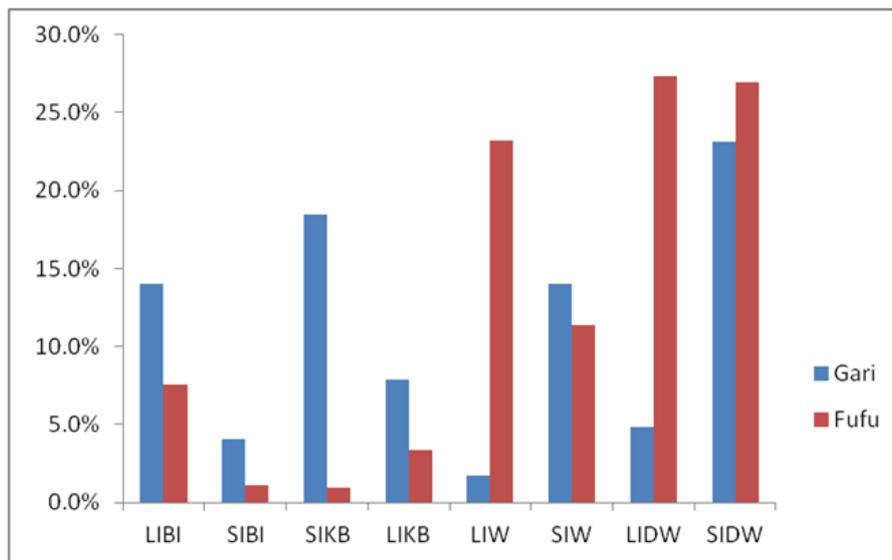
Figure 2. Percentage decrease of oxalate in 'garl' and 'fufu' samples due to inoculation

3.3 Effect of Inoculation on Thephytate Content of the Products

The phytate content in the products ranged from 177.00 ± 4.70 mg/100 g to 312.09 ± 6.41 mg/100g for 'gari'

fermented with *L. bulgaricus* from 'Baba Iya' and 'fufu' naturally fermented and made from 'Wahabi' cultivars respectively (Table 1 and Table 2). Expectedly, the values recorded for 'lafun' products (the unfermented products) and the ones obtained for 'gari' and 'fufu' products being naturally inoculated were higher than the values obtained

for 'gari' and 'fufu' products that were microbially treated. These values conformed to those obtained by [35] which ranged from 173.57 ± 0.56 mg/100g to 104.48 ± 0.68 mg/100g for Qulle flour sample and Kello varieties respectively. The results also agreed with the ones recorded by the work of [39] after fermenting cassava pulp with pure strains of some common saprophytes namely, *Aspergillus flavus, Aspergillus niger, Rhizopus oryzae* and *Saccharomyces spp.* The results of the studies showed that phytate content in microbial fermented cassava products decreased [flour (505.6 ± 4.2−748.5 ± 2.3 mg/100 g) and gari (373.5 ± 6.2−56.7 ± 5.2 mg/100 g)] when compared to naturally fermented cassava products [flour (705.1 ± 1.2 mg/100 g) and gari (633.9 ± 3.2 mg/100 g)] and unfermented cassava products [flour (874.4 ± 3.2 mg/100 g) and gari (662.8 ± 6.5 mg/100 g)].

