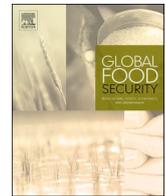




Contents lists available at ScienceDirect

## Global Food Security

journal homepage: [www.elsevier.com/locate/gfs](http://www.elsevier.com/locate/gfs)

# Opportunities and challenges for biofortification of cassava to address iron and zinc deficiency in Nigeria

Ihuoma C. Okwuonu<sup>a</sup>, Narayanan N. Narayanan<sup>b,\*</sup>, Chiedozi N. Egesi<sup>a,c,d</sup>, Nigel Ja Taylor<sup>b</sup>

<sup>a</sup> National Root Crops Research Institute, Umudike, Nigeria

<sup>b</sup> Donald Danforth Plant Science Center, St. Louis, Missouri, USA

<sup>c</sup> International Institute of Tropical Agriculture, Ibadan, Nigeria

<sup>d</sup> Cornell University, Ithaca, NY, USA

## ARTICLE INFO

## Keywords:

Micronutrient deficiency

Nigeria

Biofortification

cassava

Iron

Zinc

## ABSTRACT

Nigeria is the world's largest producer of cassava (*Manihot esculenta*), and its production is important to the country's economy. Cassava's edible storage roots act as a critical staple food for over 180 million Nigerians. Micronutrient deficiency presents a major public health issue in Nigeria and correlates with cassava consumption level across six-agro-ecological zones within the country. Though high in caloric value, cassava roots are deficient in minerals, placing populations that rely on this crop at risk of hidden hunger. Micronutrient deficiencies, especially iron and zinc, affect an estimated 6 million children in Nigeria under five years of age. Supplementation, fortification and food-based diversification are being employed to tackle micronutrient deficiencies. However, in order to achieve wider impact and sustainability, biofortification of staple foods such as cassava is also being explored. Conventional breeding of cassava is unlikely to achieve elevated storage root mineral content at nutritionally significant levels due to lack of genetic diversity for these traits within the existing germplasm. Biofortification by genetic modification provides a potential solution to this challenge. Proof of concept has demonstrated that transgenic biofortification is a reality and can produce foodstuffs with increased mineral content that could beneficially impact the health of consumers in Nigeria and elsewhere. This review is targeted towards understanding the dynamics of micronutrient deficiency across Nigeria and addresses opportunities and challenges for deploying iron and zinc biofortified cassava.

## 1. Introduction

Vitamins and minerals are micronutrients that consumers must obtain from their diet to support normal cellular and physiological functions (Merson et al., 2012). The expression "hidden hunger," is commonly used to convey micronutrient malnutrition, and has subtle and long-term effects across a population. Hidden hunger caused by micronutrient deficiencies affects around 2 billion people across the world (IFPRI, 2016). Women and children under 5 years of age are most prone to micronutrient deficiencies due to insufficient dietary intake resulting from lack of access to quality foods, uneven food distribution within the household and occurrence of infectious diseases (Bailey et al., 2015). The World Health Organization (WHO) has provided global targets for improving the nutrition of women, infants and young children (WHO, 2014). This includes a 40% reduction in stunting of children

under 5 years of age, 50% reduction of anemia in reproductive age women, 30% reduction in low birth weight and childhood wasting reduced to less than 5% (WHO, 2017). These relate to the United Nations Sustainable Development Goals on nutrition geared towards terminating all forms of malnutrition by 2025, with special focus on the needs of adolescent girls, pregnant and lactating women and the aged (<https://www.undp.org/content/undp/en/home/sustainable-development-goals.html>). Iron deficiency is one of the most common and prevalent nutritional disorders in the world, the impact of which is intensified by malaria, worm infestations and infectious diseases such as HIV and tuberculosis (WHO, 2017). Fifty percent of pregnant women and an estimated 40% of preschool children in developing countries are anemic (WHO, 2017). Zinc deficiency in humans is largely due to inadequate intake or absorption of zinc from the diet. It affects about one-third of the world's population, with estimates reaching 73% in some regions

\* Corresponding author.

E-mail addresses: [ihuomaumezurumba@yahoo.com](mailto:ihuomaumezurumba@yahoo.com) (I.C. Okwuonu), [narayanan@danforthcenter.org](mailto:narayanan@danforthcenter.org) (N.N. Narayanan), [cegesi@yahoo.com](mailto:cegesi@yahoo.com) (C.N. Egesi), [taylor@danforthcenter.org](mailto:taylor@danforthcenter.org) (N.J. Taylor).

<https://doi.org/10.1016/j.gfs.2020.100478>

Received 7 February 2020; Received in revised form 1 December 2020; Accepted 5 December 2020

2211-9124/© 2020 Published by Elsevier B.V.

(Prasad, 2013). Zinc deficiency leads to physiological disorders affecting the immune, gastrointestinal, epidermal, central nervous, skeletal and reproductive systems (Roohani et al., 2013). It is now recognized that understanding the epidemiology of micronutrient deficiencies is required to determine effective intervention strategies for a particular region and population (Bailey et al., 2015). Specific data for micronutrient status and intake in children under 5 years of age, pregnant women and women of reproductive age is therefore required at national and local levels to guide development of nutrition interventions and public health programs (Harika et al., 2017).

Iron and zinc biofortification improve the physical and cognitive status of populations impacted by hidden hunger. Independent studies carried out in countries across the world, including Rwanda, India, Mexico and Bangladesh, showed the effect of iron and zinc biofortified crops for alleviating micronutrient deficiencies (<https://www.harvestplus.org/evidence-document>). In Rwanda, regular consumption of iron-biofortified beans amongst women of reproductive age led to significant increases in hemoglobin, ferritin and total body iron within a period of 5 months. Behavioral and cognitive performance were also impacted with improved memory, increased attentiveness and enhanced performance at work and school (Haas et al., 2016). The same effect was achieved amongst adolescent children in India who consumed iron biofortified pearl millet (Finkelstein et al., 2015; Scott et al., 2018). The importance of zinc in maintaining proper functioning of the immune system cannot be overemphasized, considering that virtually all immune cells are zinc dependent (Wessels et al., 2017). Zinc biofortified crops have the potential to improve health conditions such as pneumonia and other respiratory disorders as well as susceptibility to novel viral outbreaks such as COVID-19 (<https://www.harvestplus.org/knowledge-market/in-the-news/coronavirus-pandemic-highlights-need-ensure-good-nutrition>). Antiviral activities of zinc against a variety of viruses through varying mechanisms are well documented (Read et al., 2019).

