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RESEARCH ARTICLE

COMPARATIVE ASSESSMENT OF THE ANTINUTRITIONAL COMPOSITION OF TWO CASSAVA (*MANIHOT ESCULENTA* CRANTZ) VARIETIES GROWN IN THREE MAJOR AREAS OF KOGI STATE

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ABSTRACT

Cassava (*Manihot esculenta Crantz*) is a perennial crop with an edible starchy tuberous root grown in tropical and subtropical regions. However, limited studies have assessed the antinutritional factors affecting its nutrient availability. This study evaluated the antinutritional composition of two cassava varieties (white TMS 30001 and yellow TMS 30572) grown in Ankpa, Kabba, and Okene (Kogi State) using standard methods. Tubers from Agricultural Development Project (ADP) farms were analyzed at Kogi State University Biochemistry Laboratory for cyanide, tannin, oxalate, phytate, and trypsin inhibitor content. Data were analyzed using ANOVA (SPSS version 20), with Duncan multiple range test (DMRT) for mean separation and T-test for comparing both varieties. Results showed significant differences (P≤0.05) among the antinutrient compositions. In white cassava, Okene tubers had the highest antinutrient values except for phytate (1.71±0.04mg/100g) and tannin (0.36±0.1mg/100g), which were highest in Kabba and Ankpa, respectively. In yellow cassava, Okene had the highest levels of all antinutrients except phytate (3.31±0.10mg/100g) and oxalate (0.23±0.00mg/100g), which were highest in Ankpa. The T-test indicated that white cassava had higher cyanide, oxalate, and trypsin inhibitor levels, while yellow cassava had higher phytate content. Both varieties had similar tannin levels. The lower antinutrient content of yellow cassava suggests it is more suitable for human consumption and livestock feed than white cassava across the three locations.

KEYWORDS

Cassava, Antinutrient, Cyanide, Tannin, Trypsin Inhibitor

1. INTRODUCTION

Cassava (*Manihot esculenta* Crantz), referred to as "Abacha" by the Igalas, "Rogo" by the Okuns, and "Echuka" by the Ebiras in Nigeria, is a perennial woody shrub belonging to the Euphorbiaceae family. This plant originated in tropical America and was brought to Africa by Portuguese explorers during the sixteenth century (Adenle, 2020). It is primarily grown in tropical and subtropical regions around the globe for its edible tuberous roots (Akinpelu et al., 2018). As a vital subsistence crop, cassava ranks as the third largest source of carbohydrates for human consumption worldwide, following rice and corn, with Africa being its predominant production continent (Soryotha et al., 2010). Currently, Nigeria holds the title of the world's largest cassava producer, having produced approximately 57 million metric tonnes in 2016 (FAO, 2018). Cassava is a starchy staple that produces approximately 40% more carbohydrates than rice and 25% more than maize (Tonukari, 2014). The roots of cassava are mainly utilized for food products such as garri, fufu, and lafun (Obuh, 2011). This is largely due to the high carbohydrate content of cassava roots, which ranges from 60% to 70% in Nigerian varieties. Additionally, cassava is a good source of essential minerals including calcium, phosphorus, manganese, iron, zinc, and potassium (Udoetok and Uffia, 2012).

Cassava is categorized into sweet and bitter varieties based on their cyanide levels. Sweet cassava varieties contain low levels of cyanogens and can be consumed with minimal processing. In contrast, bitter cassava varieties have high cyanogen content and require extensive processing to eliminate these compounds before they can be safely eaten (Obueh and Kolawale, 2016). The cassava plant grows faster compared to other crops and has remarkable ability to withstand harsh climatic conditions. These two qualities have made the diverse edible varieties of cassava to be an outstanding crop that saves at least 500 million of human lives from hunger worldwide (Tivana et al., 2018). Cassava plays a food security role in areas prone to drought, famine and in periods of strives and civil disturbance (Akoja and Mohammed, 2014). The crops ability to provide a staple food base is a function of its flexibility in terms of planting and harvesting strategies, and because of its relative tolerance of poor soils and pest/disease problems. This explains why it is valued in Africa, for the food security it provides. It is also grown for industrial purposes such as the production of starch and for fermentation into ethanol (Adelekan, 2020).

