

Impact of Processing on Nutrients and anti-Nutrients in Tubers and Leaves of Cassava (*Manihot Esculenta Crantz*)

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Abstract:

Purpose

The study investigated the effect of boiling and deep-frying on the levels of selected nutrients and anti-nutrients in tubers and leaves of new Migyera and MM96/2480 varieties planted in Busia County. Although cassava contains high levels of nutrients (elements and vitamins) that meet nutrient requirements when consumed as the main diet, it requires thorough cooking to reduce high levels of phytochemicals, anti-nutrients affecting nutrient bio-accessibility, and carcinogenic compounds; procedures which also affect the nutrient levels.

Methods

Tubers and leaves from the tips of the plant were harvested after 8 months. A portion of the tubers was boiled in water (100 °C, 20 minutes), while another deep-fried in vegetable cooking oil until it turned brown (5 minutes). Leaves were pounded until uniformly macerated and boiled in water (100 °C, 45 minutes). Levels of nutrients and antinutrients were determined by standard methods.

Results

The levels of nutrients and anti-nutrients were significantly higher in leaves than in tubers, and also differed significantly in different cassava varieties. Boiling of tubers significantly reduced the nutrients and anti-nutrients levels more compared to frying ($P < 0.001$), ranging from 13.7 to 97.7%. Deep frying of Migyera variety had higher nutrient reduction than in MM96/2480 variety. Essential elements and antinutrients molar ratios indicated that the elements were not bio-accessible.

Conclusion

Boiling and deep-frying of cassava tubers and leaves results in significant reduction of nutrient and anti-nutrient levels, implying that cassava cannot be used as the main diet; explaining the high level of child malnutrition in such communities.

Key words: cassava tubers; cassava leaves; boiling; deep frying; nutrients; anti-nutrients

Introduction

Cassava is a semi-arid food crop that would address malnutrition, and is commonly grown and consumed in tropical and sub-tropical countries such as Kenya. Malnutrition remains prevalent in children under the age of 5 years and has been reported in Busia (stunting 26 %, 13 % underweight, and 10.1 % wasting) and Kilifi (22 % moderately stunted and 7 % severely stunted) counties of Kenya [1-3]. Among the common cassava varieties grown in Kenya are the newly improved Migyera and MM96/2480 varieties, which were developed by the Kenya Agricultural and Livestock Research Organization (KALRO). Cassava tubers and leaves are good sources of nutrients such as protein, vitamins (C, B1, B2, B3) and minerals (Ca, Zn, Fe, K), but also contain high levels of anti-nutrients (oxalate and phytate) [4,5]. However, the variation in the levels of nutrients and antinutrients depends on the age of the plant, cultivar,

climatic conditions, and fertility of the soil [6]. In cassava roots, the levels reported are in the range of 0.3 – 3.5 mg/100 g (protein), 14.9 – 50 mg/100 g (vitamin C), 0.03 – 0.28 mg/100 g (vitamin B1), 0.03 – 0.06 mg/100 g (vitamin B2) and 1.3 – 2.8 mg/100 g, B3) [4]. These levels are sufficient to meet a good percentage of recommended dietary allowance (RDA) [7-10]. Wobeto et al. [11] and Waluchio [5] reported oxalate levels of between 1.35 g/100 g and 2.88 g/100 g and between 30.59 g/100 g and 154.7 g/100 g in cassava tubers and leaves respectively. Phytate level in tubers was reported in the range of 661.33 – 3040 mg/kg [12,13].

The bio accessibility of these nutrients is however affected by the presence of high levels of anti-nutrients such as oxalate, phytate and trypsin inhibitor in leaves. The presence of anti-nutrients in food affect bio accessibility of some nutrients; for example, by increasing elements

phytate molar ratio in food. The recommended molar ratios of phytate: zinc is < 15, that of phytate to iron is < 1 [14,15] and phytate: calcium > 0.24 [16].

Cassava tubers and leaves also contain other phytochemicals, mainly cyanogenic glycoside, which makes them poisonous and less popular [17]. On consumption, cyanogenic glycosides are broken down by the enzyme, linamarase into cyanohydrin and glucose. Cyanohydrin, being unstable, decomposes rapidly, producing hydrogen cyanide, which is poisonous [18]. Cassava tubers and leaves need therefore, to be processed by methods such as drying, fermentation, frying, pounding, and a long period of boiling to eliminate hydrogen cyanide [17,19,20].