Here, we review knowledge of staple food production and micronutrient deficiencies across geographical locations in Nigeria, with a focus on cassava production and consumption. This information will be discussed in relation to biofortification of cassava to address nutritional needs within at-risk populations. Nigeria is the focus because it stands first among African countries for the numbers of people impacted by micronutrient deficiency, with 37% of children (6 million) stunted due to malnourishment (UNICEF, 2016). The goal of this study is to illuminate the dynamics of micronutrient deficiency across the different regions of Nigeria, setting the stage for subsequent ex-ante studies and interventions for deployment of iron and zinc biofortified cassava.

There is already an existing biofortification effort in this area anchored by the Harvest Plus program. Thus a significant portion of the information used in the review was sourced from Harvest Plus publications on micronutrient deficiencies carried out in different regions of the country. More than 90% of literature reviews cited are peer reviewed research publications from internationally recognized journals. Articles from national Nigerian journals were also included in some cases due to local relevance of such studies. We intentionally focused on original research and review articles published within the last ten years. This ensured that the information presented was as current as possible. Key reports from international publically supported and trusted organizations such as FAO, ICRISAT, IFPRI, NAERLS, UNICEF, WHO were included in the review of literature.

## 2. Socio-economic status and micronutrient deficiency in Nigeria

### 2.1. Population, geographical and economic diversities in Nigeria

Nigeria is situated on the Gulf of Guinea in Western Africa between Benin in the East and Cameroon in the West. Nigeria is Africa's most populous country with a population of 195.9 million as of July 2018. Total land area is 910,800 Km<sup>2</sup> producing a population density of 215

per Km<sup>2</sup> (<http://www.worldometers.info/>). It is estimated that 50% of Nigerians are rural dwellers, most of whom are subsistence farmers and local traders (Reed and Mberu, 2013).

The country consists of six geopolitical zones divided by similarities in culture, history, background and proximity (Table 1). Climatic conditions in the different geographical areas vary depending on the combined effects of temperature, humidity and rainfall. The Southern part of the country records the highest annual rainfall at more than 2000 mm compared to 500–700 mm in the North. This plays a major role in determining the type of cropping systems, agricultural practices and staple foods available to the populace in these regions ([www.agriculturenigeria.com](http://www.agriculturenigeria.com)). High rainfall experienced in the tropical forest zone of the South supports plantation crops such as cocoa, oil palm, rubber, coffee and staple crops including yam, cassava, cocoyam and sweet potato. The lower rainfall in the North is better suited for grazing livestock including cattle, goats, sheep and camels and cultivation of crops such as sorghum, cotton and cowpea. Differences in climate, soil, vegetation, water resources and topographies also contribute to economic diversities. For instance, it is prevalent to initiate industries in the Southern part of the country due to the available natural resources and economic opportunities. This attracts investments in this region as well as immigration of the populace for better employment and improved livelihoods.

### 2.2. Staple foods and production in different regions of Nigeria

Nigeria is the largest producer of staple crops in Economic Community of West African States (ECOWAS) (Otegunrin and Sawickra 2019). Grains provide 46% of the calories and 52% of the protein consumed, while root and tuber crops provide 20% calories and 8% of protein (Chandrasekara and Kumar, 2016). Production and consumption of these staples vary across the different agro-ecological regions. The major staple foods produced in the Southern regions of Nigeria include cassava, yam, sweet potato, cowpea, maize and rice, while wheat, cowpea, sorghum, rice and millet are dominant for the Northern regions (Harris and Mohammed, 2003). In Southern Nigeria, staples are consumed along with vegetables and seafood; while in Northern Nigeria, staples are consumed with meat and dairy products.

Production of root and tuber crops including cassava, yam, sweet potato and solanum potato dominate the Nigerian agricultural system. At 240 million tons annually cultivated on 23 million hectares of land, production of root and tubers exceeds that for cereal crops (Sanginga and Mbabu, 2015). Such significant investment in root and tuber production can be attributed to its potential for addressing food and nutrition security. Cassava and yam, especially, play a vital role in ensuring food availability due to their adaptability to diverse agro-ecological environments, and their flexible harvesting period that provides on-farm food security year round (Sanginga and Mbabu, 2015).

The basic nutritional component of staple foods produced and consumed in different agro-ecological zones of Nigeria, as per the United States Department of Agriculture National Nutrient Database, is shown in Table 2 ([www.ars.usda.gov](http://www.ars.usda.gov)). Cassava, yam and sweet potato are excellent sources of carbohydrates but low in micronutrient and protein content. Dependence on these major staple foods for essential micronutrients is therefore a challenge (Table 2). Based on consumption patterns in West Africa, iron and zinc present in cassava provide only 5–8% and 13–14% of the Estimated Average Requirement (EAR) for iron and zinc, respectively, for children 2–5 years old (Stephenson et al., 2010). These analyses indicate that, the major staple foods across agro-ecological regions of Nigeria are not adequate to combat the hidden hunger resulting from insufficient micronutrients in the diet. Regular consumption of vegetables, fruits, dairy products and meats along with the staples is ideal, but they are not always accessible by low-income consumers (Ogundari and Arifalo, 2013). Dependence on meat and meat products as an alternative source for essential micronutrients is limited as production is insufficient to meet demands of the

