ORIGINAL RESEARCH

Effects of Processing Methods on Antinutrient Contents of Cassava Flour of TME419, IBA980581 and CR36-5 Varieties.

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Abstract

This study investigated the effects of processing methods on the anti-nutrients (cyanide, phytate and oxalate) contents of three cassava varieties: TME419, IBA980581, and CR36-5. The research compared the effects of air-drying, sun-drying and unprocessed cassava pulp with the aim of evaluating their impact on anti-nutrient levels of the products. Results showed that unprocessed cassava roots had the highest concentration of anti-nutrients across all varieties. Both air-drying and sun-drying significantly reduced cyanide levels in TME419 variety, with no significant difference between the two methods. However, air-drying was more effective in reducing phytate and oxalate concentrations compared to sun-drying, particularly in the IBA980581 and CR36-5 varieties. These findings revealed that air-drying is a preferable method for improving the nutritional quality of Cassava flour by reducing anti-nutient levels. The study highlights the importance of selecting appropriate processing methods to enhance the safety and nutritional value of cassava-based products.

Key words: Anti-nutrients, Processing, CR36-5, IBA980581, TME419.

Introduction

Cassava (*Manihot esculenta*), a root crop, has long been recognized as a vital food source in tropical regions, particularly in Africa, Asia, and South America (Food and Agriculture Organization, 2022). It is rich in carbohydrates and serves as a primary calorie source for millions of people worldwide (Nweke

and Tetteh, 2023). However, early studies by Ubalua and Ofoedu (2023) revealed that cassava contains elevated levels of antinutrients such as cyanogenic glycosides (notably linamarin), oxalates, and phytates, which pose serious health risks to consumers (Olusegun and Ibrahim, 2024).

To mitigate these risks, traditional processing techniques such as peeling, boiling, drying, and milling have been employed to reduce antinutrient levels (Adebowale and Oluwole, 2024). Milled cassava flour, which extends shelf life, is widely incorporated into various foods including bread, porridge, and baked goods (Balogun et al., 2023). Research by Alamu et al. (2021) identified variations in antinutrient levels among different cassava varieties, suggesting that processing methods may have differing effects depending on the variety.

Despite previous efforts to explore the effects of processing methods on cassava varieties, there remains a lack of information comparing these effects among recent hybrid varieties such as TME419, IBA980581, and CR36-5. Balogun et al. (2023) further highlighted natural differences in antinutrient content among cassava varieties, indicating that some varieties inherently contain lower levels of harmful compounds.

By investigating how various processing methods affect antinutrient content across different cassava varieties, this study aims to enhance the safety and nutritional value of cassava flour. Such improvements could lead to better cassava-based diets, especially in resource-constrained areas (Oni and Akinmoladun, 2024).

However, traditional processing methods often fail to consistently reduce cyanide and other antinutrients in cassava flour to the safe levels recommended by WHO/FAO, posing a significant public health concern. Therefore, it is important to identify and optimize processing techniques that reliably detoxify cassava flour to meet established safety standards for consumption.

Consequently, this study will provide valuable insights into optimizing cassava processing techniques and identify varieties with lower antinutrient levels, facilitating the production of safer and more nutritious flour (Udoro et al., 2021). Specifically, it aims to investigate how various processing methods of cassava tubers into flour affect the levels of cyanides, oxalates, and phytates in three different cassava varieties.

Materials And Methods

Collection and Identification of Plant Sample

Three varieties of *Manihot esculenta* roots, each with an average weight of 4000 ± 0.5 g, were sourced from the Kogi State Network of Cassava Seed Entrepreneurs' farm, which is part of the Basics II project supported by IITA and Catholic Relief Services (CRS). These roots were collected in September 2024 at GPS coordinates 7.5521774, 6.6155641 (Ajaokuta; Kogi Central; North Central, Nigeria). The varieties were identified by the President of the above organization Mr. Sumaila Onudoga Mohammed as TME 419, IBA980581 (Dixon) and CR36-5 (Ayaya: Beautiful).