Furthermore, cassava contains some antinutritional and toxic substances such as cyanide, oxalate, phytate, tannin, saponins and trypsin inhibitor (Montagnac et al., 2009). These substances interfere with its digestibility and uptake of nutrients, and might present toxic effect depending on the amount in which it is consumed. Anti-nutrients or antinutritional factors may be defined as those substances generated in natural feedstuffs by the normal metabolism of species and by different mechanisms (for example inactivation of some nutrients, diminution of the digestive process or metabolic utilization of feed) which exerts effect contrary to optimum nutrition (Gemede and Ratta, 2014).

The presence of antinutrients in cassava may reduce the acceptability of it's food products and may have negative impact on the carbohydrate and

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mineral bioavailability (Mihrete, 2019). Furthermore, of all the antinutrient present in cassava, cyanide is the most toxic factor restricting the consumption of cassava tubers. A group researchers reported that consumption of high levels of cyanide causes acute toxicity, goiter, neuropathy and in severe cases, lead to death (Gonclaves et al., 2016). High amount of phytates causes muscular weakness and paralysis, tannins: digestive tract malfunction and trypsin inhibitors inhibit digestive enzymes and cause growth depression in humans as reported by (Gemede and Ratta, 2014; Sarkiyayi and Agar 2010). However, some of these compounds may exert beneficial health effects at low concentrations (Pihlanto, 2017). For example, saponin, when used at low levels have also been shown to reduce the blood glucose and insulin responses to starchy foods (Singh et al., 2017). In addition, phytates and tannins have been related to reduce cancer risk (Silva and Bracarense, 2016). Studies have shown that processing techniques such as fermentation, heat treatment, soaking and several others aids in the reduction or removal of these antinutrients (Popova and Mihaylova, 2019). Hemce this study tends to access and compare the antinutritional composition of two cassava varieties cultivated in three major regions (Ankpa, Okene and Kabba) of Kogi State.

2. Materials and Methods

2.1 Study Area

This experimental work was carried out at the department of Biochemistry laboratory, Faculty of Natural Sciences, Kogi State University Ayingba which is located between Longitude 7º91^{1}E and 7º121^{1}E and Latitude 7º281^{1}N and 7º321^{1}N in the Eastern part of Kogi State in Dekina Local Government Area of Nigeria.

2.2 Source of Materials

Freshly harvested white and yellow tubers (TMS 30001 and 30572) were obtained from Agricultural development Program (ADP) farms in Ankpa (Latitude 7º36^{1}N and Longitude 7º62^{1}E), Kabba (Latitude 7º82^{1}N and Longitude 6º07^{1}E) and Okene (Latitude 7º55^{1}N and Longitude 6º23 in Kogi State, Nigeria. The tubers were obtained from 10-14month old plant. The tubers are shown in Plate I, II and III below:

PLATE I: White and yellow cassava from Ankpa

PLATE II: White and yellow cassava from Kabba

Plate IV: Samples labelled in beakers respectively

Plate III: White and Yellow Cassava from Okene

2.3 Preparation of Samples

The white and yellow cassava roots were properly washed to get rid of sand and dust, the peels were removed and the pulps cut into smaller sizes. The samples were crushed using mortal and pistle, and transfered into beakers. The beakers were labellled A_1(White cassva from Ankpa), A_2(Yellow cassava from Ankpa), B_1(White cassava from Kabba), B_2(Yellow cassava from Kabba), C_1 (White cassava from Okene) and C_2(yellow cassava from Okene) as shown in plate IV above. The samples were then analysed for anti-nutrients present.

2.4 Determination of Anti-nutritional Compositions

2.4.1 Cyanide Determination

Cyanide content were determined by alkaline picrate method according to wary and filled method as described (Onwuka, 2015). 5g of powdered sample was dissolved in 50ml of distilled water in a conical flask and the extraction was allowed to stand overnight. 1ml of sample filtered was mixed with 4ml alkaline picrate in a cork testube and incubated in a ||water bath for 5minutes. After colour development (Reddish brown colour), the absorbance was read at 490nm, the absorbance of the blank containing 1ml distilled water and 4ml alkaline solution was also recorded. The cyanide content was extrapolated from cyanide standered curve prepared from different concentration of KCN solution containing 5-5ug cyanide in a 500l conical flask followed by the addition of 25ml of 1NHCl.

