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Effects of Different Processing Methods on the Proximate and Cyanogenic Composition of Flour from Different Cassava Varieties.

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Research Article

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ABSTRACT

Cyanogens have long been recognized as a toxic component of edible roots of cassava and leaves. The cyanogenic content of the roots can vary from less than 10 to more than 500mg/Kg, measured as hydrogen cyanide (HCN) on a fresh weight bases. Cassava roots are processed by a variety of methods into many different food products, depending on locally available processing resources, local customs and preferences. These different processing methods are expected to affect the cyanogenic content of cassava. To investigate the effect of different processing methods on the chemical characteristics of new cassava varieties, three types of cassava flours were prepared by the method of slicing, grating and reconstitution respectively. *Yebeshie* and *Abasafitaa* cassava varieties were processed and their proximate composition, starch, non-glucosidic cyanogens, free cyanide and total cyanogens studied. The slicing method resulted in the highest protein content in both varieties. Starch contents of 65.20% and 63.40% were recorded for flour from the reconstitution method for *Yebeshie* and *Abasafitaa* respectively. The lowest total cyanogen of 0.238mg CN equivalent/Kg and free cyanide of 0.058 mg CN equivalent/Kg were observed for flour from *Abasafitaa* prepared by the reconstitution method. Flours from the slicing methods had the highest non-glucosidic cyanogens followed by flours from the grating method. The reconstitution method can therefore be effectively used to produce cassava flour with a high starch content and lower cyanogenic potential.

INTRODUCTION

Cassava (*Manihot esculenta crantz*) constitutes one of the main staple foods in many sub-Saharan countries in Africa. Cassava is a significant source of calories for more than 500 million people worldwide [1]. It plays a food security role in providing a stable food base in areas prone to drought and famine. For fresh consumption, roots are boiled or eaten raw. Leaves of cassava are also used as a fresh vegetable in several countries, often providing protein, vitamins and minerals in diets which otherwise are nutritionally marginal. Compared to grains, cassava is more tolerant of low soil fertility, and more resistant to drought, pests and diseases. One main disadvantage of cassava is its rapid post-harvest deterioration, which is a major constraint for its utilization. Once harvested, cassava roots are highly perishbable and when stored, rapid physiological and microbiological deterioration occur [2]. Thus, cassava must be processed and/or preserved immediately after harvesting.

The International Institute of Tropical Agriculture (IITA) in Ibadan, Nigeria has played a leading role in the development of improved cassava varieties, which are disease, and pest resistant, low in cyanide content, drought resistant, early maturing, and high yielding. The improved varieties have been introduced throughout Africa's cassava belt. Varieties with resistance to the major diseases give sustained yields of about 50% more. Like most root crops such as sweet potato, yam, and Irish potato, cassava contains very little fat (0.1%) and protein (2-3%). Its protein content is the lowest among the root crops. On the other hand, it is relatively rich in calcium and ascorbic acid (vitamin C), and it contains significant amounts of thiamin (vitamin B1), riboflavin (vitamin B2), and niacin [3]. Varying amounts of the elements mentioned above in the raw tubers of the cassava roots are lost when the roots are subjected to various treatments such as soaking in water, grating, grinding, fermentation and pounding during processing [4][5] . These treatments prior to drying have also been reported to significantly reduce cyanide levels in the resulting cassava flour. Though the effects of different treatments on the nutritional and cyanogenic potential of cassava have been extensively studied, it is still important that more investigations on new cassava varieties are carried out. This study therefore aims at investigating how different processing methods influence the chemical characteristics cassava varieties.

MATERIALS AND METHODS

Collection of Cassava varieties

Cassava varieties nicknamed *Yebeshie* and *Abasafitaa were* obtained from IITA inIbadan, Oyo-State, Nigeria.

Cassava Flour Preparation

Three different processing methods were used for the cassava flour preparation.