Sample preparation

The flour preparation was carried out according to the methods outlined by Lagnika et al. (2019) with some modifications. 4 kg tubers of each cassava variety were thoroughly cleaned to remove any dirt. The pinkish outer layer was then peeled off to expose the white flesh. After peeling, the cassava tubers were sliced into uniform chips of about 1.5 mm to speed up the drying process. The sliced chips were divided into two equal portions of 1500 g each. One portion was air-dried at room temperature (32°C) for seven days, while the other portion was sun-dried without fermentation for approximately 3 to 5 days. Once the drying process was completed, the samples were crushed using a clean mortar and pestle, then stored in an airtight container to maintain their quality until when needed for analysis. The unprocessed pulverized tubers served as control for the study.

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Quantification of cyanide content

The cyanide contents of the samples were determined using the alkaline Picrate method outlined by

Ikediobi et al. (1980). The cassava flour samples were homogenized and treated with hot 20%

hydrochloric acid (HCl) solution to release hydrogen cyanide vapors. These vapors reacted with alkaline

picrate test strips, resulting in the formation of a red-colored complex on the strips. The red complex was

then extracted using 50% ethanol solution, and the absorbance of the extract was measured at 510 nm

using a spectrophotometer.

Quantification of phytate content

Phytate concentration in the samples was determined using the method described by Haug and Lantzsch

(1983). First, 2 g of the sample was weighed into a clean conical flask, and 100 ml of 2% concentrated

hydrochloric acid (HCl) was added. The mixture was allowed to stand for 3 hours before being filtered

through double-layered hardened filter paper using a funnel, and the filtrate was collected.

Next, 50 ml of the filtrate was transferred into a clean conical flask, to which 107 ml of distilled water

and 10 ml of 0.3% ammonium thiocyanate solution (used as an indicator) were added. This solution was

then titrated against a standard iron (III) chloride solution. The phytate concentration was subsequently

calculated using the appropriate formula:

% Phytic acid = Titer value $\times 0.00195 \times 1.19 \times 100 \times 3.55$

Weight of sample

Ouantification of oxalate content

Oxalate content was determined using the colorimetric method described by Keith and Obzansky (AOAC, 1984).

One gram of each sample was weighed into a clean conical flask, and 10 ml of distilled water was added. Then, 1

ml of concentrated sulfuric acid (H₂SO₄) was added, and the mixture was allowed to stand for 1 hour. The volume

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was subsequently adjusted to 50 ml with distilled water.

Thereafter, 25 ml of the extract was measured and warmed to 90°C before being immediately titrated against 0.1M potassium permanganate solution. The endpoint was indicated by a persistent color change, and the burette reading was recorded once the color remained steady for several seconds. The oxalate concentration in the sample was calculated by multiplying the burette reading by 11.5.

Statistical Analysis

The data pooled in triplicates was analyzed using one-way ANOVA with Minitab statistical version 23 software. Significant means were separated using a post hoc at P < 0.05 significance level. Experimental results were expressed as mean \pm standard error of the mean (SEM)

Results

The results of this study revealed a significant difference in cyanide concentrations between unprocessed TME 419 cassava roots and non-fermented, air-dried, or sun-dried cassava flour (P < 0.05). However, no significant difference was found between the two processing methods (non-fermented/air-dried and non-fermented/sun-dried) as shown in table 1.

Regarding phytate concentration, the study found a notable difference between fresh/unprocessed TME 419 cassava and the processed flour. Unprocessed cassava exhibited a higher phytate concentration (5.20%) while the non-fermented/air-dried flour had a lower concentration (0.298%) compared to the non-fermented/sun-dried flour (0.62%). The findings regarding oxalate concentrations in TME 419 cassava followed a similar trend to those of phytate as shown in table 1 below.