Figure 3 presents the percentage decrease in the levels of phytate in the microbial fermented cassava products. The results showed that the highest percentage reduction was recorded in 'Kpace Bokun' cultivar used to produce 'fufu' and treated with *S. cerevicea* (38.24%) and the least reduction was in 'gari' from 'Wahabi' treated through *L. bulgaricus* fermentation (1.91%). Also, the ability of the organisms to detoxify 'fufu' products of phytate is higher than in the 'gari' in most cases. The decrease in the phytate content of the microbially fermented cassava products could possibly be attributed to the secretion of the enzyme phytase by microbes during fermentation. These enzymes are capable of hydrolysing phytate, thereby, decreasing the phytate content of the products [37].
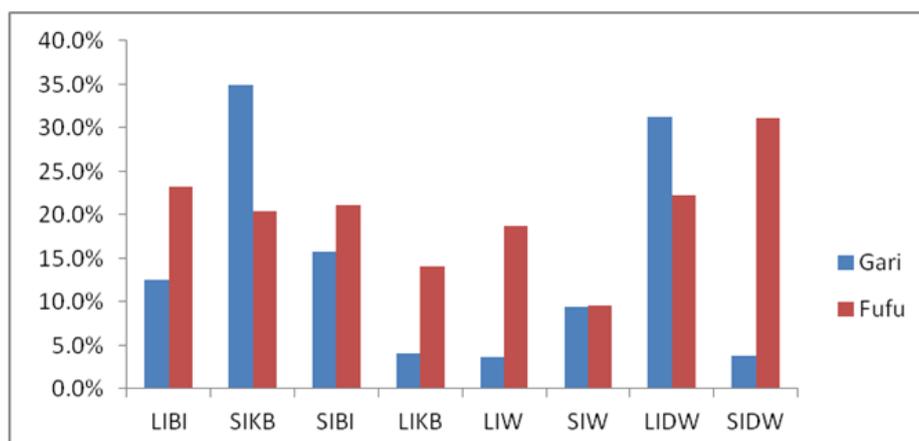
Figure 3. Percentage decrease of phytate in 'garl' and 'fufu' samples due to inoculation

3.4. Effect of Inoculation on the Tannin Composition of the Products

The amount of tannin presented in the Table 1 and Table 2 indicated that the levels of tannin ranged from 2.03 ± 1.04 mg/100g to 8.62±1.00mg/100g for 'gari' from 'Dan Warri' inoculated with *S. cerevisae* and 'lafun' from 'Kpace Bokun' not fermented respectively. There was a significant difference between the percentage obtained for the microbial treated products compared to the untreated ones. Levels of tannin in the 'lafun' products were the highest because the products were neither microbial

fermented nor naturally fermented. Reference [37] carried out similar research work and the findings indicated thatt the unfermented low-cyanide cassava flour (lafun) had 0.4 ± 0.0% tanin content and the *Rhizopus oryzae* and *Saccharomyces crevisae* fermented of the low-cyanide cassava flour recorded 0.10 ± 0.0% and 0.10 ± 0.0 0.37% tanin respectively. As for the unfermented medium-cyanide cassava flour it was found to contain 0.4 ± 0.0% tanin content while the *Rhizopus oryzae* and *Saccharomyces crevisae* fermented of the medium-cyanide cassava flour recorded 0.2 ± 0.0% and 0.2 ± 0.0% tanin respectively.
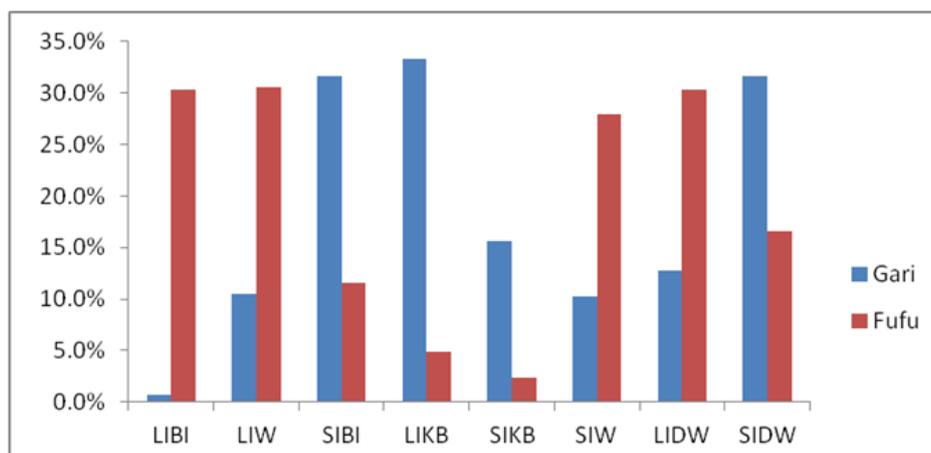
Figure 4. Percentage decrease of tannin in 'garl' and 'fufu' samples due to inoculation