Slicing method

The fresh cassava tubers were peeled, washed and sliced to 2-3cm thicknesses. These slices were spread thinly on trays before drying at a temperature of 50^{0}C for 8h. The dried chips were then milled to obtain the flour, which was then cooled and packaged in polythene bags, sealed and stored at 4^{0}C for further analysis.

Reconstitution method

This was carried out according to the method reported by [6] . Fresh cassava tubers were peeled, washed, chopped and blended with water into a paste. This was then sieved to separate the starch from the fibre using lots of water to wash the starch from the fibre. The starch was made to settle for 10h before the supernatant was decanted and the starch dried at 50^{0}C for 8h. The fibre/residue that was obtained was dried immediately at a temperature of 50^{0}C for 8h. The dried starch and fibre were then milled and mixed thoroughly using a Y cone mixer to obtain the flour. The mixed flour was then allowed to cool before packaging. These were kept in sealed polythene bags in the cold room (4^{0}C) for further analysis. This flour is referred to as the Reconstituted Cassava Flour (RCF).

Grating method

The fresh cassava tubers were washed, peeled, grated and dewatered and oven-dried immediately at a temperature of 50^{0}C for about 8h. The dried grated cassava was then milled into fine powder using an attrition mill. The flour was allowed to cool before packaging. These were kept in sealed polythene bags in the cold room (4^{0}C) for further analysis.

Chemical Composition

The proximate composition (ash, fibre, protein, moisture) was carried out by standard methods of [7].

Starch Composition

The starch composition was done according to the Linther's method of [8] in the raw cassava varieties. About 5g of sample was used and the optical rotation readings of the starch were taken in duplicates. The Luff Schoorl method [9] was used for starch determination in the cassava flour samples.

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Cyanogenic Compounds Determination for Flour Samples

The total cyanogens, non-glucosidic cyanogens and the free cyanide levels in the samples were determined by a modification of the enzymatic assays as described by [10].

Extraction of Cyanogenic Compounds

Cyanogenic compounds were extracted by swirling gently 4g of sample in 25ml of 0.1M orthophosphoric acid, H_3PO_4 for 5 minutes. It was then centrifuged for 15min at 3300rpm and the supernatant retained for the assays.

Total Cyanogens

This was determined by adding 0.1ml aliquots of sample extract to 0.4ml of pH 7 sodium phosphate buffer, followed by 0.1ml of linamarase enzyme (linamarase from cassava, BDH, Poole, UK) and then incubated for 15minutes at 30^0C. 0.6ml of 0.2 NaOH was then added to the digested extract. After 5minutes, 2.8ml of sodium phosphate buffer (pH 6.0) was added.

Colorimetric Procedure

Colorations of the sample was carried out by adding 0.1ml of chloramine T (N-chloro-p-toluene sulphonimide sodium salt) to the 4ml of buffered extract which was then mixed and left to stand for 5mins, after which 0.6ml of the colour reagent (18.5g NaOH, 35g 1,3-dimethyl barbituric acid and 28.5g isonicotinic acid per Litre, adjusted to pH 7-8) was added. The absorbance was measured using a Beckman spectrophotometer at a wavelength of 605nm after 25min. A graph of absorbance against concentration (nmols) was plotted. A reagent blank was used for the analysis

Free cyanide (HCN)

0.6ml aliquot of extract was added to 3.4ml of pH 6 sodium phosphate buffer in test tube. The treated extract was assayed colorimetrically by colouration and measurements of absorbance.

Calibration Curves for Standards

Two standard curves were plotted, using linamarin for the total cyanogens standard curve and potassium cyanide (KCN) for the free cyanide standard curve. The calibration curve plots were done by preparing serial dilutions of 20%, 2%, 0.2%, 0.02%, 0.002% from the stock solutions per tube for linamarin and potassium cyanide (KCN) for the determinations of their respective total cyanogens and free cyanide methods.

Statistical analyses

The data obtained from the analyses were statistically analyzed using Statgraphics (Graphics Software System, STCC, Inc. U.S.A). Comparisons between sample treatments and the indices were done using analysis of variance (ANOVA) with a probability $p \leq 0.05$.