**Table 1**  
Staple food crops prevalent in the geopolitical zones of Nigeria.

Geopolitical zones	Member states	Agro-ecological zones	Staples consumed
<b>North-West</b>	Zamfara, Sokoto, Kaduna, Kebbi, Katsina, Kano, Jigawa	Sahel, Sudan, Northern Guinea	Sorghum, millet, beans, legumes
<b>North-East</b>	Bauchi, Borno, Taraba, Adamawa, Gombe, Yobe	Sahel, Sudan, Northern Guinea, Southern Guinea	
<b>North-Central</b>	Niger, Kogi, Benue, Plateau, Nassarawa, Kwara, FCT	Northern Guinea, Southern Guinea	Yam, cassava, maize, rice, sorghum, millet, beans, legumes
<b>South-West</b>	Oyo, Ekiti, Osun, Ondo, Lagos, Ogun	Southern Guinea, Forest, Coastal	Cassava, yams, taro, sweetpotato, plantain, bananas, rice, legumes, maize
<b>South-South</b>	Bayelsa, Akwa Ibom, Edo, Rivers, Cross River, Delta	Southern Guinea, Forest, Coastal	
<b>South-East</b>	Enugu, Imo, Ebonyi, Abia, Anambra	Southern Guinea, Forest, Coastal	

Sources: [www.legit.ng/1094595-geopolitical-zones-nigeria-states.html](http://www.legit.ng/1094595-geopolitical-zones-nigeria-states.html); <http://www.inter-reseaux.org/publications/revue-grain-de-sel/51-special-issue-nigeria/article/staple-crop-production>.

rapidly growing population in Nigeria. In addition, poor transportation infrastructure and inadequate preservation of meat and meat products limits its availability (Adetunji and Rauf, 2012). Disruptions to production and distribution of nutritious foods of animal origin, fresh fruits and vegetables as a result of the COVID-19 global pandemic also poses a threat to nutrition security with respect to providing the lacking micronutrients in the commonly consumed staples.

### 2.3. Mineral micronutrient deficiency in Nigeria

Various studies performed in Nigeria show that micronutrient deficiency, particularly iron and zinc, is a major public health concern that requires serious attention (Abubakar et al., 2017; Harika et al., 2017). Prevalence of iron deficiency in Nigeria ranges from 19% to 80% for preschool children under 5 of age, and 20%–60% for pregnant women, while zinc deficiency ranges from 21% to 83% in preschool children (Ibeawuchi et al., 2017). Parametric analysis of malnutrition using anthropometric indices of percent stunted, underweight and wasting, show that children living in the North-West and North-East suffer greater malnutrition (50% and 40% stunted, respectively) compared to those living in the North-Central (25%), South-South/South-West (20%) and South-East (10%) (Ibeanu et al., 2012). Results of these studies indicate that inadequate intake and bioavailability of iron and zinc in staple foods are predisposing factors for iron and zinc deficiency. The main sources of these micronutrients are meat, seafood, nuts, seeds, legumes and dairy for zinc (<https://www.healthline.com/nutrition/best-foods-high-in-zinc#section1>), and whole grain cereals, liver, fish, green leafy vegetables and pulses in addition to meat and nuts for iron (Oluwatoyin and Adebukola, 2018). In some plant-based sources anti-nutritional factors such as phytic acid act to inhibit adequate absorption of these micronutrients. Studies carried out by Ibeanu et al. (2012) on nursery school children using data derived from questionnaires, anthropometry, biochemical examinations and a 3 days weighed food intake showed that children living in the North-West and North-East suffer greater malnutrition (50% and 40% stunted, respectively) compared to those living in the North-Central (25%), South-South/South-West (20%) and South-East (10%).

The prevalence of micronutrient deficiencies across the country can also be associated with the socio-economic status of families. This is determined by income, sanitary practices, and the educational levels of mothers, which affects breast feeding, timing and consumption of complementary foods (Onyemaobi and Onimawo, 2011). A study investigating prevalence of zinc deficiency in Imo state, South-East Nigeria, showed that 48% of the preschool children diagnosed with zinc deficiency were from the rural parts of the state, and comprised mostly of adults with limited formal education and low income status (Onyemaobi and Onimawo, 2011). Likewise, serum levels of zinc in preschool children in North-Central Nigeria were shown to be directly affected by the socio-economic status of their families (Abah et al., 2015). These findings indicated that strategies focused on biofortification of staple foods could be effective for reducing micronutrient

malnutrition in the country, since most rural dwellers are farmers and as such grow the staple foods they eat.

Nevertheless, other predisposing factors to iron and zinc malnutrition in Nigeria include protein-energy malnutrition (Abubakar et al., 2017), parasitic infections and competition among different minerals in the diet (Sandstorm, 2001; Ugwuja et al., 2011). Studies by Abubakar et al. (2017) reported that zinc deficiency is more prevalent among children with protein-energy malnutrition (PEM) compared to healthy children, thus suggesting that food diversification and inclusion of nutrients from animal sources are essential towards balanced nutrition for preschool children (Ibeanu et al., 2012). Protection from parasitic infections, particularly the malaria parasite is necessary for alleviating the impact of zinc deficiency in children under 5 of age, as 86% of cases observed in South-East Nigeria were precipitated by malaria (Onyemaobi and Onimawo, 2011). Studies have also shown that micronutrient bioavailability can be affected by competition among different minerals. Supplementation of iron in pregnant women diagnosed with iron deficiency anemia affects the bioavailability of zinc and copper in the bowel, while bioavailability of copper and iron in their blood was affected by zinc supplementation (Ugwuja et al., 2011). This study also showed that iron deficiency leads to increased copper levels in the liver while severe copper deficiency causes changes in iron metabolism leading to anemia and accumulation of iron in that organ (Ugwuja et al., 2011). This result illustrates concerns for the use of micronutrient supplementation as a mitigation strategy for micronutrient deficiency in pregnant women, since supplementation for one micronutrient could compete with bioavailability of another nutrient (Ugwuja et al., 2011). There is no evidence, however, that this is occurs with dietary sources of micronutrient obtained through regularly consumed biofortified and fortified foods.

