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Nutrients, the Bioavailability of Micronutrients and Antinutrient Composition of African Yam Bean Tubers

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ABSTRACT

Despite the huge nutritional and nutraceutical benefits associated with the consumption of African yam bean (AYB) seeds, AYB tubers are still underutilized in Nigeria, they are left to rot in farmers' fields. To create awareness about their nutrient density and provide information on the bioavailability of micronutrients in the tubers of AYB accessions, raw tubers of TSs 9, TSs 60 and TSs 93 were analyzed using standard laboratory procedures. The AYB tubers had the following composition on a fresh weight basis: crude protein (7.55-8.27%), carbohydrate (9.36-11.12%), crude fat (0.18-0.22%), crude fibre (0.33-0.750%), crude ash (0.44-0.59%), and moisture (79.76-80.82%). The fresh tubers were rich in minerals: potassium (532.88-557.80 mg/100 g), calcium (205.75-467.50 mg/100 g), magnesium (124.75-168.50 mg/100 g), iron (15.71-27.30 mg/100 g), zinc (8.57-14.80 mg/100 g), and manganese (0.68-1.98 mg/100 g). The phytate: Zn and the phytate: Fe molar ratios indicated the bioavailability of Fe and Zn in the raw AYB tubers. The results of this study indicate that AYB tubers are good sources of protein, energy and bioavailable Fe and Zn and should be utilized for human diets and livestock feed. With good processing, AYB tubers could contribute to food and nutrition security. There is a need for research on food products developed from AYB tubers to enhance their utilization and large-scale production in Nigeria.

Keywords: African yam bean, tubers, bioavailability, nutrient-rich, proximate, antinutrients.

Introduction

African yam bean (AYB) is an important pulse and tuber crop. AYB is one of the multipurpose tropical legumes of African origin with huge potential for food and nutrition security, and soil health that is currently underexploited. The seeds are rich in energy, protein, and fibre, low in fats and micronutrient-dense. [1] African yam bean is a promising legume that enriches the soils through the decomposition of leaves, hence, increasing soil organic matter. It produces root nodules that have been associated with high seed yield and the capacity to enhance the yield of component crops in intercropping systems.[2] AYB is however faced with some challenges that have negatively affected its production and led to its underutilization and neglect. Major constraints facing the crop include; lack of improved varieties, high production costs due to staking its vine (staking is a major agronomic practice for optimum yield), and genetic erosion of AYB biodiversity in major growing areas, especially in Nigeria. These have resulted in low yields and reduced farmers' interest in including it in their food production systems.[3, 4] This crop was once a major staple in Nigeria decades ago, but due to the preference of pulse consumers and end-users for other grain legumes to AYB seeds (especially those with shorter cooking time), poor funding and low research efforts in AYB and lack of quality food products being developed from AYB, the crop is gradually facing the risk of being extinct.

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African yam bean is grown for its edible seeds in Nigeria while most people that consume and grow AYB seeds do not know that it produces tubers. Farmers that know that the crop produces tubers do not consume them because they claim they do not know how safe the tubers are for human consumption. Hence, AYB tubers are left to rot on AYB farmers' fields across Nigeria and some West African countries. Studies on AYB tuber nutrition are scarce in literature when compared with the volume of nutrition research that has been documented for other root and tuber crops. Earlier nutritional studies on AYB tubers examined only their proximate and mineral contents.[6-7] Bioavailability of minerals in AYB tubers is also scarce in the literature. Evaluating the bioavailability of edible portions of crops is one of the strategies for providing useful information that can create awareness among potential consumers on the promising opportunities in them for enhanced human nutrition and health and livestock feed. Valid information on antinutrients in edible portions of crops is one of the major pieces of information that is needed to determine how safe they are for human and or livestock consumption. This information is especially important in countries where an edible portion of a neglected and underexploited crop is not yet utilized for human dietary preparations and livestock feed.

Bioavailability studies on food crops could also provide useful information that could guide food and nutrition experts on how to process and develop healthy diets and products from them for human consumption. It could also provide useful information that could guide the plant breeders in improving the nutritional qualities of crops and reducing their antinutritional factors through selection and hybridization. This study was initiated to determine the nutrients and anti-nutrients in tubers of AYB accessions and the bioavailability of the minerals to create awareness about its nutritional composition and how safe it is for human consumption. This could enhance AYB tuber utilization, production and improvement.

