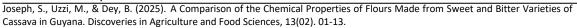
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# A Comparison of the Chemical Properties of Flours Made from Sweet and Bitter Varieties of Cassava in Guyana

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#### ABSTRACT

The use of cassava flour in flour composites and as a gluten-free alternative to wheat is on the increase. The Government of Guyana seeks to tap into this emerging market by increasing cassava flour production and exports however the absence of a regional standard for cassava flour limits exportation goals. Characteristics of prepared flours depend on cultivar type, geographical location, age of the plant and environmental conditions. This research sought to characterize cassava flours made from bitter and sweet varieties of the same age and sourced from the same geographical location. Flours were prepared from the roots of three sweet varieties; ALBS, Smokie and Yardie; and three bitter varieties; LPL67, GAA98 and WS13. The chemical characteristics observed were moisture, protein, starch, far, crude fiber, ash, residual cyanide and pH. There were no significant differences (p>0.05) in moisture, starch, crude fiber, ash and pH between bitter and sweet varieties. Differences were significant (p<0.05) for protein (bitter: 1.79%, sweet: 2.92%), fats (bitter: 2.32%, sweet: 1.62%) and cyanide (bitter: 84.24ppm, sweet: 12.41ppm). An inverse relationship (R = -0.667) existed between moisture and protein content for the sweet varieties. All flours prepared from the bitter varieties had residual cyanide levels in excess of the 10 mg/kg recommended by WHO. This preliminary data favors sweet varieties for cassava flour production and provides initial data for a local repository of cassava flour characteristics for use in generation of a local/regional standard.

**Keywords:** cassava flour, proximate analysis, sweet varieties, bitter varieties, pH, cyanide.

# INTRODUCTION

Manihot Esculenta commonly called "cassava" in Guyana is a tuberous, edible root which grows in tropical regions of the world. This crop can be classified into two broad groups based on the level of cyanogenic glycosides present in the tuber. Tubers with higher concentrations (>50 mg/Kg) of cyanogenic glycosides are termed "bitter cassava" and tubers with lower concentrations (<50 mg/Kg) of cyanogenic glycosides are termed "sweet cassava" [1]. Though

cassava cultivation and consumption have been culturally associated with the indigenous peoples of Guyana, the tuber is cultivated across all regions of the country by small scale farmers and consumed across all sections of the Guyanese society. The harvested tubers are sold at community and city markets for individual, domestic use and more recently, increasing amounts are being sold to processing facilities for the production of value-added products including cassava flour.

As a result, local agricultural authorities have ramped up encouragement and support for farmers to increase cassava cultivation for cassava flour production [2], [3], [4]. The government has encouraged this initiative with financial and technical support from the Food and Agriculture Organization of the United Nations (FAO), European Union (EU), Latin American and Caribbean consortium to support cassava research and development (CLAYUCA) and the Caribbean Community and Common Market (CARICOM)[5], [6], [7]. This initiative resulted in the establishment of cassava flour processing facilities[3], distribution of mills for cassava flour production, distribution of varieties and outreach training sessions with farmers [5]. The increased cultivation of cassava and production of cassava flour support the CARICOM initiative "25 by 2025" which seeks to reduce the food import bill of the Caribbean Region by 25% by 2025. Further, establishment of cassava flour factories is expected to boost the income potential of farmers and generate revenue for the country. The success of these initiatives is evident in the establishment of the Mainstay/Whyaka Cassava Processing Facility in November, 2024 [8], increased cassava yield - 18 tons per hectare in 2019 to 30 tons per hectare in 2024 [9] and increased cassava production – greater than 20 million pounds of cassava reaped by mid-May in 2024 [10]. Increases in cassava flour exports were also reported; in 2019, Guyana exported 30 kg of cassava flour to Barbados [11] while in 2023 export volume rose to 1330 kg, 91% of which was exported to Canada and the remaining 9% to Suriname [12]. Notwithstanding the massive increases in cassava yield, cassava flour production and exportation volumes, exports remain relatively low, due to the lack of international safety and quality certifications for the product and the production process. To realize exportation goals, cassava flour produced in Guyana needs to satisfy Hazard Analysis and Critical Control Points (HACCP) for food safety control and ISO requirements for food safety, quality management and laboratory testing for quality assurance. This research seeks to provide nutrient and antinutrient profiles of cassava flour produced in Guyana from common cassava varieties. The data will contribute to the body of knowledge on this important food product and serve to inform agricultural scientists and farmers on appropriate choices of varieties for cassava flour production. The information garnered here can also be used to provide requisite information for development of a regional standard for cassava flour and provide a framework for achieving quality certification.