The result of this present research revealed that the cyanide concentration in the IBA980581 variety exhibited a pattern consistent with that of the TME 419 variety, as reported in Table 1. Phytate showed a distinct variation between the unprocessed cassava roots and the two processing methods in this study. The unprocessed cassava had higher phytate concentrations (5.803%) compared to non-fermented air-

dried flour (0.412%) while the non-fermented air-dried flour was significantly higher in phytate concentration compared to the non-fermented sun-dried flour (0.308%). Oxalate, another anti-nutrient found in cassava, exhibited a different trend in the IBA980581 variety compared to TME 419. In this study, the fresh/unprocessed IBA980581 showed significantly higher oxalate concentrations (0.411%) than those observed in the processed forms. Interestingly, the non-fermented, air-dried IBA980581cassava flour had higher oxalate levels (3.454%) compared to the non-fermented, sun-dried cassava flour (2.26%).

The findings of this study provide valuable insights into the effects of processing on the concentration of anti-nutrients in the CR36-5 cassava variety, specifically cyanide, phytates, and oxalates. The cyanide concentrations in the CR36-5 variety followed a similar pattern to that of the TME 419 variety as in Table 1. Similarly, the cyanide content in CR6-5 showed a comparable pattern, suggesting that like TME 419, the cyanide levels in this variety could be effectively reduced with appropriate processing methods. In contrast, as reported in Table 2, the IBA980581 variety showed different trends, likely due to genetic or environmental factors influencing cyanogenic glucoside levels in that variety. Phytate concentrations in the unprocessed CR6-5 cassava were higher (5.501%) compared to the processed cassava flours, as was typically observed in other cassava varieties. This study found a significant difference in phytate concentrations between fresh roots and processed flours, with processed cassava flours showing lower phytate concentrations. Additionally, the comparison between the two processing methods revealed a higher phytate concentration in sun-dried cassava flour (0.292%) compared to air-dried cassava flour (0.196%). The oxalate concentrations in the fresh, unprocessed CR36-5 cassava were higher compared to processed flour (68.2%). In this study, there was also a significant difference between the two processing methods. Sun-dried cassava flours had higher oxalate concentrations (68.2%) compared to

air-dried cassava flours (3.432%). This suggests that the drying method can influence the oxalate reduction process, with air drying potentially being more effective at reducing oxalate levels.

Discussion

This study examined the impact of non-fermented air-drying and sun-drying on the antinutrient content of three cassava varieties: TME 419, IBA980581, and CR36-5. For TME 419, significant reductions in cyanide concentrations were observed when comparing unprocessed roots to processed flours, with both air-drying and sun-drying maintaining similar cyanide levels. This aligns with Bradbury et al. (2006), confirming that drying methods effectively reduce cyanide, primarily present as toxic cyanogenic glucosides (linamarin and lotaustralin) (Egan et al., 2018).

Phytate concentrations in TME 419 were highest in fresh roots and decreased after processing. Air-dried flour exhibited lower phytate levels than sun-dried flour, suggesting that air-drying promotes phytate breakdown, possibly through enhanced phytase activity under favorable environmental conditions (Mbithi-Mwikya and Odeny, 2002). Conversely, sun-drying's higher temperatures may cause the formation of insoluble phytate complexes, limiting phytate reduction (Raboy, 2009).

Similarly, oxalate levels followed this trend: fresh roots had the highest concentrations, with air-dried flour showing greater oxalate reduction compared to sun-dried flour. This supports findings by Lado et al. (2016), indicating that air-drying more effectively lowers oxalate content, enhancing calcium bioavailability (Singh et al., 2009).

In the IBA980581 variety, cyanide reduction patterns mirrored those of TME 419, with both drying methods yielding similar detoxification results (Okafor and Ngoddy, 1986). However, phytate and oxalate responses differed. Sun-drying reduced phytate levels more effectively than air-drying, consistent with Okello et al. (2007), likely due to enzymatic breakdown at higher temperatures. For oxalates, sun-dried samples had lower concentrations than air-dried ones, supporting reports by Van der Haar et al.

(2003) and Gomez et al. (2017) that sun exposure facilitates oxalate degradation through heat and sunlight.

The CR36-5 variety showed cyanide trends similar to TME 419, with no significant difference between air- and sun-drying methods (Eyinla et al., 2019). Phytate levels were higher in fresh roots and reduced after processing; however, sun-dried flour retained more phytates than air-dried flour, aligning with Gomez et al. (2017) who noted sun drying's limited efficacy in phytate reduction. This may be due to the higher temperatures in sun drying inhibiting phytate breakdown compared to slower drying or fermentation (Ugwu et al., 2019).

