

Effect of Processing on the Retention of Total Carotenoid, Iron and Zinc Contents of Yellow-fleshed Cassava Roots

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Abstract BACKGROUND: It was envisaged that processing of yellow-fleshed cassava roots might affect the micronutrient composition. Hence, three yellow-fleshed cassava roots were grown for 12 months in a randomized complete block design with three replications at Ibadan, Nigeria, to evaluate the effects on total carotenoid, iron, and zinc retention after processing the roots. Raw and processed storage roots were analyzed using standard methods. Percentage true nutrient retention was calculated using the concentration of each parameter adjusted for changes in weight. RESULT: There were significant genotypic differences ($P < 0.01$) for all the evaluated characteristics. The mean total carotenoid concentration of the unprocessed storage roots was 4.90 $\mu\text{g/g}$, mean iron content was 7.47 mg/kg, and mean zinc content was 8.95 mg/kg. The concentration after processing varied depending on the product. Results indicated that boiled cassava retained the highest amount of iron and zinc, also of total carotenoid (73.5%) This was followed by *gari* (44.9%) and raw *fufu* (40.8%); cooked *fufu* had the lowest (21.5%). CONCLUSION: Processing cassava storage roots resulted in a significant reduction in micronutrient retention and this depended on the processing method and genotype.

Keywords: carotenoid, iron, zinc, processing, cassava, nutrient retention

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1. Introduction

Vitamin A deficiency, particularly in developing countries, is still a major problem leading to blindness and childhood mortality [1]. The requirement for this vitamin can be satisfied either by animal food products containing preformed vitamin A or plant food products containing provitamin A carotenoid [2].

Carotenoids are major sources of vitamin A in the diets of a large proportion of the world's population. Approximately 600 carotenoids are found in nature but only three are important precursors of vitamin A in humans: β -carotene, α -carotene, and β -cryptoxanthin. Of these, β -carotene is the major provitamin A component of most carotenoid-containing foods [3]. Food composition tables are often used to estimate the amount of a provitamin A carotenoid in foods. Only recently have accurate values for food carotenoids been determined [4,5,6]. Because varieties, growing conditions, and processing methods differ in various localities, the actual content of carotenoid in a food may differ significantly from that reported in food composition tables [7].

Data on the retention or loss of carotenoid are somehow conflicting and often difficult to interpret because processing conditions are not described in detail and different foods are processed differently, making comparison of processing methods difficult, and the

procedure followed for calculating losses is not specified or the calculation is faulty [7,8].

Cassava (*Manihot esculenta* Crantz), variously known as manioc, mandioc, tapioca, or yucca, is grown principally for its swollen roots but its leaves are also consumed in some developing countries. The roots contain 25-35% starch and the leaves have a significant amount of proteins, vitamins, and minerals [9]. Currently, about half of the world's production comes from Africa [10] where total consumption more than doubled from 24 million t/year (1961 to 1965) to 58 million t/year (between 1994 and 1998) after accounting for waste [9].

From a nutritional point of view, cassava's chief advantage is that it is a source of cheap carbohydrate. The roots contain negligible amount of protein [11] although substantial amounts are derived from the leaves. Enhancement of cassava's nutritive value had not been given serious consideration, either in breeding or in processing research. However, since recent research by the Consultative Group on International Agricultural Research (CGIAR) Centers and international partners have focused on increasing the protein, mineral, and vitamin contents of the storage roots, the effect of certain processing techniques in reducing or enhancing these contents has gained importance. The storage roots have a very short shelf-life. Rapid postharvest physiological deterioration often begins within 24 h and hence the crop needs to be consumed shortly after harvest or processed

into more stable products, such as fermented paste (*fufu*), roasted granules (*gari*), chips, or unfermented flour.

Therefore, a study was undertaken to (a) determine total carotenoid, iron, and zinc contents in raw and processed storage roots, and (b) evaluate nutrient retention during the processing of selected elite yellow-fleshed clones into boiled roots, raw and cooked *fufu*, and *gari*, using traditional processing methods.