Several common varieties of cassava including both the sweet and bitter varieties are utilized in the production of cassava flour in Guyana. Some of the more common varieties include *Uncle Mack, NAREI 1, Bad Woman, Red Stem* (Sweet varieties), *Amazonian, Egg Yolk, Red Stem* and *White Stem* (Bitter varieties) [13]. Despite the drive to increase production and export of cassava flour, and existing evidence that cultivar variety affects the chemical characteristics of cassava flours [14], [15], there are no documented findings on the quality and chemical characteristics of cassava flours prepared from the tubers of varieties grown in Guyana. Studies

in Indonesia and Malaysia showed that nutrient and cyanide contents of high quality cassava flour (HQCF) varied with the variety of the cassava cultivar from which the flour was produced [14], [15]. Earlier studies on fresh cassava tubers grown in Malawi revealed variations in nutrient content based on variety of the cultivar, geographic location, age of the plant, and environmental conditions [16]. Variation in chemical characteristics of the fresh root results in variation in the chemical characteristics of value-added cassava products. The steps involved in processing cassava into other products alter nutrient content and reduce cyanide content but the extent to which these contents change tend to be constant for given methods of processing and increase with more extreme processing conditions [17]. Previous work by this research team on the cyanide content of value-added cassava products available in Guyana revealed that products such as flour and tapioca, which were prepared by more extensive processing techniques, possessed lower cyanide contents [18]. The dependence of chemical characteristics of cassava and cassava-based products on location, environmental conditions, variety and age of the plant, may extend to functional characteristics of the products [19].

In the case of cassava flour, these may include pH, colour, bulk density and foam capacity. In this research, the varieties of cassava used were all grown in the same location under similar environmental conditions and for the same period hence variations in chemical characteristics are only a function of cultivar variety. The research aimed to characterize and differentiate samples of Guyanese cassava flour made from bitter and sweet cassava varieties based on their food value and cyanide content. The parameters investigated are proximate analysis (moisture, protein, fat, starch, fiber and ash content), cyanide level and pH of the flours produced.

The following research questions are investigated.

- Is there a significant difference in the chemical characteristics of cassava flour made from bitter and sweet varieties?
- Are there correlations in chemical characteristics among the varieties?
- Are the chemical characteristics of the cassava flour produced compliant with FAO requirements?
- Can a cultivar be identified that is best suited for cassava flour production?

#### **METHODS**

# **Acquisition of Tubers**

Tubers of the "sweet" and "bitter" varieties of cassava used in this research were grown at the Ebini Research Station and collected from the National Agriculture Research and Extension Institute (NAREI), Guyana. These tubers were stored in a refrigerator for 24 hours before they were processed.

# Flour Preparation

Fresh quality tubers were peeled, cut into pieces, washed and soaked in water for 15 minutes. They were then grated and pressed through a muslin fabric to remove as much moisture as possible. The remaining pulp was dried in an air oven (memmert GmbH+Co.KG D91126) at  $50^{\circ}$ C for 24hrs. The dried pulp was then sifted through a sieve of 0.125 mm.

# **Chemical Analysis**

# **Proximate Analysis:**

Proximate analyses of fats, starch, protein, crude fiber, moisture and ash of the cassava flour samples were determined using rapid analysis with a near infrared analyzer (FOSS NIRS™ DS3). Triplicate analysis on each sample was carried out.