Supplementation has been used extensively to save lives and alleviate suffering, and remains a viable option for people at high risk (Thompson and Amoroso, 2011). However, it is not effective in addressing the root cause of micronutrient malnutrition. Importantly, the benefits are transient and do not address lasting solutions to the underlying problems. As an alternative measure, food-based approaches involving fortification, dietary diversification/modification, nutrition education and biofortification are being emphasized (Thompson and Amoroso, 2011). The fundamental concept in a food-based approach is to ensure availability, increased access and consumption of a variety of micronutrient-rich foods as a means of improving consumers' micronutrient status. To date, strategies employed in implementing food-based approaches aim to enhance the energy and nutrient density of cereal-based food forms, incorporate enhancers of micronutrient absorption and reduce the content of anti-nutrients such as phytate within cereals and legumes through soaking, germination, and fermentation.

**Table 2**  
Nutritional content of basic staple foods in Nigeria.

Components	Value per 100 g fresh weight							
	Cassava (Raw)	Yam (Raw)	Sweetpotato (Raw)	Maize (Raw)	Rice (Cooked)	Sorghum (Grain)	Wheat (Soft white)	Cowpea (Leafy tips, raw)
Water (g)	59.68	69.8	77.28	75	70.27	12.4	10.42	89.78
Energy (Kcal)	160	118	86	88	123	329	340	29
Protein (g)	1.36	1.53	1.57	3.02	2.74	10.62	10.69	4.1
Total lipid (fat) (g)	0.28	0.17	0.05	0.78	0.97	3.46	1.99	0.25
Carbohydrates by difference (g)	38.06	27.88	20.12	20.71	25.58	72.09	75.36	4.82
Fiber, total dietary (g)	1.8	4.1	3	2.1	1.6	6.7	12.7	0
Iron (mg)	0.27	0.54	0.61	0.42	0.56	3.36	5.37	1.92
Zinc (mg)	0.34	0.24	0.3	0.38	0.71	1.67	3.46	0.29
Vitamin A (RAE, µg)	1	7	709	10	0	0	0	36
Vitamin C (total ascorbic acid-mg)	20.6	17.1	2.4	6.4	0	0	0	36
Vitamin B12	0	0	0	0	0	0	0	0
Folate (DFE-µg)	27	23	11	36	9	20	41	101

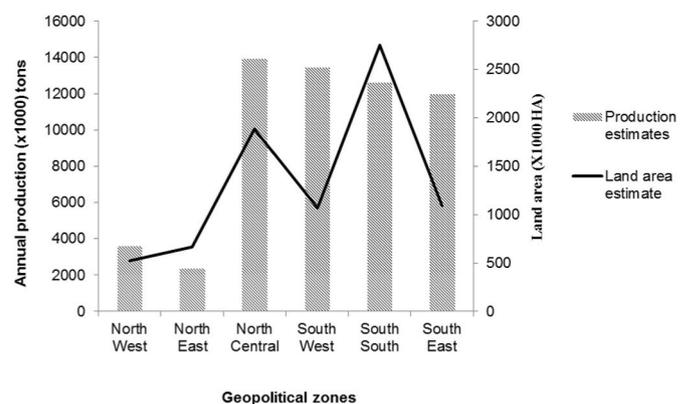
Abbreviations: RAE – Retinol Activity Equivalents; DFE - dietary foliate equivalents. Sources: (USDA, 2016)

### 3. Cassava: an important staple crop in Nigeria

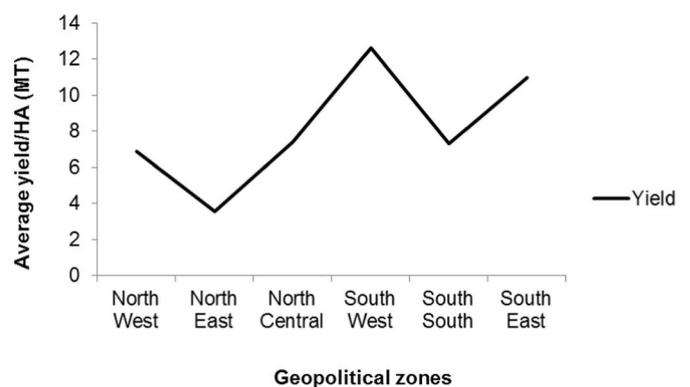
#### 3.1. Cassava production, consumption and utilization

Cassava is the most produced and second most consumed staple food in Nigeria, thereby making the country the leading producer globally with the production of 57.9 million metric tons (MT) (FAO, 2019). Due to its adaptive nature, cassava can be grown in all agro-ecological zones of the country but thrives best in the rainforest and derived savannah areas. Although two thirds of the 36 states are involved in cassava cultivation, production is most dominant in the north-central and southern parts of the country. Estimates of total land area devoted to cassava cultivation and tonnage of cassava production are shown in Fig. 1, with yield per hectare across the different zones shown in Fig. 2. While the North-Central and South-South regions have larger areas devoted to cassava cultivation, the South-West and South-East regions generate higher yield per hectare.