2. Materials and Methods

2.1. Cassava Roots and Sampling

Three yellow-fleshed genotypes (01/1371, 01/1235, and 94/0006) designated as improved Tropical *Manihot* Selections (TMS), were grown under rain-fed conditions in a randomized complete block design with three replications at the research farm of the International Institute of Tropical Agriculture, Ibadan, Nigeria. Planting was done at the beginning of the rainy season (May/June). The varieties are tolerant to the major pests and diseases of cassava (mosaic, bacterial blight, anthracnose disease, and green spider mite) in sub-Saharan Africa. No fertilizer or herbicides were applied during the course of the experiment, and hand weeding was done when necessary. The roots were harvested at 12 months after planting and processed within 24 hr as described below.

To protect samples from oxidation, all sampling following harvesting was conducted in a dimly lit room in the shortest time possible. Raw samples were dipped in liquid nitrogen before packing and freezing. All samples (raw and processed) collected for laboratory analysis were packed in sterilized Whirl-Pak bags (made by Nasco Whirl-Pak, USA) and further wrapped with aluminum foil and stored at -80°C until laboratory analysis. Analyses of total carotenoid, iron, and zinc on the raw samples were completed within 7 days and on the processed products within 14 days.

2.1.1. Boiled Cassava

A representative sample of freshly harvested roots for each genotype was sampled as follows: three plants were harvested in each replication and at least five roots/plant were collected. In the laboratory, three roots (big, medium, and small sizes) were selected for each plant, washed thoroughly with potable water to remove dirt and adhering sand particles and air-dried. The storage roots were peeled manually using a stainless steel knife and quartered longitudinally from the stem end to the root end; two opposite sections from each root were taken, combined manually, cut into small chunks, and boiled in water for 35 min.

2.1.2. Raw *fufu* Processing

Ten kg of freshly harvested roots of each genotype in each replication were peeled manually using a stainless steel knife and washed thoroughly with potable water to remove dirt and adhering sand particles. The peeled and washed roots were cut into chunks about 15 cm long using a stainless steel knife and steeped in water in a plastic bowl for 5 days at room temperature under subdued light. After 5 days, the roots were taken out, broken by hand, and the fibers were removed by passing the mash manually through a muslin mesh sieve. This result was

allowed to sediment for 24 hr in a large plastic bowl. After sedimentation, the water was decanted and the sediment/fermented paste further washed with water. The fermented mash (*fufu*) was dewatered by being placed in a hessian sack, and pressed with a hydraulic press. The resulting wet paste was subdivided into two portions. One was used for laboratory analysis and the other for further processing to cooked *fufu*.

2.1.3. Cooked *fufu* Processing

A 300 g sample was mixed with 360 ml of water and cooked in a stainless steel pot (with continuous stirring with a wooden rod) for 20 min to obtain a sample of cooked *fufu*. The sample was packaged into polyethylene bags which were further wrapped with aluminum foil and stored at -80°C until laboratory analysis.

2.2. *Gari* Processing

Ten kg of freshly harvested roots of each genotype in each replication were peeled manually using a stainless steel knife and washed thoroughly with potable water to remove dirt and adhering sand particles. The peeled and washed roots were grated into a mash using a petrol engine-driven stainless steel grater, placed in a hessian sack, and left for 3 days to ferment at room temperature (covered with black nylon to prevent exposure to light). After 3 days, the fermented mash was dewatered using a hydraulic press, then sieved manually using a stainless steel sieve to pulverize the pressed cake and separate fibrous materials. The pulverized cake was roasted in a large, shallow stainless steel pan with constant stirring with a piece of stainless steel plate for 10 min. The resulting roasted granules (*gari*) were spread on a stainless steel tray to cool and then sieved to obtain a small particle size and packaged into polyethylene bags which were further wrapped with aluminum foil and stored at -80°C until laboratory analysis.

2.2.1. Determination of Total Carotenoid Contents

Total carotene contents of raw and processed products were determined spectrophotometrically as described in the HarvestPlus Handbook for Carotenoid Analysis [12].