# pH:

The pH of the cassava flour samples was determined by a method adapted from researchers in Ghana and Sweden [20]. A slurry comprising ten (10) grams of cassava flour sample and 90mL of distilled water was prepared and left to stand for 1 hour at room temperature. The pH of the mixture was then measured in triplicates using a pH meter (Orion *Versastar Pro* from *Thermo Scientific*).

# **Residual Cyanide:**

The residual cyanide level was determined using the picrate paper method adopted from Egan and colleagues [21]. Cassava flour samples (100 mg) and deionized water (1 ml) were added to containers containing an enzyme paper disc (linamarase + phosphate buffer at pH=8). A yellow indicator paper (picrate paper) was inserted just above the level of the liquid and the container was capped. Positive and negative controls were run with each sample. The negative control did not contain a cassava flour sample, the positive control utilized a pink standard cyanide paper disk in place of the cassava flour sample. The samples were left to stand for 24 hours at 31 °C. Picrate indicator papers from the samples were then eluted in 5 mL deionized water and the absorbance of the resulting solutions were measured against the negative control at 510 nm on a Spectrophotometer (*Cary 60 UV-Vis MY1 7260006*). The total cyanide content of each sample was calculated using the equation:

$$Total\ cyanide(ppm) = Abs_{sample} - Abs_{blank}\ x\ 396$$

where  $Abs_{sample}$  is the absorbance of the sample and  $Abs_{blank}$  is the absorbance of blank (negative control)

#### RESULTS AND DISCUSSION

#### **Desirable Chemical Characteristics of Cassava Flour**

Cassava flour much like wheat flour and other flours used for food preparation are defined by extended shelf-life due to low moisture and fat contents. Conversely, high protein, crude fiber, starch and ash contents are desirable for nutrition purposes. The low moisture content inhibits the growth of microorganisms, reduces the activity of proteolytic and lipolytic enzymes resulting in high protein and high fat contents and provides more efficient processing and handling of the flour. The low fat content of flours enhances the shelf-life since reduced lipid content results in reduced lipolytic oxidation and consequent rancidity of the flour. High crude protein content provides an appreciable source of protein particularly for vegans and also increases functional properties of the flour such as water absorption, cohesiveness, viscoelasticity, dough strength, texture, loaf volume, and crumb grain [22], [23]. Cassava is used as a staple in several communities in the tropics because of its high starch and fiber contents so these components are needed for a high nutrient-value product. The ash content refers to the minerals and inorganics left after the moisture and other volatile compounds have been

removed from the food sample due to heating to very high temperatures. The ash content is therefore a measure of the mineral content of the flour so high ash content is directly related to higher mineral nutrition value of the flour. In addition to the characteristics which define food value in cassava flour, attention is given to the cyanide content since cassava is well known for its high content of cyanogenic glycosides and the toxicity of these compounds are well documented [24], [25], [26], [27]. Consequently, cassava flour for food consumption should have low cyanide contents, the WHO recommends concentrations less than 10 mg of cyanogenic glycoside per kg of cassava flour [28].

## **Chemical Characteristics of Bitter and Sweet Varieties**

The cassava flour samples were comprised of mainly starch and moisture. The flours produced from bitter and sweet varieties possessed similar concentrations of starch, moisture, crude fiber and ash. However, cassava flour produced from the sweet varieties had significantly lower quantities of cyanide (70% less) and fat (30% less) than its counterpart which possessed significantly higher protein content (63% more) (Table 1). The noted characteristics of the flours derived from sweet varieties along with their apparently lower moisture contents indicate sweet cassava as the preferred choice for producing cassava flour. Chemical characteristics of specific varieties of the sweet varieties are highlighted below.