Oxalate concentrations in CR36-5 were also highest in fresh roots. Interestingly, air-dried flour had lower oxalate levels than sun-dried flour, suggesting air-drying's slower dehydration at lower temperatures may better facilitate oxalate degradation. This contradicts some previous findings (Gomez et al., 2017) and highlights the complexity of oxalate breakdown, which depends on temperature, drying duration, and humidity.

In summary, this study emphasized the importance of selecting appropriate processing methods tailored to cassava varieties to optimize antinutrient reduction. Air-drying appears preferable for reducing phytates and oxalates in TME 419 and CR36-5, while sun-drying may be more effective for phytate and oxalate reduction in IBA980581.

Conclusion

This study evaluated the effects of non-fermented air-drying and sun-drying on cyanide, phytate, and oxalate levels in three cassava varieties: TME 419, IBA980581, and CR36-5. Both drying methods effectively reduced cyanide concentrations across all varieties, with similar detoxification patterns observed. However, phytate and oxalate reductions varied by variety and method: air-drying was more effective for TME 419 and CR36-5, while sun-drying better reduced these antinutrients in IBA980581.

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These findings highlight the need to tailor processing techniques to specific cassava varieties to optimize nutritional quality and safety.

Author's Contribution:

AE conceptualised the research . AE, OJM handled the data curation while statistical analysis was done by AE. The experiments were conducted by AE, EIJ, SVD, AM ASO and MSO. All authors contributed to the development of the final manuscript and approved its submission.

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Table 1: Effects of Processing Methods on Cyanide, Phytate, and Oxalate Levels in Cassava Flour from Tme419 Variety

VARIETY	TYPE OF PROCESSING	CYANIDE	PHYTATE (%)	OXALATE
		(mg/kg)		(mg/100g)

TME 419 TME 419	Fresh roots without processing Non-fermented air-dried flour	$2.501 \pm 0.003^{b} \\ 0.081 \pm 0.002^{a}$	5.202 ± 0.001^{c} 0.298 ± 0.013^{a}	0.600± 0.002 ^a 2.12 ±0.130 ^b
TME 419	Non-fermented sun-dried flour	0.076 ± 0.002^{a}	0.62 ± 0.015^b	3.46 ± 0.041^{c}

Means on the same column with different letter superscripts are significantly different (p<0.05).

Table 2: Effects of Processing Methods on Cyanide, Phytate, And Oxalate Levels in Cassava Flour from **Iba980581variety**

VARIETY	TYPE OF PROCESSING	CYANIDE (mg/kg)	PHYTATE (%)	OXALATE (mg/100g)
IBA980581	Fresh roots without processing	2.715 ± 0.013^{b}	$5.803 \pm 0.005^{\circ}$	0.411 ± 0.013^{c}
IBA980581	Non- fermented air-dried flour	0.080 ± 0.001^{a}	0.412 ± 0.008^{b}	3.454 ± 0.011^{b}
IBA980581	Non- fermented sun-dried flour	0.101 ± 0.001^{a}	0.308 ± 0.022^{a}	2.260 ± 0.114^{a}

Means on the same column with different letter superscripts are significantly different (p<0.05).

Table 3: Effects of Processing Methods on Cyanide, Phytate, and Oxalate Levels in Cassava Flour from Cr36-5 Variety

VARIETY	TYPE OF PROCE	ESSING	CYANIDE (mg/kg)	PHYTATE (%)	OXALATE (mg/100g)
CR36-5	Fresh roots processing	without	5.412 ± 0.105^{b}	5.501 ± 0.101^{c}	$68.2 \pm 0.100^{\circ}$
CR36-5	Non-fermented flour	air-dried	0.090 ± 0.001^{a}	0.196 ± 0.011^{a}	3.432 ± 0.023^{a}
CR36-5	Non-fermented flour	sun-dried	0.1006 ± 0.001^{a}	0.292 ± 0.008^{b}	4.5000.100 ^b

Means on the same column with different letter superscripts are significantly different (p<0.05).