The different processing methods have varied effects on nutrient and anti-nutrient levels. According to Ismaila et al. [21], the addition of palm oil during fermentation significantly increased the crude protein content of the bitter cassava product, yellow gari from 1.03±0.00 % to 1.22±0.01 % (P<0.05). Montagnac [3] observed that fermentation is the most efficient processing method to remove phytate (85.6 %) from cassava roots. During frying and boiling, there are high losses of phytate, which might be a result of thermal degradation and dissolution in water [22]. On the other hand, Omosuli [23] reported no significant difference in the ash, protein, crude fiber, sodium, magnesium, calcium, and phosphorus content in raw and boiled cassava tubers. According to Achidi [24], pounding or grinding cassava leaves before cooking both reduced ascorbic acid and thiamine content, with the loss being slightly higher with grinding (78.61% for ascorbic acid and 38.95% for thiamine) than pounding (76.87% for ascorbic acid and 35.14% for thiamine). The reduction was attributed to the leaching of the nutrients in boiling water. Ujong & Deede [25] reported a reduction of proteins by 4.40% on boiling cassava leaves.

It is expected that processing methods should retain high levels of nutrients but minimize levels of anti-nutrients and cyanogenic glycosides in different varieties of cassava to make consumable and provide required nutrients. The aim of the study was to investigate the impact of boiling and deep frying on the levels of nutrients [protein, minerals (K, Ca, Fe, Zn), and vitamins (C, B1, B2, B3)], and anti-nutrients (oxalate and phytate) in two new cassava varieties (Migyera and MM96/2480) grown in Busia County, Kenya.

2 Materials and methods

2.1 Equipment and chemicals

High Performance Liquid Chromatography, (HPLC) (Shimadzu 20A series), with a Hypsil C18 column was used for the measurement of vitamins and anti-nutrients. The Kjeldahl apparatus was used for measurement of protein, and an Atomic Absorption Spectrophotometer, (AAS) (Shimadzu AA-7000) was used for determination of minerals. Standards for minerals, vitamins, oxalate and phytate, and all other chemicals were obtained from Sigma-Aldrich, Germany.

2.2 Sampling and sample preparation

Migyera and MM96/2480 cassava varieties were planted in Busia County in June 2020 (rainy season). The county is located within the coordinates, 0° 28' 0''N/ 34° 6' 0''E. It covers an area of 1695 km² and has a population of 893,681 [26]. It is covered mostly with sandy loam soils although the northern and central parts are covered with dark clay soils. The county receives an annual rainfall of between 750 mm and 2000 mm [27]. The mean temperature in the county is between 21 and 27 °C [28]. Busia County is the largest producer of cassava in Kenya, with a production of 16.4 tons per hectare [29].

A 19 by 19 m plot was tilled using a hoe, and planting of cuttings obtained from the Kenya Agricultural and Livestock Research Organization (KALRO) done the following day at intervals of 1 by 1 m. Neither fertilizer application nor irrigation were done. Weeding was done

monthly, and the tubers and the first three leaves from the tips of the plant were harvested after 8 months. The preparation and processing of both cassava tubers and leaves were done according to home-based protocols. The tubers were washed to remove soil particles, peeled, split longitudinally into two halves, and cut into pieces of about 4 cm long. The pieces were washed using tap water and then divided into three portions. The first portion was packed raw in plastic containers. The second portion, consisting of 500 g was placed in 500 mL of water in a pan, then boiled at 100 °C until it became tender (approximately 20 minutes), cooled, and then packed in plastic containers. The third portion (500 g) was deep fried in 1 L of vegetable cooking oil until it turned brown (5 minutes), then cooled and packed in plastic containers.

The leaves from each variety were pounded in a mortar and pestle until uniformly macerated, then divided into two portions. The first portion was analyzed raw, while the second portion (500 g) was boiled in 1 L of water at 100 °C for 45 minutes and packed in plastic containers. Both tubers and leaf samples in plastic containers were placed in a cooler box kept at around 0 °C. All the samples were then transported to Department of Food Science and Technology laboratories, Jomo Kenyatta University of Agriculture and Technology (JKUAT) within 8 hours where they were frozen at around -10 °C while awaiting the analysis.