Table 1: One-way ANOVA for class of cultivar on chemical characteristics of Cassava

	Mean $\pm$ SD (g/100g)		F-value	p-value
	Bitter	Sweet		
Moisture content	11.77 ± 3.26	10.48 ± 4.32	0.511	0.485
Protein content	1.79 ± 1.01	2.92 ± 1.94	2.39	0.142*
Fat content	$2.32 \pm 1.08$	1.62 ± 0.43	3.22	0.092**
Starch content	63.94 ± 5.52	59.85 ± 6.44	2.10	0.167
Crude Fiber content	$2.31 \pm 0.53$	$2.13 \pm 0.40$	0.629	0.439
Ash content	$2.02 \pm 0.49$	2.18 ± 0.39	0.572	0.460
Cyanide content (mg/kg)	84.24 ± 71.95	12.41 ± 11.96	8.73	0.009**
pH (no unit)	6.73 ± 0.57	6.76 ± 0.17	0.025	0.876

<sup>\*</sup>denotes variations in the parameter are significant at the 85% confidence interval

# **Moisture Content**

Micro-organisms naturally present in flour start to grow and multiply at high moisture contents, resulting in undesirable odors and flavors [29]. The FAO recommends a maximum moisture content of 13% w/w for cassava flour [30]. Lower moisture contents cause reduced activity of proteolytic and lipolytic enzymes leading to slower breakdown of proteins and fats respectively. While high protein content is desirable for nutritional value and functional properties of flour such as increased water absorption, foaming and gel-forming capacities [22], [23]. High fat content in flours may lead to reduced shelf-life caused by the oxidation of fats to fatty acids and consequent rancidity, it is therefore desirable for cassava flour to possess inherently low fat contents. High moisture content of flours negatively affects processing and handling techniques since wetter flours agglomerate more readily, causing hoppers and conveyors to block resulting in laborious cleaning efforts and wasteful downtimes [23]. Two of the six varieties studied (one

<sup>\*\*</sup>denotes variations in the parameter are significant at the 90% confidence interval

<sup>\*\*\*</sup>denotes variations in the parameter are significant at the 95% confidence interval

from each class) had moisture content slightly greater than the maximum threshold as stipulated by the FAO (Tables 2 and 3).

Although the mean moisture content of the sweet cassava varieties appeared to be lower than the mean moisture content of the bitter varieties, this difference was not statistically significant (Table 1). Nonetheless, the sweet cultivar - ALBS had the lowest moisture content of all six varieties, a value that was significantly lower (at the 90% confidence interval) than that of Yardie - a sweet cultivar (Figure 1). The ALBS cultivar is therefore an appropriate choice for making cassava flour and other value-added cassava products which require low moisture contents. The extended shelf-life expected from these products would have a positive impact on food security by reducing post-harvest waste and scarcity when cassava crops are affected by pests or natural disasters.

# **Crude Protein Content**

Crude protein is a measure of the total nitrogen present and it approximates the amount of protein in food samples. The levels of protein in flours impact functional properties of dough such as dough elasticity, water absorption, mixing tolerance, gas retention, and texture. Highprotein flour form strong gluten networks, ideal for bread and chewy products, while lowprotein flours create tender, crumbly textures suitable for cakes and pastries [31], [32]. The cassava root from which flours are made has very low quantities of protein values [33], [34], most notably the gluten protein is absent and this has negative impacts on the organoleptic properties of the baked product. However, these properties may be improved by increasing the percentage of wheat flour used in cassava-wheat composites. In the cassava flours examined, the mean protein content was 2.35% ± 1.61% but cassava flours made from sweet varieties had significantly higher crude protein content, at the 85% confidence interval, than the flours made from the bitter varieties (Table 1). Additionally, cassava flour made from ALBS had significantly higher protein content (at the 90% confidence interval) than the cassava flours made from the other two sweet varieties (Figure 1). There was an inverse correlation (R = -0.667\*) between moisture content and protein content among the flours made from the sweet varieties where flour derived from ALBS had the highest protein content and lowest moisture content (Figure 1). These characteristics reinforce the ALBS cultivar as an appropriate choice for cassava flour production.

# **Fat Content**

Flours made from cereals, grains and vegetable roots are generally not significant sources of fat. The very low fat content of cassava flours enhances the shelf-life of the flours [33], [34]. The fat present in flours are typically long-chain organic compounds with C=C double bonds and carboxylate functional groups. These C=C double bonds are very reactive and get oxidized to ketones, aldehydes and short-chain fatty acids resulting in "sour" taste and rancid odors. This process is facilitated by the enzymatic action of lipoxygenase and formation of the intermediate lipid hydroperoxide compounds. The flours produced by both bitter and sweet varieties of cassava had mean fat content  $1.97 \pm 0.88$  g/100g. These values were higher than proximate crude fat values reported in other studies [35], [36]. This may be a cultivar-specific trait since varieties used in this research also presented lower starch contents than findings in other studies [35], [37], [38].