RESULTS AND DISCUSSION

Chemical Characteristics

Moisture content of flour is very important regarding its shelf life, lower the flour moisture, the better its storage stability [11] . In general the moisture contents of all the flour samples were low; it ranged from 6.32% to 10.31% for both varieties. The moisture contents of the flour samples from the method of grating was method of grating was the highest amongst the processing methods used. This should be expected because the grating resulted in larger surface area for the faster loss of water from the samples. Considering the two cassava varieties used, cassava flour produced from the *Yebeshie* cassava variety by the method of slicing had the lowest moisture content of 6.32% as indicated in Table 1. The moisture contents of the different flour samples from the *Abasafitaa* cassava variety were higher than those of the *Yebeshie* variety even though the moisture content of its tubers was lower than that of the tubers of *Yebeshie*. For the *Abasafitaa* cassava variety, the flour produced from the method of reconstitution recorded the lowest moisture content.

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Table 1

Cassava Variety	Samples	[1]Moisture (%)	**[1]Protein (%)	[1]Ash (%)	Crude Fibre (%)	Starch (%)
Yebeshie	Cassava Flour	58.79±0.14	1.43±0.01	2.84+0.01	5.45	35.9
	Flour from Slices	6.32 ±0.02	1.07±0.00	1.93±0.07	5.40	48.95
	Flour from Grates	8.12 ±0.14	0.88±0.00	0.82±0.02	4.25	59.27
	*RCF	7.93±0.01	0.71±0.00	0.79±0.02	3.57	65.20
Abasafitaa	Cassava Flour	55.44±0.00	0.90±0.04	1.90±0.01	3.62	47.1
	Flour from Slices	9.65±0.09	1.24±0.00	3.66±0.08	3.59	35.85
	Flour from Grates	10.31±0.02	1.07±0.00	3.23±0.01	3.26	35.09
	*RCF	9.24±0.17	0.35±0.00	0.97±0.01	2.87	63.40

1: Average of two determinations ±standard deviation
*RCF – Reconstituted Cassava Flour
**NX6.25