2.3 Determination of crude protein

Protein was determined by the Kjeldahl method, according to Maehre et al. [30]. All the procedures were carried out in triplicate. One gram of the sample was digested using 15 mL concentrated H₂SO₄ and 0.5 g of CuSO₄ catalyst. The mixture was heated until the digest color turned blue. It was cooled, transferred into a 100 mL volumetric flask, and topped up with distilled water. A blank digestion was also done. Ten (10) mL of digest was transferred into a distilling flask, which was connected to another flask that contained boric acid, and distillation was done to a volume of about 60 mL distillate. The distillate was titrated using 0.02 N-HCl to an orange color of the mixed indicator. Total nitrogen was calculated using equation 1.

$$N (\%) = ((\text{mL } 0.02 \text{ N sample} - \text{mL } 0.02 \text{ N blank}) \times 0.0014 \times N \text{ HCl} \times 100) / (\text{Weight of sample}) \quad \text{Equation 1.}$$

Protein content was determined from total nitrogen by multiplying the concentration of nitrogen in food by a conversion factor of 6.25.

2.4 Determination of vitamins

Vitamins C, B1, B2, and B3 were determined using a reversed phase (RP) HPLC procedure adopted from procedures described by Martin et al. [31], Tanmay et al. [32] and Sudipta et al. [33]. All procedures were done in triplicate. Exactly 20 cm³ of deionized water were added to a 5 g food sample. The mixture was homogenized for 1 min then centrifuged for 10 min at 14000 g. Then 10 mL of homogenized and centrifuged food sample was loaded on an activated stationary phase and eluted with 10 mL methanol and 10 mL water whose pH had been adjusted to 4.2 at a flow rate of 1 mL min⁻¹. The eluent was collected in a bottle and evaporated to dryness. The residue was dissolved in the mobile phase, filtered with a 0.45 µm Millipore filter and 20 µL of the filtrate was infused into the HPLC system. The separation was achieved by a C18 column (internal diameter 4.6 mm; pore size 3.5 µm) using a mobile phase of 0.1 M KH₂PO₄ (pH 7): methanol (9:1) at a flow-rate of 0.7 mL min⁻¹ and 25 °C. The column elute was monitored with a photodiode-array detector at 265 nm, 234 nm, 266 nm and 261 nm for vitamin C, thiamine, riboflavin, and niacin, respectively. Identification and quantification of compounds were achieved by comparing their peak areas with those of standards of known concentrations using the calibration curves.

2.5 Determination of minerals

In the determination of the levels of minerals, standard calibration curves were developed from the calcium, zinc, iron, and potassium standards

series (0-2 µg/mL). All the procedures were done in triplicate. A clean dry crucible was weighed, and about 5 grams of sample were weighed into it. The crucibles were placed on a hot plate under a fume hood, and the temperature was slowly increased until smoking ceased and the samples were thoroughly charred. They were then placed in a muffle furnace and heated for 1 hour as temperature increased gradually to 250 °C. The temperature was increased to 600 °C and incinerated for about 5 hours. The temperature was then decreased to 300 °C, the crucibles were removed and they were cooled to room temperature. The ash was transferred quantitatively to 100 mL beaker using 20 mL of 0.5 N HNO₃, then stirred and allowed to settle. This was then transferred to a 100 mL volumetric flask and filled to the mark using 0.5 N HNO₃. Insoluble matter was filtered, and absorbance of the solutions was read by an Atomic Absorption Spectrophotometer (AAS) at 422.7 nm, 213.9 nm and 248.3 nm for calcium, zinc, and iron respectively. Emission was read by an Atomic Emission Spectrophotometer (AES) for potassium at 766 nm and concentrations of all the minerals were obtained from their respective calibration curves developed from their standards.