Further, flours produced by the sweet varieties had significantly lower crude fat content (at the 90% confidence interval) than flours produced by the bitter varieties (Table 1) however, there were no significant differences in the crude fat content among sweet varieties. This characteristic presents cassava flour as a low-fat dietary option.

#### **Starch Content**

Cassava tubers and by extension products made of cassava flour are consumed mainly for their carbohydrate value. Starch is the major form of carbohydrate in cassava flour and it is also the most abundant nutrient present, typically accounting for more than 80% of the flour by weight [39], [40], [41]. The complex carbohydrate nature of starch makes it a good source of sustained energy since during digestion it is slowly broken down by amylose into simpler carbohydrates like glucose and fructose. The slow release of the simple carbohydrates reduces the incidences of spikes in blood sugar levels which are precursors to insulin resistance and type II diabetes. The cassava flour samples in this study had a mean starch content of  $61.89\% \pm 6.18\% \text{ w/w}$  which was similar to the findings of [42] but much lower than other studies [35], [37], [38] all of which reported mean starch content greater than 80%. There were no significant differences in the mean starch content between flours made from the bitter and sweet varieties (Table 1), nor were there any significant differences in starch contents of the various flours.

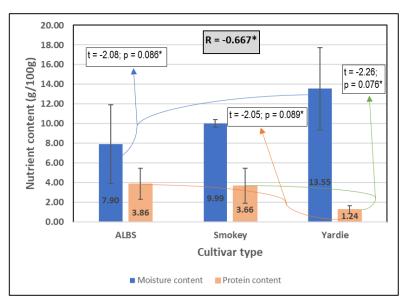


Figure 1: Moisture and protein contents in cassava flours made from the sweet varieties

#### **Crude Fiber Content**

Fiber is an indigestible complex carbohydrate found in plants. Foods with high fiber content aid in digestion, appetite suppression and overall improved gut health. The fiber content of cassava flour is highly dependent on the processing technique; flours that include the skin have much higher fiber content (2.8%) than those that do not (1.8%) [43], also flours that passed through smaller sieves have lower fiber contents than those that passed through larger sieves or those that were not sieved [42]. The cassava flours prepared in this research did not include the skins and yielded mean crude fiber content of 2.22%  $\pm$  0.47% w/w - a value consistent with those

reported by other researchers. There were no significant differences observed in the fiber contents of the flours investigated nor between flours derived from bitter varieties versus sweet varieties.

Table 2: Proximate Analysis of the three 'sweet' cassava varieties

	ALBS	Smokey	Yardie
Moisture (%)	7.90 ± 3.99	9.99 ± 0.39	13.55 ± 4.18
Protein (%)	3.86 ± 1.58	3.66 ± 1.78	1.24 ± 0.39
Fat (%)	1.63 ± 0.43	$1.67 \pm 0.43$	1.57 ± 0.36
Crude Fiber (%)	$2.08 \pm 0.27$	2.14 ± 0.29	2.19 ± 0.53
Ash (%)	2.04 ± 0.22	2.11 ± 0.47	2.38 ± 0.26
Starch (%)	58.30 ± 5.88	60.43 ± 6.94	60.80 ± 4.92

Table 3: Proximate Analysis of the three 'bitter' cassava varieties

0111011111111				
	LPL67	GAA98	WS13	
Moisture (%)	11.32 ± 0.30	13.31 ± 3.34	10.68 ± 3.64	
Protein (%)	2.03 ± 0.95	2.25 ± 0.94	1.09 ± 0.41	
Fat (%)	1.95 ± 0.87	$2.74 \pm 0.78$	2.29 ± 1.20	
Crude Fiber (%)	2.50 ± 0.65	$2.16 \pm 0.03$	$2.17 \pm 0.41$	
Ash (%)	1.88 ± 0.37	2.17 ± 0.41	$2.01 \pm 0.53$	
Starch (%)	68.47 ± 3.24	58.88± 3.57	64.47 ± 3.38	