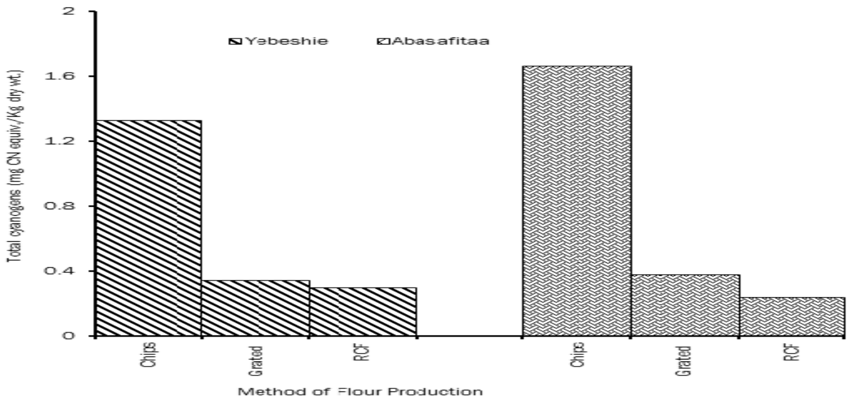

Figure 1

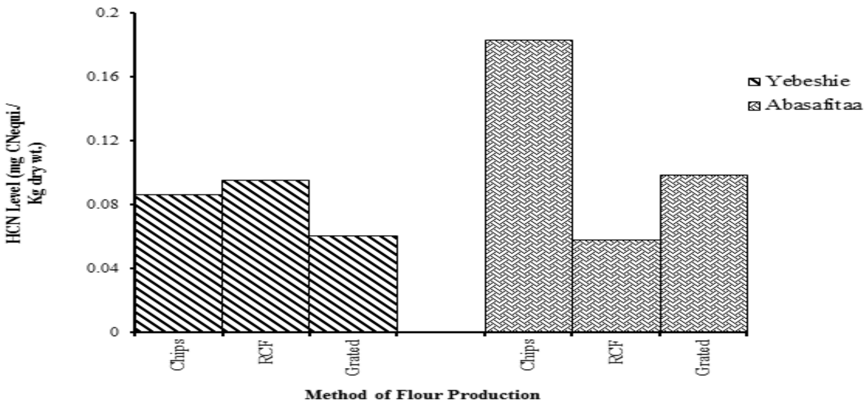

Figure 2

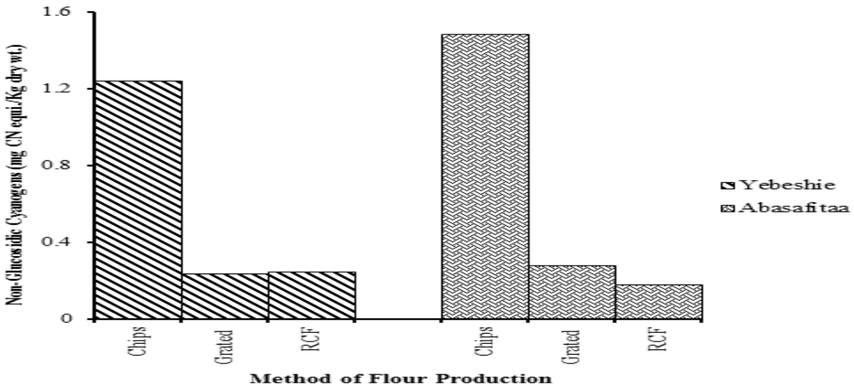

Figure 3

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The protein content of the cassava samples were significantly ($p<0.05$) affected by the different methods used. The method of reconstitution resulted in cassava flour samples with very low protein contents. Like most root crops such as sweet potato, yam, and Irish potato, cassava contains very protein (2-3%). Its protein content is the lowest among the root crops [3] . Table 1 indicates that the RCF method significantly reduced the protein content of the *Yebeshie* cassava variety from 1.43% to 0.71%. The cassava flour of the *Yebeshie* had more protein as compared to the *Abasafitaa* cassava. With respect to the protein content, the slicing method resulted in flour samples with relatively high protein contents. For example, the protein contents of the cassava flour samples from the *Abasafitaa* and *Yebeshie* cassava varieties produced from the method of slicing recorded protein contents of 1.24% and 1.07% respectively. The ash content of the cassava varieties used were also significantly ($p<0.05$) by the different processing methods used. Once again the reconstitution method significantly reduced the ash content of the samples. This means the limited minerals in the cassava varieties are significantly reduced by the methods of reconstitution. The method of slicing resulted in the flour samples with high ash contents meaning the method was able to retain most of the minerals in the cassava samples. According to [12] the average fibre content for cassava flour is about 0.80%. The fibre contents of the different flour samples ranged from 2.87% to 5.45%. The fibre contents of the flour prepared by the reconstitution method were lower than the other samples. This may be attributed to the fact that some of the fibre may have been lost during its separation from the starch of the cassava varieties. Some of the fibre may again have been lost during the process of grating hence resulting to its lower fibre content that that from the flours produced by slicing. The ash content was reduced to the minimum by the method of reconstitution. Starch is one of the most important plant products to man. It is an essential component of food providing a large proportion of the daily calorific intake. In West Africa, cassava flour and *gari* (a processed cassava product) are consumed in large quantities. Cassava starch is recommended for use in extruded snacks for improved expansion. It is also used as a thickener in foods that are not subject to rigorous processing conditions. Cassava starch, which is very bland in flavor, is used in processed baby foods as a filler material and bonding agent in confectionary and biscuit industries [13] . High starch contents of 65.2% and 63.4% for flour samples from the *Yebeshie* and *Abasafitaa* varieties respectively were recorded for the varieties compared to 24% reported by [13]. The reconstitution method of the flour production resulted in the highest starch contents of the three methods. This could be due to the separation of the starch from the fibre of the fresh cassava. The separation could have prevented the loss of starch during the processing of the flour. The starch content recorded for the flour samples from *Abasafitaa* produced by the methods of slicing and grating were comparable.