2.4.2 Phytate Determination

Analysis of phytate determination in samples followed the method (AOAC, 2012). Phytate was extracted with 0.5mol/L HNO_3 solution and digested with 0.5ml of perchloride acid ($HClO_4$.). The digested sample was made up to 25ml volume with distilled water in a standard volumetric flask. From this extract, 2.5ml of sample was taken and added to 2.5ml of nitric acid and the phytic phosphorous present in the extract reacted with 25ml of Vanadium molybdate (solution) reagent to produce a yellow-orange

complex. The absorbance (OD) was measured at 450nm wavelength using a spectrophotometer (P7 UV/Vis spectrophotometer). The phytate content was calculated from the 2mg of phytic acid standard concentration with a reagent blank treated as sample above.

$$\text{Phytate (Mg/100g)} = \frac{\text{Sample absorbance} \times \text{Standard Concentration}}{\text{Standard absorbance} \times \text{weight of sample}} \times 100$$

2.4.3 Oxalate Determination

Total oxalates were determined according to the procedure of fasset (2016). The extraction was done by weighing 1g of each sample and soaked with 100ml of distilled water. These were allowed to stand for 3hours and each was filtered through a double layer of filter paper. 10, 20, 30, 40 and 50ppm standard solution of oxalic acid were prepared and read on the spectrophotometer at 420nm for the absorbance. The absorbance of filtrate from each samples were also read on the spectronic 20.

2.4.4 Tannin content Determination

Analysis of tannin content was determined by the AOAC (2012) method. Sample (5g) was dispensed in 50ml of distilled water and shaken. The mixture was allowed to stand for 30minutes at 28°C before it was filtered through whatman no.4 grade of filter paper. The extract (2ml) was dispensed into a 50ml volumetric flask. Similarly, 2ml standard tannin solution (0.1mg/ml tannic acid) and 2ml distilled water were put in a separate volumetric flask to serve as standard. 2.5ml of saturated sodium carbonate (Na_2CO_3) solution and 1ml of follin-C reagent were added up to each flask and volume made up to 50ml and mixed well. After standing for $1^{1/2}$ hour, the sample was filtered using whatman no.4 grade of filter paper and the absorbance measured at 760 nm against reagent blank.

$$\text{Tannin (Mg/100g)} = \frac{\text{Standard concentration} \times \text{Sample absorbance}}{\text{Standard Concentration} \times \text{weight of sample}} \times 100$$

2.4.5 Trypsin inhibitor Activity Determination

Trypsin inhibitor activity was assayed by a procedure of (AOAC, 2012). Sample extract (0.1ml) and 0.9ml of 0.1mol/L phosphate buffer of pH 8.0 was mixed with the same volume of trypsin and pre incubated at 37°C for 5minutes; 1ml of 0.03% (w/v) Bovine serum Albumin (BSA) was added to the mixture and incubated for 30mins at 37°C after which the reaction was stopped by the addition of 2ml of 5% (w/v) Trichloroacetic acid (TCA) solution. The mixture was filtered. To 1ml of the filterate, 5ml of 0.55 mol/L Na_2CO_3 and 0.1ml of follin- C reagent. The resulting colour absorbance was determined at 660nm wavelength. Standard sample was prepared in the absence of inhibitors.

$$\%\text{Trypsin Inhibitor} = \frac{T - T^{\circ}}{T} \times 100$$

Where T is the absence of inhibitors; T° is absorbance in the presence of inhibitors.

2.5 Data Analysis

The data obtained were subjected to analysis of variance (ANOVA) using Statistical Package for Social Sciences (SPSS) Version 20 to compare the means among different antinutritional compositions of the two (2) Cassava varieties in the different locations. Where significant differences exist, Duncan's multiple range test (DMRT) was used to separate the means at p≤0.05 level of significance. T-test was also used to compare the antinutritional compositions between the two (2) varieties.