2.6 Determination of Oxalate

The sample procedure followed for the determination of oxalate was done according to Karanja et al. [34]. Standard calibration curves were developed from the oxalate standard series (0-200 µg/mL). All the procedures were done in triplicate. Exactly 0.5 g fresh weight of the sample from the freezer was homogenized with 4 mL of 0.5 N HCl, heated at 80 °C for 10 minutes with intermittent shaking, and then distilled water was added to a volume of 25 mL. An aliquot of 3 mL of the solution was withdrawn and centrifuged at 24149 g for 10 minutes to get 1 mL of supernatant that was filtered through a 0.45µm microfilter for injection into the HPLC column. HPLC detection was done using a Shimadzu UV-VIS detector, and a Hypsil C18 column (5µ M, 4.6 mm *250 mm) equipped with waters 550 was used as the static phase. Mobile phase was 0.25 % dehydrogenate phosphate and 0.0025 M, Tetrabutyl Ammonium hydrogen sulphate buffered at pH 2.0 with orthophosphoric acid. Flow rate of 0.6 mL min⁻¹, pressure of 62 kgf and a detection wavelength of 314 nm.

2.7 Determination of phytate

Phytates were analyzed by the HPLC method according to Camire & Clydesdale [35]. Standard calibration curves were developed from the phytate standards (0-200 µg/mL range). All the procedures were done in triplicate. Approximately 0.5 g of sample was extracted with 10 mL of 3 % H₂SO₄. The contents were filtered, and the filtrate was transferred to a boiling water bath for 5 minutes, followed by the addition of 3 mL of FeCl₃ solution (6 mg ferric iron per mL in 3 % H₂SO₄). The contents were heated for 45 minutes to complete precipitation of the ferric phytate complex. They were then centrifuged at 1048 g for 10 minutes, and the supernatant was discarded. The precipitate was washed with 30 mL distilled water, centrifuged again at the same speed and time, and the

supernatant was discarded. A 3 mL of 1.5 N NaOH was added to the residues, and the volume was brought to 30 mL with distilled water. The contents were heated for 30 minutes in a boiling water bath to precipitate the ferric hydroxide. Cooled samples were centrifuged, and the supernatant was transferred into a 50 mL volumetric flask. The precipitate was rinsed with 10 mL distilled water, centrifuged (1048 g for 10 minutes) and the supernatant was added to the contents of the volumetric flask. This was microfiltered, and HPLC analysis was carried out. The mobile phase was 0.005 N sodium acetate in distilled water, and detection was done at 500 nm.

2.8 Determination of molar ratio of phytate/mineral

The molar ratio of phytate to minerals was determined as described by Norhaizan & Faizadatul [36]. The mole of phytate and minerals was determined by dividing the weight of phytate and minerals by their atomic weights (phytate: 660 g/mol; Fe: 56 g/mol; Zn: 65 g/mol; Ca: 40 g/mol). The molar ratio between phytate and minerals was obtained by dividing the mole of phytate with the mole of minerals.

Molar ratio of phytate/ mineral = (moles of phytate)/(moles of mineral)
Equation 2.

2.9 Method validation procedures

The linearity of standard calibration curves was checked by calculating the correlation coefficient R². The accuracy of the methods was verified by determining the percentage recovery after spiking the samples. Precision was determined by calculating the coefficient of variation (CV) of repeated measurements of the test samples.

2.10 Data analysis

The levels of nutrients and anti-nutrients in cassava tubers and leaves under different processing as well as in different varieties were compared using one way ANOVA, using the SPSS program (version 28) to determine a significant difference at P < 0.001. The calculation of the mean separations was done by standard error [37].

3 Results and discussion

3.1. Method validation

The method validation results are shown in **Table 1**. The percentage recoveries ranged from 98.000 % to 100.002 % which confirms that the methods of analysis used were accurate and suitable for the analysis of each parameter [38]. The coefficient of variations (CV) ranged from 0.6897 to 2.9851, which was within the sufficiently precise range [39]. The degree of linear relation between the signal and concentration shown by the correlation coefficient R² ranged from 0.9921 to 1.0000, which indicates a 99.21-100 % instrumental response, and that established that the calibration curves used were linear over the respective range of the concentration of the standards [40].