Cassava production in Nigeria remains significantly below its potential, illustrated by the fact that average yield/hectare is only 7.7 MT compared to 22–23 MT in Indonesia and Thailand (FAO, 2014). This disparity can be attributed to lack of quality cassava planting material, minimal mechanization and constraints from biotic stresses such as Cassava mosaic disease (CMD), that limit production by Nigerian farmers. Until recently, cassava cultivation in Nigeria was largely limited to subsistence farming, with women being the major stakeholders in production, processing and marketing. These low-income farmers resort to manual weeding and harvesting which is labour intensive, time consuming, tedious, and expensive if labourers are engaged. Greater than 90% of smallholder farmers use stem cuttings from previous cropping cycle to establish new fields, with no assurance of the disease status or quality of the planting material. Subsequent yields are, therefore, often compromised. Mitigating strategies towards ensuring the certification, availability and accessibility of high-quality cassava seed in Nigeria have been employed by the BASICS (Building an Economically Sustainable Seed Systems) project which aims at empowering cassava farmers economically (<http://www.rtb.cgiar.org/basics/>). The HarvestPlus Nigeria program is another project contributing to production and dissemination of quality planting cassava material (<https://nutritionconnect.org>). Across Nigeria, cassava storage roots are mostly processed and consumed as traditional meals in the form of *gari*, *fufu*, *abacha* and *lafun* (Fig. 3) (Akinpelu et al., 2011). Eighty percent of the cassava produced in Nigeria is processed into these traditional foods, with the remaining going to industrial products such as ethanol, starch, syrup, chips and high quality cassava flour (HQCF). Available data on cassava consumption patterns are shown in Fig. 4 with the highest cassava consuming states being Abia, Akwa Ibom, Anambra, Benue, Cross River, Ebonyi, Enugu, Imo, Ondo and Rivers. In urban



**Fig. 1.** Estimates of land area and tonnage of cassava production in the six agro-ecological zones of Nigeria. Source: NAERLS 2013 annual report



**Fig. 2.** Average cassava yield per hectare across the six agro-ecological zones of Nigeria. Source: NAERLS 2013 annual report

areas consumption of cassava foodstuffs in low-income households is double that of high-income households, while in rural areas consumption of cassava in low-income households is triple that of high-income households (Akinpelu et al., 2011).

#### 3.2. Overlap between cassava consumption and Fe/Zn deficiencies

A study conducted by Gegios et al. (2010) showed that 89% and 31% of Kenyan and Nigerian children respectively consuming cassava as a

staple food derived 21% of their energy from cassava. This proportion of dietary energy obtained from cassava was found to be inversely correlated with iron ( $r = -0.31$ ), zinc ( $r = -0.11$ ) and vitamin A ( $r = -0.15$ ) intake. It can be inferred from this study, that consumption of cassava might be related to risk factors for insufficient iron, zinc and vitamin A intake (Gegios et al., 2010) against data on household cassava consumption (<http://dataportal.opendataforafrica.org/>) (Fig. 5) shows that the number of households affected by micronutrient deficiency was highest for South-South and South-East regions, and that these areas also have highest levels of cassava consumption (Ibeanu et al., 2012). Strategic intervention to elevate iron and zinc levels in cassava has potential, therefore, to mitigate hidden hunger in populations depending on this staple.

#### 4. Iron and zinc biofortification

##### 4.1. Conventional breeding efforts in Fe/Zn biofortification

Biofortification is a process of improving the micronutrient content of staple crops through conventional plant breeding, genetic modification or agronomic practices. Biofortification of staple food crops brings advantages due to long-term cost-effectiveness and its potential to reach and deliver sustained beneficial impact within urban and rural populations. Investment in developing and delivering biofortified crops provides poor farmers with nutrient rich crops at low cost which can be further adapted to cultivars suited for different geographical locations.

Numerous international organizations, national programs and private seed companies are addressing the challenge of hidden hunger in Nigeria through the development of biofortified crops. HarvestPlus is a leading program, integrating the efforts of international partners and national programs to improve the micronutrient content of major staple crops (<https://www.harvestplus.org/evidence-document>). Seven staples including beans, cassava, maize, sweet potato, pearl millet, rice and wheat are being targeted. These were selected using the biofortification priority index (BPI) tool to determine the preferred staple(s) relevant for

delivering maximum benefit within a given country (Asare-Marfo, 2014). This was accomplished by a) exploring the available genetic diversity for minerals that can be used within breeding programs, b) breeding to develop germplasm combining high levels of one or more micronutrients, c) understanding genotype x environment (GxE) by studying micronutrient expression in different growing environments within target countries and d) release of nutritionally enhanced planting materials after multi-location testing over multiple seasons and with approval of national government agencies (Bouis and Saltzman, 2017). Progress made by HarvestPlus include increases in hemoglobin and total body iron in iron-depleted women in Rwanda after 4.5 months consumption of iron biofortified beans (Haas et al., 2016). Similarly, 4 months consumption of iron enhanced pearl millet by secondary school children in Maharashtra, India resulted in a significant improvement in serum ferritin and total body iron (Finkelstein et al., 2015). Recently, iron biofortification of pearl millet led to improvements in functional cognitive development that could highly impact women and teenagers' daily lives in India (Scott et al., 2018).

HarvestPlus targeted Vitamin A biofortification of cassava and maize as a priority for Nigeria (Asare-Marfo, 2014). Genetic diversity present within the germplasm have been used to generate biofortified cassava and maize using conventional breeding approaches. In a large, diverse country such as Nigeria significant differences exist across agro-ecological and climatic conditions and in social economic, educational and cultural practices. There is a need, therefore, to determine regional requirements when considering best strategies to mitigate micronutrient deficiencies within the country.

Presently, iron and zinc biofortification is being addressed by collaborative efforts of the Nigeria Sorghum Value Chain, Agricultural Transformation Agenda Support Program (ATASP-1), and Harvest Plus. Two new, nutritional-high sorghum varieties, improved Deko (12KNICSV-188) and improved Zabuwa (12KNICSV-22) rich in iron and zinc have been released. The improved Deko has three times higher iron content, at 126 ppm compared to 40 ppm dry weight derived from local varieties (ICRISAT, 2016) while Zabuwa had an iron gain of 54 ppm



**Fig. 3.** Processing cassava storage roots in Nigeria. a) Peeling of cassava roots. b) Milling of storage roots into cassava flour c) Roasting of cassava flour into gari. d) Stirring of cassava flour to make fufu.