2.2.2. Determination of Zinc And Iron Contents

Samples of raw and processed products for iron and zinc measurements were sent to Waite Analytical Services, Adelaide, Australia, for analysis. Iron and zinc contents were determined using Inductively Coupled Plasma Atomic Emission Spectrometry (ICPAES), and the method described by Zarcinas *et al.* [13]. A sample of 0.6 g of the ground material was cold digested in 50 mL tubes overnight using 11 mL of nitric/perchloric acid mixture (10:1) and made to a final volume of 25 mL. Aliquots of the digested samples were analyzed for iron and zinc using the ICPAES (made in Switzerland by ARL model 3580 B). Samples were analyzed in duplicate. Three random samples were selected from the total batch of samples and re-analyzed to check on the accuracy and reproducibility of the method.

2.2.3. Determination of True Nutrient Retention

Studies on retention require the pairing of the raw and cooked/processed samples so that sample variation does

not affect the analytical results. For each genotype in each replication, three roots (big, medium and small sizes) were selected, washed thoroughly with potable water to remove dirt and adhering sand particles and air-dried. The roots were peeled manually using a stainless steel knife and quartered longitudinally from the stem end to the root end. Two opposite sections from each root were taken and combined, manually cut into small pieces and mixed thoroughly. A sample was taken and analyzed immediately for total carotenoid, iron, and zinc contents. These measurements represented the concentration of the nutrients in the raw roots. The samples to be processed were weighed before and after processing.

True total carotenoid, iron, and zinc retention (% TR) was calculated as indicated below [14].

% True Retention

$$= \frac{\left(\frac{\text{Nutrient content / g of processed food}}{\text{×g of food after processing}} \right)}{\left(\frac{\text{Nutrient content / g of raw cassava roots}}{\text{×g of food before processing}} \right)} \times 100$$

2.3. Statistical Analysis

Data were subjected to analysis of variance and descriptive statistics using the Statistical Analysis System

software [15]. Means were separated using Fisher's protected least significance difference test at $P < 0.05$.

3. Results and Discussion

3.1. Total Carotenoid

Boiled roots: Table 1 represents total carotenoid in raw and boiled roots, raw and cooked *fufu*, and *gari* prepared from yellow fleshed genotypes and their percentage true carotenoid retention (%TR). Significant differences ($P < 0.01$) were observed for each of the evaluated characteristics (Table 1). The mean total carotenoid concentration of the raw roots was 4.90 $\mu\text{g/g}$ and ranged from 2.6 to 7.3 $\mu\text{g/g}$. After boiling, the mean total carotenoid concentration was lower than the mean concentration of the raw roots (Table 1). The genotype TMS 01/1371 had a higher concentration of total carotenoid than TMS 94/0006. Mean total carotenoid retention was 73.6% and ranged from 48.4 to 96%. The percentage true total carotenoid retention was highest for TMS 01/1235, intermediate for TMS 01/1371, and lowest for TMS 94/0006. The mean percentage true retention observed in this study is similar to that reported by Chavez et al. [24] who found a mean retention of 66% after evaluating 28 genotypes that were boiled for 30 min.

Table 1. Total carotenoids ($\mu\text{g/g}$ FW) concentrations and percentage true retention in selected cassava products prepared from yellow-fleshed cassava genotypes¹

Genotype	Raw Roots	Boiled roots	%TR ²	Raw <i>fufu</i>	%TR ²	Cooked <i>fufu</i>	%TR ²	<i>Gari</i>	%TR ²
TMS 01/1371	7.3	6.1	76.3	12.3	39.6	5.3	36.7	15.9	38.1
TMS 01/1235	4.8	4.8	96.0	9.1	37.1	3.0	15.0	10.8	49.8
TMS 94/0006	2.6	1.4	48.4	4.5	45.6	2.6	12.7	5.1	46.8
Mean	4.90	4.09	73.57	8.64	40.78	3.64	21.45	10.63	44.90
SE	1.34	1.40	13.81	2.26	2.50	0.84	7.65	3.11	3.51
P level	**	**	**	**	**	**	**	**	**

¹ Means of three replicates; ** = $P < 0.01$.

² = % TR- percentage true retention.