Analyte	Equation	R ²	% Recovery	% CV
Vitamin C	y= 42299x	0.9999	99.996	2.686
Vitamin B ₁	y= 33361x	0.9936	99.940	2.838
Vitamin B ₂	y= 128202x	0.9999	100.002	0.690
Vitamin B ₃	y= 211142x	1.0000	99.940	1.859
Calcium	y= 0.06x+0.0104	0.9921	98.000	2.728
Zinc	y= 1.065x-0.0109	0.9966	99.800	2.985
Iron	y=0.1136x+0.0024	0.9975	99.200	2.687
Potassium	y= 8572x+0.0399	0.9939	99.400	1.361
Phytate	y= 12745x	0.9991	99.998	1.019
Oxalate	y= 43493x	0.9992	99.994	2.871

Table 1: Methods validation parameters

Where x is concentration, and y is absorbance

3.2 Levels of proteins and vitamins

The mean levels of proteins and vitamins in raw and processed cassava tubers and leaves are shown in Table 2. The levels in raw tubers and leaves, which ranged from 0.056±0.001 to 0.219±0.001 mg/100 g for protein, 2.137±0.037 to 23.863±0.491 mg/100 g for vitamin C, 1.163±0.033 to 13.353±0.146 mg/100 g for vitamin B1, 0.145±0.001 to 0.485±0.008 mg/100 g for vitamin B2, and 0.767±0.011 to 8.875±0.079 mg/100 g for vitamin B3 agree with those reported by USDA (2008). Leaves had levels within the ranges reported in kale and spinach [41],

which are common green leafy vegetables consumed in Kenya. Cassava leaves had significantly higher ($P < 0.001$) levels of protein and vitamins than tubers, which agrees with the findings of the USDA (2008). This may be attributed to the fact that nutrients are synthesized in leaves, and tubers only act as storage sites. The roots are the main part of cassava that is consumed because the leaves contain higher levels of anti-nutrients and cyanogenic glucosides that are not easily removed by boiling, which is the main method of processing green vegetables [42]. This explains why a cassava tuber diet does not provide necessary nutrients.

P<0.001					
Vitamin B ₁ (mg/100 g)	<i>Migyera</i> tuber	1.163±0.033 ^{cA}	0.319±0.009 ^{aA} (72.6%)	0.688±0.015 ^{hA} (40.8%)	P<0.001
	<i>Migyera</i> leaves	13.353±0.146 ^{bD}	0.924±0.016 ^{aD} (93.1%)		
	MM96/2480 tubers	1.190±0.020 ^{cB}	0.335±0.006 ^{aB} (71.8%)	0.993±0.028 ^{hB} (16.6%)	
	MM96/2480 leaves	4.212±0.030 ^{bC}	0.392±0.010 ^{aC} (90.7%)		
P<0.001					
Vitamin B ₂ (mg/100 g)	<i>Migyera</i> tuber	0.145±0.001 ^{bA}	0.031±0.001 ^{aC} (78.6%)	0.067±0.0018 ^{aA} (53.8%)	P<0.001
	<i>Migyera</i> leaves	0.485±0.008 ^{bD}	0.020±0.005 ^{aB} (95.9%)		
	MM96/2480 tubers	0.403±0.004 ^{cC}	0.049±0.001 ^{aD} (87.8%)	0.085±0.0009 ^{hB} (78.9%)	
	MM96/2480 leaves	0.394±0.005 ^{bB}	0.009±0.001 ^{aA} (97.7%)		
P<0.001					
Vitamin B ₃ (mg/100 g)	<i>Migyera</i> tuber	1.399±0.026 ^{cB}	0.358±0.004 ^{aA} (74.4%)	0.575±0.012 ^{hA} (58.9%)	P<0.001
	<i>Migyera</i> leaves	8.875±0.079 ^{bD}	2.775±0.032 ^{aD} (68.7%)		
	MM96/2480 tubers	0.767±0.011 ^{cA}	0.585±0.009 ^{aB} (23.7%)	0.610±0.011 ^{hB} (20.5%)	
	MM96/2480 leaves	6.021±0.118 ^{bC}	1.377±0.014 ^{aC} (77.1%)		
P<0.001					

Table 2: Mean levels (DM) of protein and vitamins in raw and processed tubers and leaves of cassava varieties

Mean values followed by a different small letter within the same row differ significantly from one another. Mean values followed by a different capital letter within the same column differ significantly from one another (One - Way ANOVA, $P < 0.001$, SNK - test, $\alpha = 0.05$).