3. Results and Discussion

3.1 Results

3.1.1 Anti-nutritional Compositions of White Cassava (Manihot esculenta) from three different locations within Kogi State

Table 1 below showed the mean antinutrient compositions of white cassava variety from three different locations within Kogi State. Cassava tubers from Okene had the highest cyanide and trypsin inhibitor compositions (3.60±0.01mg/100g and 20.00±0.00%) respectively, while samples from Ankpa (0.51 ± 0.00mg/100g and 8.88 ±0.00%) had the lowest compositions respectively. Tannin contents was found to be highest in cassava from Ankpa (0.31 ± 0.1mg/100g) and lowest in Kabba and Okene(0.10±0.00mg/100g). Furthermore, cassava varieties from Kabba (1.71±0.04mg/100g) had the highest phytate compositions while Ankpa (0.61 ± 0.04mg/100g) had the lowest. However, the oxalate content of white cassava from Kabba (0.18 ± 0.00mg/100g) was found to be the lowest, with Okene (0.36 ± 0.00mg/100g) having the highest composition at P ≤ 0.05.

Table 1: Anti-nutritional Compositions of White Cassava (*Manihot esculenta*) from three different locations within Kogi State

Location	Cyanide (Mg/100g)	Tannin (Mg/100g)	Phytate (Mg/100g)	Oxalate (Mg/100g)	Trypsin Inhibitors (%)
Ankpa	0.51 ± 0.00^{c}	0.31 ± 0.1^{a}	0.61 ± 0.04^{c}	0.21±0.00^{b}	8.88 ±0.00^{b}
Kabba	3.27 ± 0.01^{b}	0.10 ± 0.00^{b}	1.71 ± 0.04^{a}	0.18±0.00^{c}	5.00 ± 0.00^{c}
Okene	3.60 ± 0.01^{a}	0.10 ± 0.00^{b}	0.80 ± 0.07^{b}	0.36±0.00^{a}	20.00 ± 0.00^{a}

Means were expressed as mean ± Standard deviation (S.D). The level of significance was taken at P ≤ 0. 05. Ducan multiple range test was used to separate the means where significant differences exist

3.1.2 Anti-nutritional Compositions of Yellow Cassava (Manihot esculenta) from three different locations within Kogi State

Table 2 below showed that; at P ≤ 0.05, Cassava tubers from Okene had the highest cyanide and trypsin inhibitor (1.80±0.01mg/100g and 15.38±0.00%) compositions respectively, while samples from Ankpa (0.31±0.00mg/100g and 4.10±0.82%) had the lowest compositions respectively Also, phytate and oxalate compositions was found to be highest in cassava from Ankpa (3.31± 0.10mg/100g and 0.23±0.00mg/100g) and lowest in Okene (1.78±0.04mg/100g and 0.20±0.00mg/100g) Furthermore, Tannin content of cassava from Okene (0.33±0.00mg/100g) was found to be highest with Kabba and Okene (0.09±0.00mg/100g) having the lowest compositions.

Table 2: Anti-nutritional Compositions of Yellow Cassava (*Manihot esculenta*) from three different locations within Kogi State

Location	Cyanide (Mg/100g)	Tannin (Mg/100g)	Phytate (Mg/100g)	Oxalate (Mg/100g)	Trypsin Inhibitor (%)
Ankpa	0.31 ± 0.01^{c}	0.09 ± 0.00^{a}	3.31 ± 0.10^{c}	0.23±0.00^{b}	4.10 ±0.82^{b}
Kabba	1.65 ± 0.00^{b}	0.09 ± 0.00^{b}	2.70 ± 0.07^{a}	0.22±0.00^{c}	8.33 ± 1.67^{c}
Okene	1.80 ± 0.01^{a}	0.33 ± 0.00^{b}	1.78 ± 0.04^{b}	0.20±0.00^{a}	15.38 ± 0.00^{a}

Means were expressed as mean ± Standard deviation (S.D). The level of significance was taken at P ≤ 0.05. Ducan multiple range test was used to separate the means where significant differences exist.

3.1.3 Comparism between the White and Yellow Cassava

The result from the T-test in Table 3 below showed that; significant differences were observed in the Cyanide (0.042), Phytate (0.00) and Oxalate (0.017) and Trypsin inhibitor (0.481) content between both varieties of cassava from the different locations in this study. However, no significant differences was observed between the yellow and white cassava in their Tannin (0.983) content. The white cassava variety was found to highest in cyanide (2.46±1.46mg/100g), oxalates (0.28±0.08mg/100g) and trypsin inhibitor (11.29±6.74%) compositions while the yellow cassava was highest in phytate (2.59±0.67mg/100g) and both varieties have the same tannin content (0.17± 0.10mg/100g) .