Source: NextGen Gender responsive research @NRCRI, Umudike

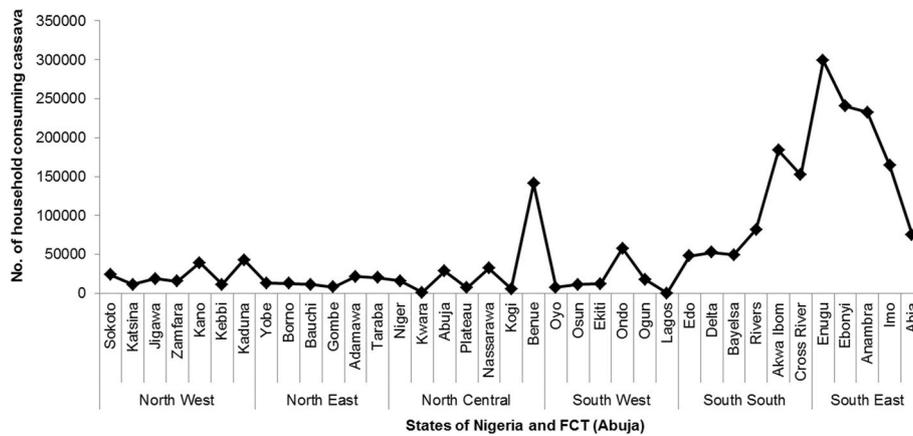


Fig. 4. Daily cassava consumption across thirty six states and the federal capital territory (Abuja) of Nigeria.

Source: <http://nigeria.opendataforafrica.org/>

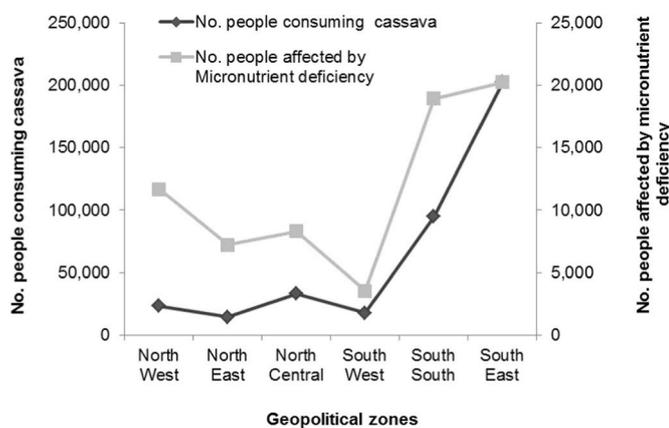


Fig. 5. Cassava consumption and micronutrient deficiencies across geopolitical zones of Nigeria.

Source: <http://nigeria.opendataforafrica.org/> Ibeanu et al., 2012

(<http://www.tnpp.org/2016/02/new-sorghum-varieties-released-in.html>). These results are encouraging but not adequate to handle the consequences of iron and zinc deficiencies in the country, as sorghum is a staple food for only a fraction in the northern part of the country. There is a strong need for iron and zinc biofortification in crops such as cassava that are widely consumed. There is insufficient genetic variation in iron and zinc accumulation within the cassava germplasm (6–10 µg/g DW Fe and 7–10 µg/g DW Zn). Therefore, conventional breeding is unlikely to achieve elevated mineral content in cassava storage roots to reach nutritionally significant levels (Chavez et al., 2000).

#### 4.2. Transgenic efforts in Fe/Zn biofortification

Transgenic approaches can be an effective method for achieving biofortification, especially when limited genetic variation for a given nutrient is available within the existing germplasm (Zhu et al., 2007). Transgenic methods can be employed to overexpress genes controlling mineral uptake and transport (Johnson et al., 2011), reduction in anti-nutrients (Kruger et al., 2012), and redistribution of nutrients across plant tissues (Ishimaru et al., 2010). Production of transgenic biofortified crops requires significant investment in early-stage development, but over time can be a cost-effective and viable approach (Garg et al., 2018). Some cereal crops, notably rice, have been genetically modified to enhance their micronutrient content. Overexpression of rice *Nicotianamine synthase* (*OsNAS2*) and soybean ferritin (*SferH-1*) resulted in elevated iron (15 µg g<sup>-1</sup>Fe) and zinc (45.7 µg g<sup>-1</sup>Zn) in polished

grain, to deliver 30% Estimated Average Requirements (EAR) for both minerals (Trijatmiko et al., 2016). Product development for high iron and zinc in adapted rice genotypes is now underway with expected release in 2022 in Bangladesh (Tohme and Beyer, 2014). Success in iron biofortification of rice was translated to wheat to achieve a 52 ppm iron compared to 30 ppm by expressing *OsNAS2* under the control of maize ubiquitin promoter (*Ubi1*) (Singh et al., 2017). Recent study shows increased mineral bioavailability in wheat products is possible by understanding the mechanisms that control the mineral deposition and transport within the developing wheat grains (Balk et al., 2019).

Two major international programs are engaged in transgenic approaches to achieve biofortification of Nigerian staple crops. The African Biofortified Sorghum (ABS) project (<https://www.biosorghum.org/>) aims to develop transgenic sorghum with increased levels of lysine, vitamin A, iron and zinc for subsequent introgression into high yielding farmer-preferred varieties. With the aid of its technology partner Cor-teva Agriscience, the ABS project has produced transgenic sorghum with improved protein profile [lysine (30–120%), tryptophan (10–20%), threonine (30–40%)], elevated levels of pro-vitamin A (5.7–21.0 µg/g beta-carotene) and reduced phytate (35–65%). Bioavailability studies also showed that increased iron (20–30%) and zinc (30–40%) absorption was achieved along with the reduced phytate levels. The Virus Resistant Cassava for Africa (VIRCA Plus) project (<https://www.danforthcenter.org/scientists-research/research-institutes/institute-for-international-crop-improvement/crop-improvement-projects/virca-plus>) brings together success from two previous projects, VIRCA (Taylor et al., 2012) and BioCassava Plus (Sayre et al., 2011). Its goal is to develop virus resistant, iron and zinc enhanced cassava varieties to address micronutrient deficiency and improve the livelihood of consumers in Nigeria.