Although the cassava varieties were grown under similar conditions, the levels of protein and vitamins varied significantly ($P < 0.001$) between the *Migyera* and MM96/2480 varieties. For instance, protein in tubers of the *Migyera* variety was 0.114±0.003 mg/100 g while it was 0.056±0.001 mg/100 g in tubers of the MM96/2480 variety. Tubers of the *Migyera* variety were also higher in vitamins; C, and B3, minerals (Zn and K) and anti-nutrients (oxalate and phytate) than the MM96/2480 variety. The leaves of *Migyera* had higher levels of vitamins (C, B1, B2 and B3), minerals (calcium, zinc, and potassium), and anti-nutrients (phytate). This may be attributed to the genetic differences of the different cassava varieties [43].

The levels of proteins and vitamins were reduced significantly ($P < 0.001$) with boiling and deep frying. Boiling reduced nutrients in the ranges of 80.4 % - 92.1 % (protein), 46.0 % - 92.4 % (vitamin C), 71.8 % - 72.6 % (vitamin B1), 78.6 % - 87.8 % (vitamin B2), 23.7 % - 74.4 % (vitamin B3), which agrees with the results reported by Alamu et al. [44] and Zipporah et al. [45] for vitamin C and B1. Frying reduced nutrient levels

in tubers by 51.8 % - 88.6 % (protein), 39.6 % - 43.1 % (vitamin C), 16.6 % - 40.8 % (vitamin B1), 53.8 % - 78.9 % (vitamin B2), 20.5 % - 58.9 % (vitamin B3). Boiling significantly reduced the levels of proteins more than deep frying, mainly due to leaching [23,46]. Frying is a better processing method for tubers than boiling, as it retains relatively higher levels of water-soluble nutrients. Both processing methods significantly reduced nutrient levels ($P < 0.001$) hence, a small fraction of the total nutrients in raw cassava will be available in processed cassava products, resulting in lower bioavailability. This may explain why there are cases of malnutrition in communities that rely on a cassava-based diet.

3.3 Levels of minerals and anti-nutrients

The mean levels of minerals and anti-nutrients in raw and processed tubers and leaves are shown in Table 3. The ranges were in 32.075±0.875 - 50.134±0.204 mg/100 g for calcium, 0.544±0.003 - 1.160±0.009 mg/100 g for zinc, 0.707±0.019 - 3.141±0.028 mg/100 g for iron, 298.397±3.107 - 485.778±6.609 mg/100 g for potassium, 284.274±6.296 - 613.456±17.611 mg/100 g for oxalate, and 452.774±13.922 - 561.281±5.720 mg/100 g phytate. The levels are in the same range as reported by Tagesse & Tesfaye [47] and the USDA [48]. Similar levels were reported in kale for calcium [49], iron [41] and zinc [50], while the USDA [48] reported results in the same range in spinach for calcium (99

mg/100 g), zinc (0.53 mg/100 g), iron (2.71 mg/100 g) and potassium (558 mg/100 g). In other studies, oxalate levels ranged between 1.35 and 2.88 g/100 g and between 30.59 and 154.7 g/100 g in cassava tubers and leaves, respectively [5,11], while phytate levels in tubers were reported in the range of 661.33 – 3040 mg/kg [12,13,51]. The levels of the nutrients are high and meet a percentage of the RDA. The leaves of MM96/2480 variety had significantly (P<0.001) higher levels of Ca, Fe, and K than the Migyera variety. The tubers of MM96/2480 had significantly (P<0.001) higher levels of Ca and Fe than the Migyera. This may be attributed to the genetic differences of the different cassava varieties [43] as both varieties were planted in the same soil under the same environmental conditions. The leaves of Migyera variety had significantly higher levels of Ca, Zn and Fe than tubers.

The elements and anti-nutrients were significantly reduced (P<0.001) by the processing methods of boiling and deep frying for tubers and leaves.

Boiling reduced calcium by 77.7 % - 79.3 %, zinc 17.5 % - 22.8 %, iron 13.7 % - 43.9 %, potassium 22.0 % - 32.4 %, oxalate 31.8 % - 74.3 %, and phytate 58.6 % - 73.9 %. On the other hand, deep frying reduced calcium by 32.5 % - 36.8 %, zinc 2.6 % - 16.4 %, iron 6.6 % - 13.3 %, potassium 0.02 % - 8.6 %, oxalate 12.6 % - 58.9 %) and phytate 47.3 % - 51.7 %. The percentage reductions were lower than for protein and vitamins under the same processing conditions. The leaves experienced a significantly higher reduction in the elements and anti-nutrients as a result of leaching and evaporation, especially when the leaves were pounded before boiling [23,46]. The losses of phytate due to frying and boiling might be a result of thermal degradation and dissolution in water [22]. Phytate is soluble in water and relatively heat stable during normal household boiling, but experiences higher losses in industrial processing such as canning or extrusion cooking where high temperatures are used [52].