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Materials and Methods

African yam bean tuber collection and preparation

The fresh tubers of AYB accessions analyzed in this study were collected from the agronomic evaluation of AYB accessions on the 6th of December, 2022. The AYB seeds used for the agronomic evaluation were gotten from the AYB collections of the International Institute of Agriculture (IITA) Ibadan and preserved at the gene bank of the Genetic Resources Centre of IITA. The mature AYB tubers of TSs 9, TSs 60 and TSs 93 were harvested from the trial field. The trial was conducted at the Research farm of the Ikole Campus of the Federal University, Oye-Ekiti, Nigeria in 2021. The proximal and the distal ends of the fresh AYB tubers were cut off and washed with distilled water. The leftover distilled water on the washed tubers was properly drained from them. Thereafter, the tubers were cut into pieces and ground using mortar and pestles. The ground samples of the AYB tubers were sealed in plastic containers, refrigerated and used for the various biochemical analyses using standard laboratory procedures outlined below.

Determination of the proximate composition of the fresh AYB tubers
The AOAC[8] approach was used for the proximate analysis. Kjeldahl AOAC technique was used to calculate crude protein (955.04). Using the oven-drying AOAC method, the moisture content of fresh AYB tubers was determined (930.15). The AOAC method for acid hydrolysis was used to determine the fresh AYB tuber's crude fat content (954.02). The dry ashing AOAC method was used to determine the fresh tubers' ash content (920.117). By employing the AOAC method, crude fibre analysis was performed (978.10). The following formula was used to determine the quantity of carbohydrates present in fresh AYB tubers:

AYB tuber carbohydrate content = 100% - (%moisture + %protein + %fat + %ash + %fibre).

Determination of the mineral composition of the fresh AYB tubers
To analyze minerals, AOAC[8] method 968.08 was used. Using a Buck Scientific Atomic Absorption Spectrophotometer (Model: 210VGP) and particular cathode lamps for each metal, the metal concentrations in samples of digested fresh AYB tuber were calculated. With the use of a flame photometer, sodium and potassium were measured (Model: FP10). Based on calibration curves of metal standard solutions, the metals were quantified. Each batch of analysis contained blanks and verified reference standards were utilized to evaluate the analytical method's precision.

Anti-nutrient determination in the fresh African yam bean tubers
The hydrogen cyanide contents of the fresh AYB tubers were determined using the Onwuka method[9]. The analytical technique developed by Obdoni and Ochuko was used to determine their saponin contents.[10] Total alkaloid was determined in the fresh AYB tubers using the Harbone method.[11] Total phenolic content of the samples was determined according to Xu and chang with slight modifications[12]. The phytate content of the fresh AYB tubers was determined by the method of Davis and Reld as modified by Abulude[13]. AOAC[8] technique 915.03 was used to determine the oxalate concentration of fresh AYB tubers.

Calculation of molar ratios
The phytate: Zn, phytate: Fe, Ca: phytate, Ca x phytate: Zn molar ratios were calculated using the as previously described by Norhaizan and Faizadatul.[46] Molar ratios between the minerals and antinutrients were obtained by dividing the mole of the phytate by the moles of the minerals.

Statistical analysis
Utilizing the R statistical analysis software version 4.1.1,[14] all data collected were analyzed. The significance of the treatment means was determined by Fisher's least significant difference (F-LSD) at the 5% probability level in the proximate qualities, antinutrients, minerals and antinutrients studied in the fresh tubers of the AYB accessions.