Uncooked *fufu*: When the raw roots were processed to uncooked *fufu*, there was a significant increase in total carotenoid for all the genotypes (Table 1). The mean total carotenoid concentration was 4.9 $\mu\text{g/g}$ in the raw roots and 8.6 $\mu\text{g/g}$ in the processed product. The mean percentage true total carotenoid retention was significantly higher for TMS 94/0006 than for TMS 01/1235. This result may suggest that losses of total carotenoid content may be dependent on the genotype and the concentration before processing as TMS 94/0006 had the lowest total carotenoid content in the raw roots.

Cooked *fufu*: When the raw fermented paste was further processed by cooking to thick dough, there was a significant reduction in total carotenoid from 8.6 $\mu\text{g/g}$ to 3.6 $\mu\text{g/g}$ (Table 1). The genotype TMS 01/1371 had the highest concentration and TMS 94/0006 had the lowest. After adjustments for weight and moisture changes during processing to cooked *fufu*, the mean percentage total carotenoid retention was 21.5%. Cooking time and temperature as well as cooking method may also affect nutrient loss; the higher the temperature and the longer the heat applied, the greater the loss [16]. Provitamin A carotenoids are easily destroyed by exposure to light and during processing, heating, and storage [7]. A combination of multiple preparation and processing methods is known

to result in substantial losses of provitamin A carotenoids [17], as observed in the present study.

***Gari*:** There was a significant increase in mean total carotenoid concentration, from 4.9 $\mu\text{g/g}$ in the raw roots to 10.6 $\mu\text{g/g}$ in the *gari*. The mean percentage true retention was 44.9% for total carotenoid with a range of 38.1 to 49.8%. The genotype TMS 01/1235 had an intermediate total carotenoid concentration in the raw roots but the highest retention of total carotenoid in the *gari* compared with TMS 01/1371 which had the highest total carotenoid content in the raw roots.

The retention of carotenoid decreases with a longer processing time and after a higher processing temperature, also when food is cut or pureed [8]. The effect of processing on nutrient content will depend on the sensitivity of the nutrient to the various conditions prevailing during the process, such as heat, oxygen, pH, and light. Carotenoids are destroyed by heat, and oxidize and isomerize when exposed to heat and light. In the present study, the low percentage retention indicates that substantial losses occurred in preparing raw *fufu*. However, concentration on a $\mu\text{g/g}$ basis is higher in raw *fufu* than in peeled and raw roots. This may be due to a greater extractability of carotenoid from processed samples, unaccounted loss of moisture, and leaching of soluble

solids during processing. The mean moisture content of the peeled raw roots was 80.5% and that of the raw *fufu* was 43.1%. Therefore, the observed high concentration of carotenoid in raw *fufu* may have been as a result of both moisture loss and exposure to light, especially during sieving.

Chemical transformations that occur in heat treatment involve isomerization and epoxidation of carotenoids and not their formation [8]. The observed increases in total carotenoid concentration during the processing of roots to *gari* could be due to the greater ease with which carotenoids in cooked or processed samples can be extracted compared with those in fresh foods where the carotenoids are physically protected and/or combined with other food components that impede solvent penetration and extraction [7]. The higher concentration may also be caused by unaccounted losses of moisture and soluble solids which would concentrate and increase the total carotenoid/unit weight of the processed food. The mean moisture content of the peeled raw roots was 74.5% and that of the *gari* was 12.5%.

Nutritional value is better defined by the concentration which remains once the crop has been processed before consumption. In Africa, cassava roots are usually processed into various products. The observed differences among the studied genotypes for carotenoid retention are in agreement with similar observations by Iglesias *et al.* [18] who reported that the stability of carotenes in response to different processing methods was genotype-dependent. In the present study, boiled cassava retained the highest amount of total carotenoid (73.5%) followed by *gari* (44.9%), raw *fufu* (40.8%), and cooked *fufu* (21.5%).