With minimal genetic diversity for iron and zinc concentration (6–10 µg/g DW Fe and 7–10 µg/g DW Zn respectively) (Chavez et al., 2000) in known cassava germplasm, VIRCA Plus aims to reach nutritionally significant levels for consumers by elevating Fe and Zn in storage root tissues to 40–50 µg/g DW. Earlier efforts to improve the mineral concentration of cassava tuberous roots met with limited success. Fourteen gene constructs comprising seven different transgene combinations were introduced into cassava. Strategies found to be successful in cereals, such as upregulation of genes involved in long distance transport (*OsNAS2* and *OsYSL*), were not efficacious in cassava, and in many cases proved to be toxic and/or caused developmental problems in regenerated plants (Narayanan et al., 2019). Alternative strategies were pursued to enhance mineral source sink dynamics in cassava storage root tissues. Overexpression of *FEA1*, iron assimilatory protein from *Chlamydomonas reinhardtii* accumulated 3 times higher iron concentration in greenhouse-grown plants (Ihemere et al., 2012), but

was not functional under field conditions. Overexpression of zinc transporters *AtZIP1* and *AtMTP1* in cassava elevated zinc concentration in storage roots but greenhouse- and field-grown plants were compromised for shoot development (Gaitán-Solís et al., 2015). Overexpression of the *Arabidopsis thaliana* vacuolar iron transporter gene *VIT1* in cassava was more successful (Narayanan et al., 2015). When tested under field conditions, *AtVIT1* transgenic cassava plants accumulated 3–7 times higher iron concentration (60 µg/g vs 9 µg/g DW) in storage roots compared to non-transgenic controls, but as expected brought no increase in zinc accumulation (Kennedy and Meyers, 2005). Co-expressing the *A. thaliana* iron transporter *IRT1* and ferritin *FER1* resulted in elevation of both minerals to reach target levels in storage roots, achieving up to 18 times higher iron (130 µg/g vs 7 µg/g DW) and 10 times higher zinc concentrations (103 µg/g vs 10 µg/g DW) than non-transgenic controls under field conditions (Kennedy and Meyers, 2005).

The potential nutritional impact of cassava storage roots biofortified by overexpressing *VIT1* and *IRT1+FER1* was assessed by calculating their potential contribution to Estimated Average Requirement (EAR) (Kennedy and Meyers, 2005) for iron and zinc. EAR is a method to calculate the average daily nutrient intake estimated to meet requirements of half the healthy individuals of a specific gender and life stage group within a given population. EAR is determined by dividing the physiological requirement of the nutrient for a particular group by the fractional nutrient absorption. Non-modified cassava storage roots provide only 5–8% and 13–14% EAR for iron and zinc respectively for 1-6-year-old children and non-lactating, non-pregnant West African women (Taylor et al., 2016). Analysis of *IRT1+FER1* biofortified storage roots for iron and zinc retention and bioavailability after processing into common West African foodstuffs determined that the modified tissues could provide 40–50% EAR for iron and 60–70% EAR for zinc. This data indicates that consumption of *IRT1+FER1* biofortified cassava foodstuffs has the potential to beneficially impact the health of consumers in West Africa and elsewhere (Narayanan et al., 2019).

#### 4.3. Requirements for deploying iron and zinc enhanced cassava

Biofortification for iron and zinc in cassava offers a powerful strategy for combating the consequences of micronutrient deficiency in Nigeria and elsewhere in sub-Saharan Africa. To be effective in delivering these benefits, enhanced cassava planting materials must be developed and delivered to at-risk populations. Review of existing literature and the sub-national biofortification priority index identified the North-Central, South-West, South-East and South-South regions as the major cassava production and consumption areas (Herrington et al., 2018 (<https://ageconsearch.umn.edu/record/277092/?ln=en>)). Multiple cassava cultivars with varying agronomic and culinary traits are grown across the different regions and sub-regions within Nigeria. Effective biofortification intervention requires identification of cassava varieties relevant to food use in these regions. For example, enhancing iron and zinc content in a cultivar grown primarily for beer or alcohol production would fail to deliver the desired goals. A program for transgenic biofortification in a vegetatively propagated crop such as cassava requires a) selection of relevant farmer and breeder preferred cultivars for modification b) development of genetic transformation capacity in the target cultivars and production of transgenic cassava plants lines, c) molecular characterization of the transgenic lines and assessment of mineral accumulation and phenotype, d) a confined field testing program in target region(s) to identify lead lines e) multilocational confined field trials for collection of regulatory data and compilation of regulatory dossiers for evaluation by regulatory authorities, f) evaluation of selected events for bio-accessibility and availability of iron and zinc in cassava foods consumed within the target regions, 7) regulatory approval and variety registration before release of biofortified planting materials to farmers and breeders.

Success of these processes requires an enabling policy environment.

Recent approval for Bt-cowpea indicates important progress in obtaining approval for deployment of biotech crops to farmers in Nigeria (<https://geneticliteracyproject.org/2020/06/02/landmark-approval-of-gmo-bt-insect-resistant-cowpea-leads-nigeria-toward-sustainable-farming/>). Engagement with cassava farmers and farmer groups, processors and marketers acquainted with the limitations associated with cassava production and utilization, and with a basic understanding of the benefits of transgenic technology is essential in achieving product acceptance and adoption. National biosafety committees play an important role in regulatory decisions as well as collaborating with the government policies. Additionally, there is a need for capacity building for effective communication and program development on the importance of iron and zinc enhanced cassava as a means of ameliorating the effect of hidden hunger impacting countries such as Nigeria. There is also need to create awareness about transgenic crops within farmers and consumers. Recent studies of cultivating provitamin A GM cassava in Nigeria among smallholder farmers showed three distinct groups including low, medium and high opposition. It was estimated that only 25% of the surveyed population were highly opposing to transgenic technology (Oparinde et al., 2017). Knowledge sharing between the scientists, regulators and policy makers can help in developing guidelines for public. Golden rice a provitamin A biofortified rice is an appropriate example for transgenic biofortified crop which has completed food safety evaluation and regulatory approvals (Potrykus, 2010). Collaboration and knowledge transfer from these different biofortified crops will also help in achieving the product acceptance.