Nutrients and Anti-nutrients (mg/ 100 g)	Cassava variety/part	Raw Mean ± SD	Process		P-Value
			Boiled Mean ± SD (% reduction)	Fried Mean ± SD (% reduction)	
Calcium	Migyera tuber	32.075±0.875 ^{cA}	6.644±0.146 ^{aA} (79.3%)	21.646±0.534 ^{bA} (32.5%)	P<0.001
	Migyera leaves	50.134±0.204 ^{bD}	20.288±0.221 ^{aC} (59.5%)		
	MM96/2480 tubers	47.461±0.634 ^{cC}	10.571±0.124 ^{aB} (77.7%)	29.986±0.214 ^{bB} (36.8%)	
	MM96/2480 leaves	39.149±0.381 ^{bB}	33.021±0.240 ^{aD} (15.7%)		
P<0.001					
Zinc	Migyera tuber	0.737±0.022 ^{bB}	0.608±0.0151 ^{aD} (17.5%)	0.618±0.016 ^{aB} (16.4%)	P<0.001
	Migyera leaves	1.160±0.009 ^{bD}	0.350±0.005 ^{aA} (69.8%)		
	MM96/2480 tubers	0.544±0.003 ^{bA}	0.420±0.011 ^{aB} (22.8%)	0.530±0.006 ^{bA} (2.6%)	
	MM96/2480 leaves	1.014±0.021 ^{bC}	0.466±0.012 ^{aC} (54.0%)		
P<0.001					
Iron	Migyera tuber	0.707±0.019 ^{bA}	0.610±0.018 ^{aB} (13.7%)	0.613±0.015 ^{aA} (13.3%)	P<0.001
	Migyera leaves	2.381±0.034 ^{bC}	1.476±0.017 ^{aC} (38.0%)		
	MM96/2480 tubers	0.715±0.020 ^{bB}	0.401±0.009 ^{aA} (43.9%)	0.668±0.013 ^{bB} (6.6%)	
	MM96/2480 leaves	3.141±0.028 ^{bD}	1.779±0.011 ^{aD} (43.4%)		
P<0.001					
Potassium	Migyera tuber	485.778±6.609 ^{cD}	328.570±3.754 ^{aD} (32.4%)	375.798±3.777 ^{bB} (22.64%)	P<0.001
	Migyera leaves	375.798±3.777 ^{bC}	185.454±2.092 ^{aB} (50.7%)		
	MM96/2480 tubers	326.43±5.037 ^{cB}	254.473±3.280 ^{aC} (22.0%)	298.212±1.281 ^{bA} (8.6%)	
	MM96/2480 leaves	298.397±3.107 ^{bA}	163.301±2.542 ^{aA} (45.3%)		
P<0.001					
Oxalate	Migyera tuber	613.456±17.611 ^{cD}	157.406±2.307 ^{aB} (74.3%)	252.267±5.497 ^{bA} (58.9%)	P<0.001
	Migyera leaves	284.274±6.296 ^{bA}	115.562±3.478 ^{aA} (59.3%)		
	MM96/2480 tubers	413.863±8.087 ^{cB}	282.252±2.855 ^{aC} (31.8%)	361.529±7.187 ^{bB} (12.6%)	
	MM96/2480 leaves	526.982±0.177 ^{bC}	294.280±4.121 ^{aD} (44.2%)		
P<0.001					
Phytate	Migyera tuber	561.281±5.720 ^{cD}	146.343±0.942 ^{aC} (73.9%)	271.163±2.486 ^{bB} (51.7%)	P<0.001
	Migyera leaves	555.994±5.718 ^{bC}	61.520±0.787 ^{aB} (88.9%)		
	MM96/2480 tubers	463.774±5.750 ^{cB}	192.229±3.641 ^{aD} (58.6%)	244.508±2.787 ^{bA} (47.3%)	
	MM96/2480 leaves	452.774±13.922 ^{bA}	32.089±0.338 ^{aA} (92.9%)		
P<0.001					