Table 3: Comparism of anti-nutrient compositions between the white and yellow cassava

Variety	Cyanide (Mg/100g)	Tannin (Mg/100g)	Phytate (Mg/100g)	Oxalate (Mg/100g)	Trypsin Inhibitors (%)
White	2.46 ± 1.46[a]	0.17 ± 0.10[a]	1.04 ± 0.51[b]	0.28 ± 0.08[a]	11.29 ± 6.74[a]
Yellow	1.26 ± 0.71[b]	0.17 ± 0.10[a]	2.59 ± 0.67[a]	0.21 ± 0.01[b]	9.27 ± 5.02[b]
P-Value	0.042	0.983	0.00	0.017	0.481

Means were expressed as mean ± Standard deviation. The level of significance was taken at P ≤ 0.05. T-test was used to compare the anti-nutrient compositions between the white and yellow cassava

3.1 Discussion

From the results above in tables 4.1, and 4.2, it was observed that the mean cyanide compositions of cassava samples from the three locations (Ankpa, Kabba and Okene) in Kogi State showed significant differences at P ≤ 0.05 and ranged between 0.51±0.00mg/100g and 3.60±0.01mg/100g for the white and 0.31±0.01mg/100g and 1.80 ±0.01mg/100g for the yellow variety. These values falls below the range of the findings of a group researchers that recorded between 0.98±0.05mg/100g and 6.85±0.00mg/100g in their work on six varieties of cassava grown in international institute for tropical agriculture (IITA), Ibadan, Nigeria and the work of that recorded between 11.29±0.19mg/100g and 19.29±0.19mg/100g on six varieties of cassava grown and sold in Selangor Malaysia (Oresugun et al., 2016; Siti and Aishah, 2016). The cyanide content of cassava are the most toxic compounds limiting the consumption of cassava tubers (Emmanuel et al., 2012). It was reported by a group researcher that, consumption of high amounts of cyanide or improperly processed cyanide for a long time could induce certain diseases such as neuropathy, goiter, epilepsy and often lead to death (Gonclaves et al., 2016). However, in a very limited study, cyanides have been shown to have beneficial cardiovascular effects at very low concentrations (Parikh et al., 2018).

Tannin however, impair with cassava starch and disaccharide assimilation and interact with proteolytic enzymes inhibiting their activity (Krupa, 2008; Agarwal, 2016). The results obtained in this study showed that; total tannin contents for white cassava ranged between 0.10± 0.1mg/100g and 0.31 ± 0.01mg/100g while yellow cassava had values between 0.09±0.00mg/100g and 0.33±0.00mg/100g within the three locations in this study. This observed value agrees with the value observed by a researchers that recorded between 0.20±0.10mg/100g and 0.30±0.00mg/100g in their work on two cassava varieties grown and eaten in Ovia south local government area of Edo State, Nigeria. Phytates on the other hand bind with minerals such as zinc, calcium and iron to form a soluble complex, thereby reducing mineral bioavailability (Obueh and Kolawale, 2016; Kumar et al., 2009). The result in this study indicates that white cassava tubers has a phytate composition ranging between 0.61 ± 0.04mg/100g and 1.71 ± 0.04mg/100g while yellow cassava ranged between 1.78 ± 0.04mg/100g and 3.31 ± 0.10mg/100g . This observed value is lower than the values observed by who recorded between 112.82 ± 0.00mg/100g and 225.64 ± 0.00mg/100g on his work on pupuru produced by steeping cassava in different types of water and the work of that recorded from 216 mg/100g to 304 mg/100g phytate composition (Oyetayo, 2006; Sarkayiya and Agar, 2010). The significant differences between the recorded values and the work above is due to different locations involved in the study. Consequently, Phytates have been shown to cause muscular weakness and sometimes paralysis when in high concentration in the body (Soetan and Oyewole, 2009). However, *invitro* and *invivo* studies have shown that phytic acid may have beneficial effects in the prevention and treatment of several pathological conditions and cancer (Silva and Bracarense, 2016).