Results and Discussion

The results of the proximate composition of the AYB tubers are shown in Table 1. The proximate composition was significantly ($p<0.05$) influenced by accession except for the crude protein (CP) and moisture contents of the tubers. The crude protein content of the AYB tubers ranged from TSs 93 (7.55%) to TSs 60 (8.27%). Crude ash contents of the AYB tubers were significantly ($p<0.05$) influenced by accession. TSs 60 (0.59%) was significantly ($p<0.05$) higher than TSs 93 (0.44%) which had the least ash content. The crude fibre was significantly ($p<0.05$) higher in the tubers of TSs 93 (0.83%) than TSs 60 (0.75%) although it was statistically ($p>0.05$) similar to TSs 9 (0.81%). The crude fats of the AYB tubers were significantly ($p<0.05$) influenced by accession and ranged from TSs 9 (0.18%) to TSs 60 (0.22%). The carbohydrate content of the AYB tubers differed significantly ($p<0.05$). TSs 9 (11.12%) was significantly higher than TSs 60 (9.36%) but was statistically similar to the carbohydrate content of the tubers of TSs 93 (11.10%). The fresh tubers of the AYB accessions analyzed were statistically ($p>0.05$) similar in their moisture contents and ranged from TSs 9 (79.76%) to TSs 60 (80.82%). The results of the proximate analysis have revealed that AYB tubers are rich in protein and energy but low in fat. The fresh AYB tubers analyzed in this study were superior in terms of protein content to fresh cassava roots with 1.20% protein.[15] The CP contents of the fresh AYB tubers analyzed in this study were also higher than the mean CP of 4.32% (on a dry weight basis) reported for tubers of 43 yam genotypes.[16] The AYB tubers studied had protein levels that were higher than most of the commonly consumed tuber crops. This implies AYB tuber could contribute significantly to mitigating the effects of protein-energy malnutrition among those that grow and consume it. This could also contribute to the protein content of animal/livestock feed when AYB tubers are being utilized in livestock feed production.
The results of the mineral composition of the fresh tubers of the AYB accessions are shown in Table 2. The mineral concentrations of the fresh AYB tubers varied significantly ($p<0.05$) except for potassium. Potassium ranged from TSs 60 (532.88 mg/100 g) to TSs 9 (557.80 mg/100 g). Fresh tubers of TSs 60 (168.50 mg/100 g) were significantly ($p<0.05$) higher in magnesium concentration while TSs 9 had the least magnesium content (124.75 mg/100 g). Fresh tubers of TSs 60 had high calcium (467.50 mg/100 g) which was more than double the calcium contents of TSs 9 (207.75 mg/100 g). Fresh tubers of TSs 9 (27.30 mg/100 g) had the highest iron content followed by TSs 60 (19.35 mg/100 g) while TSs 93 (15.71 mg/100 g) had the least iron content. A similar trend was also recorded in the zinc content of the fresh AYB tubers. TSs 9 (14.80 mg/100 g) was significantly highest in zinc while the fresh tubers of TSs 93 (8.57 mg/100 g) recorded the least zinc content. Fresh tubers of TSs 60 (1.98 mg/100 g) were most prominent for manganese content while TSs 93 (0.68 mg/100 g) had the least manganese content.
The fresh AYB tubers analyzed were very rich in minerals. The AYB tubers were especially rich in potassium, magnesium, calcium and iron and zinc. The tubers were richer in these minerals than what has been reported for yam tuber by Bhandari et al.[17] and cassava root by Adeniji et al.[18] and higher than what has been reported for potassium, calcium, magnesium, zinc for white yam and cocoyam by Alinor and Akalez.[19] Micronutrient malnutrition (hidden hunger) has remained a major public health challenge among both the elites and the resource poor in Nigeria. Considering their nutrient density, the consumption of processed AYB tubers by humans and livestock could help to reduce micronutrient malnutrition in Nigeria where the use of food supplements is not common.

Anti-nutrient contents of the African yam bean tuber
Table 3 shows the anti-nutrient contents of the fresh AYB tubers. Most of the anti-nutrients were not significantly different ($p>0.05$) influenced by AYB accession; except hydrogen cyanide (HCN) and phenol. TSs 9 (0.98 mg/100 g) had the least HCN content while TSs 60 (1.40 mg/100 g) recorded significantly higher HCN than the other

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two accessions. The lethal dose range for humans for HCN taken by mouth is estimated to be 0.5 to 3.5 mg/kg body weight.[21] The AYB tubers were generally very low in HCN and were much lower than what has been reported in sweet cassava roots (60 mg/kg) and the 1000 mg/kg found in the roots of bitter cassava varieties. [15] They found out that conventional cooking methods could reduce the HCN of the cassava root to a harmless level.[15] Traditional processing methods that include cooking have been found to reduce HCN in cassava by up to 99%.[20] The results implied that the HCN levels found in the AYB tubers were much lower than the safe levels for HCN. Hence, the consumption of well-processed products made with AYB tubers should not lead to HCN toxicity.