Cyanogenic Compounds in Flour Samples

The major cyanogenic glycoside in cassava is linamarin, with a small amount of lotaustralin (methyl linamarin). In the presence of the enzyme linamarinase, linamarin is rapidly hydrolysed to glucose and acetone cyanohydrin and lotaustralin hydrolysed to a related cyanohydrin and glucose. Under neutral conditions, acetone cyanohydrin decomposes to acetone and hydrogen cyanide [5]. The effects of the different processing methods on the cyanide content of the cassava varieties used are shown in Figs. 1, 2& 3. Generally, the cyanogens levels observed in the *Abasafitaa* cassava variety were higher than in the *Yebeshie* cassava variety. The amount of glucosides mainly consisting of linamarin (about 90%), can reach 1500mg CN equivalent per Kg dry weight in fresh roots, particularly in those of bitter varieties grown for their higher yields [16] but with efficient processing methods, this can be reduced to negligible levels. There is a great need for development of improved processing methods to greatly reduce the total cyanide content of cassava flour [14]. The safe level of cyanide in cassava flour has been set by the WHO as 10 ppm [15] while the acceptable level in Indonesia is 40 ppm [16]. The different processing methods significantly influenced the cyanogens levels in the cassava varieties. It has also been reported by [20] that methods which use grating and crushing are very effective in removing cyanide because of the intimate contact in the finely-divided wet parenchyma between linamarin and the hydrolysing enzyme linamarase, which promotes rapid breakdown of linamarin to hydrogen cyanide gas that escapes into the air. The level of HCN was very high in the *Abasafitaa* cassava flour prepared from the slicing method. If the cassava plant is not adequately detoxified during the processing or preparation of the food, it is potentially toxic because of the release of this preformed hydrogen cyanide. The reconstitution method produced flour samples with the least total cyanogen content (Fig. 1). Values of 0.295 and 0.238 mg CN equivalent/ Kg of total cyanide were recorded for flour samples from the *Yebeshie* and *Abasaafita* cassava varieties respectively produced by the method of reconstitution. The total cyanogen was significantly very high in flour samples produced by the method of slicing from both cassava varieties. This means that the method of slicing should not be considered when the aim of processing cassava flour is to reduce the total cyanogens in cassava. The methods of grating and reconstitution significantly reduced the non-glucosidic cyanogens in the flour samples compared to the non-glucosidic content in the flour produced by slicing. The non-glucosidic

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cyanogens content of the flour samples was highest in the flour sample produced by the method of slicing for both varieties (Fig. 2). The method of grating resulted in flour samples with non-glucosidic cyanogens content of 0.247 and 0.279 mg CN equivalent/ Kg for the *Yebeshie* and *Abasafitaa* respectively. The least free cyanide as HCN for the two varieties was observed in the flour produced by the method of grating from the *Abasafitaa* cassava variety (Fig. 3). This observation may be attributed to the fact that the separation of the starch and the fibre and drying them separately might have resulted in effective removal of free cyanide in the cassava as HCN. The free cyanide content of the different flours from the Yebeshie cassava variety was no significantly different. However for the *Abasafitaa* cassava variety, very high free cyanide content was observed in the flour produced by the method of slicing

CONCLUSIONS

The different methods of cassava processing influenced the nutritional characteristics of the cassava varieties used. The starch content for instance was marginally higher in flour samples produced by the method of reconstitution. The method of reconstitution significantly reduced the total cyanogen contents of the cassava flours produced. This implies that the method of reconstitution can effectively be used to produce cassava flour that is very low in cyanogens. Flour samples produced by the method of slicing had very high cyanogen content.

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