Cassava processing involves a combination of activities which are performed in stages. Such activities are (i) peeling; (ii) chipping, crushing, milling, slicing or grating; (iii) dehydration by pressing, decanting, or drying in the sun; (iv) fermenting by soaking in water, heaping, or stacking; (v) sedimentation; (vi) sieving; and (vii) cooking, boiling, toasting, or steaming. The number of steps required and the sequence vary with the product being made [19,20]. This sequence of activities may result in losses of total carotenoid through enzymatic and non-enzymatic oxidation at each stage of processing and may

be responsible for the any observed differences in nutrient retention of different cassava products.

The enzymatic and non-enzymatic changes occurring in carotenoids during processing have been reviewed by several authors [7,8,16,21,22]. Loss of carotenoids occurs during processing through physical removal (peeling), isomerization, and enzymatic or non-enzymatic oxidation [7]. Carotenoids are highly unsaturated and therefore are prone to isomerization and oxidation. Isomerization of *trans*-carotenoids to the *cis*-isomers is promoted by contact with acids, heat treatment, and exposure to light, resulting in some loss of color and alteration of biological activity [8].

In general, information on stability or loss of total carotenoid on cassava storage roots is limited; much has been done on total carotenoid and/or β -carotene in dark-green leafy vegetables. A large loss of carotene (31%) was observed in baked sweet potato [23]. When Chavez *et al.* [24] processed yellow-fleshed cassava genotypes into different products (boiled, oven-dried, sun-dried, shadow-dried flour, or *gari*) they reported that oven-drying, shadow-drying, and boiling resulted in the highest levels of retention of β -carotene; *gari* had the lowest. In another study conducted by Iglesias *et al.* [18] boiling resulted in 34% reduction in carotene content.

3.2. Iron Retention

Boiled roots: Table 2 revealed the iron concentration and its percentage true retention during the processing of raw roots to boiled cassava, raw and cooked *fufu*, and *gari*. Mean iron concentration was 7.47 mg/kg with a range of between 6.7 mg/kg and 8.5 mg/kg for raw roots. Chavez *et al.* [24] evaluated 601 genotypes and reported that storage roots averaged 9.6 mg/kg for iron content. The genotype TMS 01/1371 had a higher concentration of iron than TMS 94/0006. The percentage mean iron retention was 85.24% with a range from 79.4 to 96.4%. The genotypes TMS 94/0006 and TMS 01/1235 had a similar percentage true retention for iron and it was below the mean. The results for iron are in agreement with those observed by Bell [25] who reported that boiling produced a significant reduction in the amount of iron in different species of West African yam.

Table 2. Iron concentrations (mg/kg) and percentge true retention in selected cassava products prepared from yellow-fleshed cassava genotypes¹

	Raw Roots	Boiled roots	%TR ²	Raw <i>fufu</i>	%TR ²	Cooked <i>fufu</i>	%TR ²	<i>Gari</i>	%TR ²
TMS 01/1371	6.7	7.2	96.4	5.5	19.7	7.0	31.5	7.9	20.5
TMS 01/1235	8.5	7.1	79.4	8.9	21.6	8.5	39.8	8.5	22.3
TMS 94/0006	7.1	5.9	79.9	7.0	37.2	9.2	26.3	8.0	25.3
Mean	7.47	6.72	85.24	7.13	26.15	8.25	32.54	8.17	22.70
SE	0.55	0.40	5.59	0.98	5.54	0.65	3.94	0.20	1.39
<i>P</i> level	**	**	**	**	**	**	**	**	**

¹ Means of three replicates; ** = $P < 0.01$.

² = %TR- percentage true retention.

Uncooked *fufu*: Iron concentration of raw *fufu* was high in TMS 01/1235, intermediate for TMS 94/0006, and low for TMS 01/1371. The genotype TMS 94/0006 retained more iron than TMS 01/1371 and TMS 01/1235.

Cooked *fufu*: When the raw fermented paste was further processed by cooking to thick dough there was a slight increase in mean iron concentration from 7.1 mg/kg to 8.3 mg/kg. This slight increase may be due to a

concentration factor or to contamination through the water used for cooking. After adjustment for weight and moisture changes during the processing to cooked *fufu*, the mean percentage retention was 32.5% for iron (Table 2). Cooking time and temperature as well as cooking method can also affect nutrient loss; the higher the temperature and the longer the heat is applied, the greater the loss [16].