The saponins contents of the AYB tubers ranged from TSs 93 (0.86 mg/100 g) to TSs 9 (0.92 mg/100 g) (Table 3). The AYB tubers analyzed in the study were also very low in saponin compared to what has been reported in other leguminous crops. Duhan et al.[22] found 2164 mg/100 g seed saponin content in *Cajanus cajan*. Avanza et al.[23] found 110-820 mg/100 g seed saponin content in *Vigna unguiculata* varieties. The level of saponins in AYB tuber therefore should not be a threat to its consumption, because the values were in the fresh AYB tubers. These levels of antinutrients in the AYB tubers could be reduced with appropriate processing methods. Saponins have, however, been reported to reduce the risk of cardiovascular diseases, and increase the elimination of bile acids.[26] Studies have reported that saponins have anti-carcinogenic, anti-mutagenic, immune modulatory activity and hypocholesterolemic properties.[24-26] Cooking, fermentation, sprouting, boiling and soaking are useful in the reduction in saponin contents of foods to safe levels. [24-26]

The alkaloid contents of the AYB tubers were not significantly ($p>0.05$) influenced by accession. TSs 60 (30.32 mg/100 g) had the least alkaloid content while TSs 9 (31.85 mg/100 g) recorded the numerically higher alkaloid content than the other two accessions although the accessions were statistically similar for this parameter (Table 3). Alkaloids in the AYB tubers were higher than what has been reported in yam tubers,[27] but lower than the alkaloid level reported in Bambara groundnut.[28] Akpe *et al*.[27] however reported that cooking reduced the alkaloids in yellow yam, sweet potatoes and Irish potatoes by 74.5%, 71.4%, and 62.7% respectively.

Table 1: Proximate composition (%) of African yam bean tubers

Accession	Crude protein	Crude Ash	Crude Fibre	Crude Fat	Carbohydrate	Moisture
TSs 9	7.56^{a}	0.58^{a}	0.81^{a}	0.18^{b}	11.12^{a}	79.76^{a}
TSs 60	8.27^{a}	0.59^{a}	0.75^{b}	0.22^{a}	9.36^{b}	80.82^{a}
TSs 93	7.55^{a}	0.44^{b}	0.83^{a}	0.19^{b}	11.10^{a}	79.89^{a}
Mean	7.789	0.54	0.797	0.19	10.52	80.16
F-LSD$_{(0.05)}$	NS	0.08	0.04	0.02	0.99	NS
CV$_{(\%)}$	6.736	9.46	3.35	6.48	5.88	0.89

Means in the same column with the same letters are not significantly different at 95% level of confidence

Table 2: Mineral composition (mg/100 g) of African yam bean tubers

Accession	Potassium	Magnesium	Calcium	Iron	Zinc	Manganese
TSs 9	557.80^{a}	124.75^{b}	205.75^{c}	27.30^{a}	14.80^{a}	1.21^{b}
TSs 60	532.88^{a}	168.50^{a}	467.50^{a}	19.35^{b}	12.18^{b}	1.98^{a}
TSs 93	538.75^{a}	133.50^{b}	354.00^{b}	15.71^{b}	8.57^{c}	0.68^{c}
Mean	543.14	142.25	342.42	20.79	11.85	1.29
F-LSD	NS	33.07	89.5	4.90	1.95	0.28
CV$_{(\%)}$	13.80	14.53	16.35	14.73	10.29	13.48

Means in the same column with the same letters are not significantly different at 95% level of confidence NS= non-significant at 95% level of confidence ($p>0.05$)