For oxalates, the recorded values ranged from 0.18±0.00mg/100g to 0.36±0.00mg/100g for the white variety and 0.20 ±0.00mg/100g to 0.23+0.00mg/100g for the yellow variety. Compared to the work of most of researchers that recorded an oxalate content of 13.56mg/100g and 1.76±1.5mg/100g respectively, the observed values were found to be lower (Olaoye et al., 2015; Wobeto et al., 2007). High amount of oxalates in the body bind calcium, leading to formation of crystals or excretion through urine. The crystals formed majorly contribute to kidney stones in humans (Massey et al., 2007). The low oxalate content compared to other works indicates that dwellers of kogi State who consume these cassava varieties are not at risk of kidney stones compared to other regions.

Furthermore, trypsin inhibitor compositions was recorded between 8.88±0.00% and 20.00±0.00% for white cassava and between 4.10±82% and 15.38±0.00% for the yellow cassava. The value observed was found to be a little higher than the value recorded by which is 4.0±0.00% for white cassava variety (Sarkiyayi and Agar, 2010). The observed value however, is in alignment with the findings of who worked on African yam bean and recorded 6.67±0.33% trypsin inhibitor content (Ndidi et al., 2014). High amount of trypsin inhibitors in foods have been shown to cause pancreatic enlargement and growth depression in humans (Soetan and Oyewole, 2009). It can however be reduced in cassava grown in Kogi state and other regions via processing strategies like soaking, heat treatment, and fermrntation (Parul, 2014).

Summarily, from table 4.3 above, significant differences were observed in all the parameters studied except for tannin and trypsin inhibitors. The white cassava variety was found to be higher in cyanide (1.26±1.46mg/100g) oxalates (0.28±0.08mg/100g) and trypsin inhibitor (11.29±6.74%) compositions while the yellow cassava was highest in phytate (2.59±0.67mg/100g) and both varieties have the same tannin content (0.17±0.10mg/100g). The white cassava was observed to be the cassava variety with the highest concentration of all compared antinutrients except for phytates and tannins.

Anti-nutrients present in cassava and other crops can be reduced by several processing methods such as soaking, cooking, boiling, germination, autoclaving, fermentation, sprouting and other processing methods, which however may interfere with the level of protein and fibre contents used as indicator of high nutritive value as reported (Liyanage et al., 2014). However, fermentation drastically reduce or totally eradicate the cyanide and most antinutritional content of cassava and substantially improve the nutritional quality. Heat treatment such as cooking or boiling also have a greater efficiency in the elimination of heat labile antinutrient (oxalates, cyanides, trypsin inhibitors) since they are very sensitive to standared temperatures (Gemede and Ratta, 2014; Mihrete, 2019).

4. Conclusion

From the study, there was significant variations in the anti-nutrients compositions of the two cassava varieties grown and eaten in three areas (Ankpa, Kabba and Okene) of Kogi state. From the locations studied, white cassava tubers from Okene had the highest value for all the anti-nutrients analysed except for phytate and tannin where Kabba and Ankpa was highest respectively. Similarly, Okene had the highest composition of all the antinutrient in the yellow cassava except for phytate and oxalate where Ankpa took the lead. Therefore, families living in Okene could be at risk of several health complications such as goitre, cretinism among children, neuropathy and growth depression compared to other locations and as such, cassava should be extensively processed in this area prior to consumption. The white cassava variety had the highest content of all the anti-nutritional factors analyzed except in phytates where the yellow cassava outperformed it and tannins where both varieties had the same value. Hence, the yellow cassava variety will be safer and healthier for use in human consumption and livestock feed formulation across the three locations in Kogi State compared to the white cassava.

Recommendation

The anti-nutritional factor of the two cassava varieties (White and yellow varieties) used in this work can provide useful information regarding the selection of both desirable and potential variety for human and animal consumption; it is therefore recommended that further research work be carried out on this variety to determine the best of them for incorporation in human and livestock feed. Also, other nutritional analysis should be conducted on both varieties to know other compositions they contain. However, it is important to note that before cassava can be used in human and animal feed in Kogi state and other regions, it has to be processed and detoxified by several processes such as soaking, boiling, heat treatment and fermentation to the level which is safe before it can be incorporated in their feed. This will help in reducing the negative effect of the anti-nutritional factors and as well improve the nutritional quality of these varieties.

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