#### Ash Content

The ash content represents the mineral content of the food sample. The most common minerals present in foods are calcium, magnesium, sodium and potassium and to lesser extents, manganese, zinc, iron and a few other trace minerals. Minerals are essential micronutrients that are needed for various physiological functions including nerve transmission, hormone regulation, enzyme activation and immune system support. The ash contents of the cassava flours ranged between 1.88% and 2.18% with a mean value of 2.10 %  $\pm$  0.43% w/w. These values all fell within the FAO standard of a maximum 3% for ash content in cassava flour and no significant differences were observed between flours made from different varieties. A 2019 study on the proximate compositions and functional properties of cassava flour in Ghana revealed 2.3% to 3.58% ash content [37]. The researcher reported that the values were influenced by the drying method used in the preparation of the flour; oven-dried flours had the highest ash content due to the higher temperatures that they were exposed to. This caused greater removal of moisture and resulted in higher nutrient concentrations.

# **Cyanide Content**

Cassava is arguably the most known plant which contains cyanogenic glycosides. The cyanogenic glycosides are hydrolyzed in a two-step, enzyme-catalyzed reaction called cyanogenesis to yield cyanide which is considered an anti-nutrient due to its toxic effect on the human body [25]. Cassava and cassava products can still be safely eaten provided that the method employed for processing the cassava removes sufficient cyanide to give residual cyanide of less than 10 mg/kg as recommended by the FAO [30]. The residual cyanide in cassava products is a function of the cyanide content in the fresh tuber, the processing method

used [44], total processing time and the number of processing techniques employed [17]. Common methods used include boiling, frying, fermentation, steaming, baking, air-drying and combinations of two or more of these. The more extreme processing conditions, such as those which include high temperature heat treatments have greater effect on reducing the level of cyanide in the food product particularly if the cassava was previously ground to create a high surface area. The residual cyanide levels of two of the three flours made from the sweet varieties of cassava had cyanide levels within the FAO limits of 10 mg/kg. However, none of the flours made from the bitter varieties fell within this range (Figure 2) indicating that changes in the processing of the flours from these varieties are needed. Longer soak times along with increased drying times may be able to bring these values in alignment with the permissible levels required. The ALBS cultivar which showed the most desirable moisture and protein contents had a cyanide level 150% greater than the permissible level as stipulated by the FAO. Conversely the Yardie cultivar which had the lowest residual cyanide level also possessed the lowest protein content and a moisture content marginally higher than the recommended 13%. The Yardie cultivar could therefore benefit from longer drying times to remove more moisture while the ALBS cultivar could benefit from longer soak times and increased drying times to promote greater reduction of cyanide content.

#### CONCLUSION AND RECOMMENDATIONS

A comparison of the chemical properties of cassava flours produced from six varieties is the first of this kind of research in Guyana. The research found that cassava flours made from both the bitter and sweet varieties had similar pH and starch, moisture, crude fiber, ash contents. However, cassava flours made from sweet varieties had lower crude fat and cyanide contents and higher protein content.

One significant relationship amongst chemical characteristics of the cassava flours was found; an inverse correlation between moisture content and protein content of the sweet varieties. The chemical characteristics of the flours were largely compliant with FAO and WHO regulations, exceptions include cyanide content in the three bitter varieties and one sweet variety exceeding the 10 mg/kg limit and crude fiber content of all varieties exceeding the 2.0% limit. Additional research into the physicochemical and functional properties must be completed before cultivar/s best suited for flour production can be identified. Despite the absence of the aforementioned data, the data collected in this study is valuable as it can be the first added to a repository of such data for the many cassava varieties that are known to be cultivated in Guyana. This data can then be used to produce cultivars with enhanced nutrient profiles. Currently there is no existing local or regional standard for cassava flour. Given that cassava flour production and export is projected to increase, the results of this research can also be used to create product specifications in the development of such a standard.

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