Tubers of TSs 60 (92.03 mg/100 g) were significantly ($p<0.05$) higher than TSs 9 (61.05 mg/100 g) in their phenol concentration (Table 3). The phenolic compounds in the AYB tubers were lower than documented levels in dry beans 117.8-157.6 mg/g, Faba bean cultivars, 11.2 -48.3 mg/g in dry beans[28-29] and the 0.57-1210 mg/100 g reported for various pulses.[26] Phenolic compounds have been implicated in health. They are considered bioactive molecules. Different studies reported that phenols have cardio-protective, anti-microbial, anti-hypertensive, anti-inflammatory properties, anti-carcinogenic, antioxidant, and immune-modulating effects, lowering the risk of colon cancer and osteoporosis.[25,30-32,28] There is a need for further studies on the effect of various processing methods on the phenolic contents of AYB tubers. Decoating, cooking, soaking, boiling, fermenting and milling have been found to enhance the reduction of phenols in legumes seeds.[24]

The phytate contents of the AYB tubers ranged from TSs 93 (116.67 mg/100 g) to TSs 60 (128.26 mg/100 g). The phytate contents of the fresh AYB tubers were within the range of phytate contents (0.8-2291 mg/100 g) reported for major staples[33-35;26]. Nevertheless, phytate has important health benefits such as anti-carcinogenic properties, antineoplastic effects and anti-oxidant activities.[26] Maphosa and Jideani,[25] and Geraldo et al. [26] have reported the effectiveness of autoclaving, fermenting, germinating, roasting, steaming, boiling, soaking, de-hulling and gamma irradiating in reducing phytate contents in legumes.

The oxalate contents of the fresh AYB tubers ranged from TSs 93 (158.30 mg/100 g) to TSs 60 (169.70 mg/100 g). The oxalate contents of the AYB tubers were lower than what has been reported in *Dioscorea bulbifera* (aerial yam) tubers[17] and falls within the range of the oxalate contents of major grain legumes.[34,36-37] The levels of oxalate in these AYB tubers should not be a major limitation to consuming them by normal healthy humans, as more than 1 kg of AYB tubers analyzed in this study would be consumed at once to ingest 2 g of oxalate which is considered to be a fatal dose to humans.[38] Steaming, boiling, soaking and combining with foods with high calcium content have been found to greatly reduce oxalate concentration in foods.[39-40]

The effects of cooking and other processing methods on antinutrients require further studies in AYB tubers. One of the major reasons why millions of tonnes of AYB tubers are left to rot in the soil in Nigeria and other parts of West Africa is due to the uncertainty of their safety for food and livestock feed. The results of this study have shown that the anti-nutrients analyzed in the AYB tubers were within the range of what is found in common staples and did not differ significantly from what has been documented for antinutrients in raw grain legumes and tuber crops.

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Table 3: The anti-nutrient concentrations (mg/100 g) of African yam bean tubers

Accession	Hydrogen Cyanide	Saponins	Alkaloids	Phenols	Phytate	Oxalate
TSs 9	0.98 ^{c}	0.92^{a}	31.85^{a}	61.05 ^{b}	124.16^{a}	162.95^{a}
TSs 60	1.40^{a}	0.89^{a}	30.32 ^{a}	92.03^{a}	128.26 ^{a}	169.70^{a}
TSs 93	1.27 ^{b}	0.86^{a}	31.74^{a}	84.05^{ab}	116.67 ^{a}	158.30^{a}
Mean	1.22	0.89	31.30	79.04	123.03	163.65
F-LSD$_{(0.05)}$	0.11	NS	NS	26.20	NS	NS
CV$_{(\%)}$	5.695	14.61	9.37	20.72	9.72	6.194

Means in the same column with the same letters are not significantly different at 95% level of confidence NS= non-significant at 95% level of confidence ($p>0.05$)

Table 4: Molar ratios of micronutrients of African yam bean tubers

Accession	phytate: Zn (± SD)	phytate: Fe (± SD)	Ca: phytate (± SD)	Ca:phytate:Zn (± SD)
TSs 9	0.85 ± 0.19^{b}	0.39 ± 0.01^{b}	27.45 ± 1.72^{b}	4.39 ± 1.26^{b}
TSs 60	1.05 ± 0.08^{b}	0.58 ± 0.13^{a}	59.73 ± 8.87^{a}	12.13 ± 1.46^{a}
TSs 93	1.36 ± 0.19^{a}	0.63 ± 0.02^{a}	50.74 ± 10.05^{a}	11.84 ± 0.56^{a}
F-LSD$_{(0.05)}$	0.26	0.119	12.48	1.85
CV$_{(\%)}$	14.89	14.09	16.97	12.26