The percentage loss in tannins occurred in all the cultivars fermented with *L. bulgaricus* and *S. cerevisae* as shown in Figure 4. For tannins, the reductions were markedly high in *S. cerevisae*-inoculated 'gari' from 'Kpace Bokun' (33.86%), 'Wahabi' (30.33%), *L. bulgaricus*-treated 'gari' from 'Wahabi' (47.81%) and 'Baba Iya' (33.27%) . Tannin contents of 'Dan Warri's 'gari' treated with *S. cerevisae* and that of 'fufu' of the same 'Dan Warri' decreased by 43.12% and 51.73 % respectively. Decrease in tannin content can be attributed to the degradation of polyphenols as a result of different microorganisms that are involved in fermentation and the corresponding microbial enzymes that are released during fermentation period [35]. From the results, it can be inferred that the two organisms were good at bringing about a reasonable reduction of tannin in the 'gari' products compared to 'fufu' products, while there was no appreciable difference between the abilities of the two organisms to reduce tannin contents in cassava products.

3.5. Effect of Inoculation on the Saponin Content of the Products

From Table 1 and Table 2, the result of saponin content showed that the levels obtained for the inoculated cassava products were higher than those values recorded for the uninoculated counterparts. This trend remained the same irrespective of the variety of cassava and the anti-nutritional component under consideration. There is no available data specifically on saponins in cassava products or even cassava tubers from literature.

The result of saponin content presented in Figure 5 indicated that the highest reduction in saponin occurred in *S. cerevisae*-inoculated 'fufu' from 'Baba Iya' (70.11%) and the least reduction was in 'gari' treated with *L. bulgaricus* produced from 'Wahabi' (2.43%). From these results it is obvious that *S. cerevisae* had the ability to detoxify casssava products more than the *L. bulgaricus*, since most of the treatments with L. *bulgaricus* recorded lower reductions in the anti-nutrient.
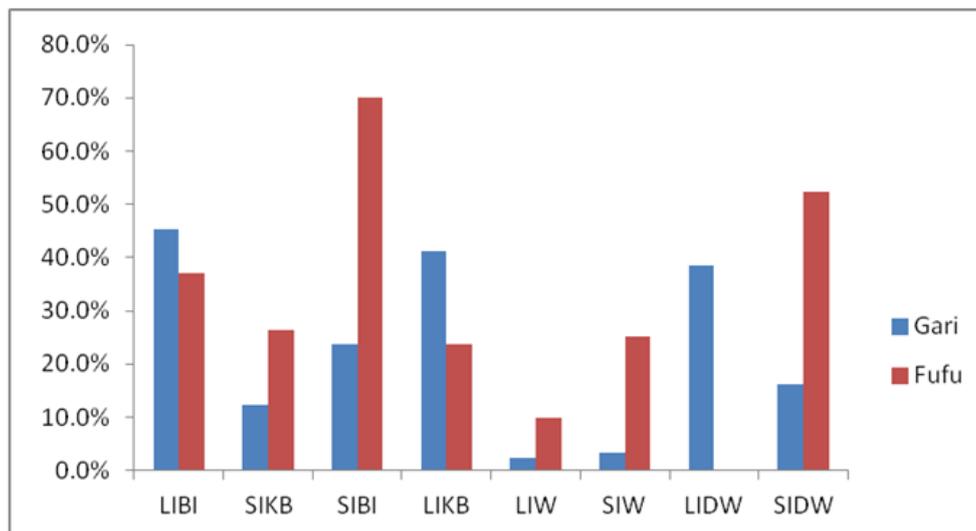
Figure 5. Percentage saponin decrease in 'garl' and 'fufu' samples due to inoculation

4. Conclusion

The inference drawn from the study are that inoculation of cassava pulp with *S. cerevisae* and *L. bulgaricus* during fermentation enhanced the nutritional status of the products obtained by drastically reducing the anti-nutrients components determined. The nature of organism used for fermentation significantly affected the quality of the products.

S. cerevisae had greater ability to a large extent (about 40%) to detoxify the cassava products than the *L. bulgaricus*. The two organisms (*S. cerevisae* and *L. bulgaricus*) showed greater potency to improve the quality of 'fufu' products than the quality of 'gari' products. There was no clear trend regarding varietal compositions between the improved and local varieties investgated. With the significant reductions in the anti-nutrients determined, health disorders associated with the consumption of excess anti-nutrients, particularly among people who are already malnourished could reduce drastically.

Aknowledgements

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