Table 3: Mean levels (DM) of minerals and anti-nutrients in raw and processed tubers and leaves of cassava varieties

Mean values followed by a different small letter within the same row differ significantly from one another. Mean values followed by a different capital letter within the same column differ significantly from one another (One - Way ANOVA, P < 0.001, SNK - test, α = 0.05). (% reduction levels on processing)

Even though boiling and frying lead to significant reductions in oxalate and phytate, the levels that remain are higher than the recommended molar ratios (Table 4). The ratios for phytate to elements were: zinc < 15, iron < 1 and Calcium < 0.24 [14-16]. The results show that although raw cassava tubers and leaves contain high levels of proteins, vitamins, and

minerals, a higher percentage of nutrients is lost during boiling and deep frying. Boiling and deep frying are the main methods used to prepare cassava for consumption. The processed cassava tubers and leaves therefore contribute very little towards meeting the RDAs, which are, 0.83 g/kg (protein), 40 mg – 120 mg/day (vitamin C), 0.2 mg – 1.4 mg/day

(vitamin B1), 0.3 mg – 1.6 mg/day (vitamin B2), 2 mg – 18 mg/day (vitamin B3), 200 mg – 1300 mg/day (Ca), 8 mg – 11 mg/day (Zn), 1.8 mg/day (Fe) and 4700 – 5100 mg/day (K) [7-10]. Both boiled and fried cassava tubers and leaves cannot therefore be relied on to alleviate malnutrition.

Mineral	Cassava variety/part	Phytate:Mineral Molar ratio			Recommended molar ratios (Hurrell and Egli, 2010; Magallanes-Lopez <i>et al.</i> , 2017; Morris & Ellis, 1985)
		Raw	Boiled	Deep fried	
Calcium	<i>Migyera</i> tubers	1.061	1.335	0.759	<0.24
	<i>Migyera</i> leaves	0.672	0.184		
	MM96/2480 tubers	0.592	1.102	0.494	
	MM96/2480 leaves	0.701	0.059		
Zinc	<i>Migyera</i> tubers	75.003	23.705	43.213	<15
	<i>Migyera</i> leaves	47.204	17.311		
	MM96/2480 tubers	83.961	45.075	45.435	
	MM96/2480 leaves	43.976	6.782		
Iron	<i>Migyera</i> tubers	67.360	20.356	37.533	<1
	<i>Migyera</i> leaves	19.813	3.537		
	MM96/2480 tubers	55.036	40.674	31.057	
	MM96/2480 leaves	12.231	1.530		

Table 4: Phytate to minerals molar ratios in raw and processed cassava products

4 Conclusions

The levels of nutrients and anti-nutrients are not only significantly higher in leaves than in tubers but also significantly different among cassava varieties. The leaves of the MM96/2480 variety are more nutritious than those of the *Migyera* variety, while the tubers of the *Migyera* variety are more nutritious than those of MM96/2480 variety. The levels of nutrients in both tubers and leaves are significantly affected by boiling and deep frying, reducing the levels by up to 97 %. Hence, boiled and deep-fried cassava products cannot be relied on to reduce malnutrition in communities. However, deep-frying is a better processing method for tubers than boiling, as it retains relatively higher levels of water-soluble nutrients. Both boiling and frying significantly reduce the anti-nutrients (oxalate and phytate) in both tubers and leaves, although levels that remain still affect the bio-accessibility of the nutrients due to high molar ratios. This may explain why there is a high level of child malnutrition in communities that rely on cassava food.

Statements and Declarations

Ethical approval

The study followed national guidelines and legislation, as appropriate permission for the study was received from the National Council of Science Technology and Innovation, Kenya (NACOSTI).

Consent for publication

The manuscript does not have details, images, or videos relating to an individual person and thus requires no consent to publish.

Availability of data and materials

The data obtained and analyzed during the current study is available from the corresponding author on reasonable request.

Competing interest

The authors declare that they have no competing interests.

Funding

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Authors contribution

All authors contributed to the study conception and design. Data collection and analysis, and the drafting of the manuscript was by Ogombe, Charles Ojiambo. The other authors read, commented on previous versions and approved the final manuscript. The authors have agreed both to be personally accountable for the author's own contributions and to ensure that questions related to the accuracy or integrity of any part of the work.

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