Means in the same column with the same letters are not significantly different at 95% level of confidence

Selecting and breeding for low antinutritional factors in AYB tuber should be one of the objectives of future AYB breeding programmes. Some of the earlier reported studies have variously documented the effectiveness of various processing methods in reducing antinutrients. It is therefore important for nutritionists and food scientists to begin to look into the effects of various processing methods on the nutritional and antinutritional factors in AYB tubers. This will enhance the utilization of this edible part of AYB, in Nigeria and West Africa. AYB tubers are already being consumed by humans in Southern African Countries.[5]

Bioavailability of zinc, iron and calcium in the fresh tubers of AYB accessions

Phytate: Zinc

Table 4 shows the inter-relationships among zinc (Zn), iron (Fe), calcium (Ca) and phytate (phy) in the fresh tubers of the AYB accessions. The phy: Zn ranged from TSs 9 (0.85) to TSs 93 (1.36). The calculated phy: Zn molar ratios for the raw AYB tubers were less than the levels suggested to be critical level (ratios less than 15 are regarded as excellent for zinc bioavailability) (Morris and Ellis (1989).[41] This suggests that Zn bioavailability (absorption) in the tubers of the AYB accessions studied will not be impaired in AYB tubers. The phytate: Fe (phy: Fe) ratio in the fresh tubers of the AYB accessions ranged from TSs 9 (0.39) to TSs 93 (0.63). The phy: Fe molar ratios were below 1 (indicative of favourable iron bioavailability).[42] The low phytate: Fe indicates good iron absorption in AYB tubers. The Ca: phytate (Ca: phy) ranged from TSs 9 (27.45) to TSs 60 (59.73). The critical value for Ca: phy is 6.1.[43] The Ca: phy was above the critical value in the raw AYB tubers, which suggests calcium absorption may be affected in raw AYB tubers. A better picture of Ca: phy molar ratios in the AYB tubers will be better seen in the processed AYB tuber. Therefore the need for proper processing of AYB tubers to reduce the phytate levels and adding calcium-rich foods in the preparation of AYB tuber meals are required for enhanced nutrition of AYB tuber based-meals. A more sustainable means of reducing phytate in AYB tuber is through selecting and breeding low-phytate AYB tuber varieties that combine with high nutrient density. Ca: phy higher than the critical levels have also been reported in *Dioscorea* spp.[16;44] The Ca x phy: Zn ratio was lowest in TSs 9 (4.39), while TSs 60 (12.13) had the highest Ca x phy: Zn content. Ellis et al.[45] reported that the ratio of Ca x phy: Zn is a better predictor of zinc bioavailability. They stated that when Ca x phy: Zn is greater than 0.5 mol/kg, there will be interferences with the availability of zinc. The Ca x phy: Zn of the raw AYB tubers were higher than the critical values, suggesting that Zn absorption may be impaired in the raw AYB tubers. Most tropical root and tuber crops are not consumed raw without processing. Impaired Zn bioavailability will therefore not be a challenge in processed food products made from AYB tubers. Processing methods like cooking, soaking, steaming and roasting could be explored in reducing phytate in AYB tubers in order to further enhance the Zn absorption. Ojewumi[48] noted that the rate at which indigenous crops including legumes are being neglected and going into extinction has become increasingly alarming. A major reason for the neglect has been attributed to a lack of relevant information about their nutritional and medicinal values.[48] Selecting and breeding for improved nutritional composition and especially low phytate in AYB tubers will further enhance the bioavailability of minerals for enhanced nutrition of humans and livestock that will consume products made from AYB tubers.

Conclusion

This study has revealed AYB tubers as nutrient-dense, and especially rich in protein, energy and minerals. Tubers of AYB are promising and good sources of bioavailable Fe and Zn and could help to reduce protein energy malnutrition and micronutrient deficiencies when food products and livestock feed are made from them. Its antinutrient contents are within the range of what has been reported for antinutrients in other staples. Future AYB breeding programmes could further enhance the bioavailability of minerals and other nutrients in AYB tubers through selection, hybridization and other crop improvement strategies. There is a need for research on food products developed from AYB tubers to enhance their utilization and large-scale production in Nigeria.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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