Gari: There was a slight increase in iron concentration in the processed product. The mean percentage true retention for iron was 22.7% with a range from 20.5 to 25.3%. Genotype TMS 01/1235 had intermediate retention for iron compared with TMS 01/1371.

Nutritional value is better defined by the concentration which remains once the crop has been processed before consumption. In the present investigation, boiled cassava retained more iron (85.2%) followed by cooked *fufu* (32.5%), raw *fufu* (26.2%), and *gari* (22.7%).

3.3. Zinc Retention

Boiled storage roots: Mean zinc concentration was 8.95 mg/kg with a range from 6.5 to 10.8 mg/kg (Table 3). The genotype TMS 01/1371 had concentrations that were higher than the mean for all the parameters except iron.

Table 3. Zinc concentrations (mg/kg) and percentage true retention in selected cassava products prepared from yellow-fleshed cassava genotypes¹

Genotype	Raw Roots	Boiled roots	%TR ²	Raw <i>fufu</i>	%TR ²	Cooked <i>fufu</i>	%TR ²	<i>Gari</i>	%TR ²
TMS 01/1371	10.8	9.3	78.3	4.5	10.2	5.0	35.0	4.4	7.2
TMS 01/1235	9.5	8.6	86.4	7.9	15.2	7.5	38.0	4.6	10.7
TMS 94/0006	6.5	6.4	97.3	4.3	24.0	6.2	29.2	3.3	12.1
Mean	8.95	8.09	87.34	5.58	16.46	6.23	34.07	4.15	10.00
SE	1.29	0.86	5.51	1.16	4.05	0.72	2.58	0.40	1.46
P level	**	**	**	**	**	**	**	**	**

¹ Means of three replicates; ** = $P < 0.01$.

² = %TR- percentage true retention.

Cooked *fufu*: When the raw paste was further processed by cooking to thick dough there was a slight increase in the mean zinc content from 5.6 to 6.2 mg/kg (Table 3). This may be due to a concentration factor or contamination through the water used for cooking. After the adjustment for changes in weight and moisture during the processing of cooked *fufu*, the mean percentage retention was 34.1% for zinc and ranged from 29.2 to 38% (Table 3).

Gari: All the genotypes investigated exhibited a significant loss in zinc concentration. The mean percentage true retention was 10% with a range from 7.2 to 12.1%. The genotype TMS 01/1235 had intermediate retention for zinc in *gari* compared with TMS 01/1371.

In the present study, boiled cassava retained the highest amount of zinc (87.3%) followed by cooked *fufu* (34.1%), raw *fufu* (16.5%), and *gari* (10%).

Cassava processing involves a combination of activities which are performed in stages as described earlier. The number of steps required and the sequence vary with the product being made [19,20]. This sequence of activities may result in losses of zinc through leaching at each stage of processing which may explain the observed differences in nutrient retention by the various products.

4. Conclusion

The results from this study indicate that processing cassava storage roots resulted in a significant reduction in nutrient retention and the loss may be dependent on the processing method and genotype. In the present study, boiled storage roots had the highest retention of nutrients compared with the other food products.

Chavez *et al.* [24] evaluated 601 genotypes and reported that storage roots averaged 6.4 mg/kg for zinc content. After boiling, the mean zinc concentrations were lower than the mean concentration of the raw roots (Table 3). The genotype TMS 01/1371 had a higher concentration of zinc than TMS 94/0006. The genotypes TMS 94/0006 and TMS 01/1235 retained the highest amount of zinc compared with TMS 01/1371. The percentage mean zinc retention was 87.34% with a range from 78.3 to 97.3%. Although the genotype TMS 01/1371 had the lowest percentage true zinc retention and it was below the mean, it had the highest zinc concentration.

Uncooked *fufu*: When the raw roots were processed, raw *fufu* of genotypes TMS 94/0006 and TMS 01/1371 had a significantly lower concentration of zinc than TMS 01/1235 (Table 3).

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