#### 5. Concluding remarks

This review highlights mineral deficiencies in the diet associated with geographical locations, the importance of the staple food crop cassava within populations in Nigeria. A recent report describing genetic modification of cassava storage roots for mineral accumulation indicates the potential to enhance cassava foodstuffs to provide 40–50% EAR for iron and 60–70% EAR for zinc within vulnerable populations. Consumption of biofortified cassava foodstuffs has potential to beneficially impact the health of consumers in West Africa. Iron and zinc biofortification in cassava would have a significant impact on not only on people's health but also on their economy. Apart from the underlying technology, factors such as regulatory policies and consumer acceptance is required through outreach and education if biofortification is to reach its maximum impact. For commercialization of biofortified crops, taste studies (palatability), increasing awareness to the farmers and public via consumer marketing, television, newspaper and workshops are necessary. Moving forward, biofortified cassava developed through VIRCA Plus and other development programs could collaborate with diverse partners including regional organizations, food processing companies, and local development banks. Collaborative efforts by public and private sectors can support biofortification and ultimately promote cognitive development of children towards building a stronger and self-sustaining nation "Nigeria".

#### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

#### Acknowledgements

This work was supported by the Bill & Melinda Gates Foundation (BMGF) through the Global 339 Challenges for Global Health Program (Grant no: OPPGD1484).

## References

- Abah, R.O., Okolo, S.N., John, C., Ochoga, M.O., 2015. Prevalence of zinc deficiency among school children in a rural setting in North-Central Nigeria. *Int. J. Public Heal. Res.*
- Abubakar, N., Atiku, M., Alhassan, A., Mohammed, I., Garba, R., Gwarzo, G., 2017. An assessment of micronutrient deficiency: a comparative study of children with protein-energy malnutrition and apparently healthy controls in Kano, Northern Nigeria. *Trop. J. Med. Res.* <https://doi.org/10.4103/1119-0388.198124>.
- Adetunji, M., Rauf, M.O., 2012. Analysis of household demand for meat, in Southwest, Nigeria analysis of household demand for meat, in Southwest, Nigeria analysis of household demand for meat, in Southwest, Nigeria. *Glob. J. Sci. Front. Res. Agric. Biol.*
- Akinpelu, A.O., Amangbo, L.E.F., Olojede, A.O., Oyekale, A.S., 2011. Health implications of Cassava production and consumption. *J. Agric. Soc. Res.* 11, 118–125.
- Asare-Marfo, D., 2014. Biofortification prioritization index (BPI) | IFPRI [WWW Document]. URL, accessed 4.22.19. <http://www.ifpri.org/publication/biofortificati-on-prioritization-index-bpi>.
- Bailey, R.L., West, K.P., Black, R.E., 2015. The epidemiology of global micronutrient deficiencies. *Ann. Nutr. Metab.* <https://doi.org/10.1159/000371618>.
- Balk, J., Connorton, J.M., Wan, Y., Lovegrove, A., Moore, K.L., Uauy, C., Sharp, P.A., Shewry, P.R., 2019. Improving wheat as a source of iron and zinc for global nutrition. *Nutr. Bull.* 44, 53–59. <https://doi.org/10.1111/mbu.12361>.
- Bouis, H.E., Saltzman, A., 2017. Improving nutrition through biofortification: a review of evidence from HarvestPlus, 2003 through 2016. *Glob. Food Sec.* <https://doi.org/10.1016/j.gfs.2017.01.009>.
- Chandrasekara, A., Josheph Kumar, T., 2016. Roots and tuber crops as functional foods: a review on phytochemical constituents and their potential health benefits. *Int. J. Food Sci.*
- Chavez, A.L., Bedoya, J.M., Sánchez, T., Iglesias, C., Ceballos, H., Roca, W., 2000. Iron, carotene, and ascorbic acid in cassava roots and leaves. *Food Nutr. Bull.* <https://doi.org/10.1177/156482650002100413>.
- FAO, IFAD, UNICEF, WFP and WHO, 2019. The State of Food Security and Nutrition in the World 2019. Safeguarding against Economic Slowdowns and Downturns. FAO, Rome.
- FAO, 2014. FAOSTAT. Food and Agriculture Organization of the United Nations, Rome, Italy.
- Finkelstein, J.L., Mehta, S., Udipi, S.A., Ghugre, P.S., Luna, S.V., Wenger, M.J., Murray-Kolb, L.E., Przybyszewski, E.M., Haas, J.D., 2015. A randomized trial of iron-biofortified pearl millet in school children in India. *J. Nutr.* <https://doi.org/10.3945/jn.114.208009>.
- Gaitán-Solís, E., Taylor, N.J., Siritunga, D., Stevens, W., Schachtman, D.P., 2015. Overexpression of the transporters AtZIP1 and AtMTP1 in cassava changes zinc accumulation and partitioning. *Front. Plant Sci.* <https://doi.org/10.3389/fpls.2015.00492>.
- Garg, M., Sharma, N., Sharma, S., Kapoor, P., Kumar, A., Chunduri, V., Arora, P., 2018. Biofortified crops generated by breeding, agronomy, and transgenic approaches are improving lives of millions of people around the world. *Front. Nutr.* <https://doi.org/10.3389/fnut.2018.00012>.
- Gegios, A., Amthor, R., Maziya-Dixon, B., Egesi, C., Mallowa, S., Nungo, R., Gichuki, S., Mbanaso, A., Manary, M.J., 2010. Children consuming cassava as a staple food are at risk for inadequate zinc, iron, and vitamin A intake. *Plant Foods Hum. Nutr.* <https://doi.org/10.1007/s11130-010-0157-5>.
- Haas, J.D., Luna, S.V., Lung'aho, M.G., Wenger, M.J., Murray-Kolb, L.E., Beebe, S., Gahutu, J.-B., Egli, I.M., 2016. Consuming iron biofortified beans increases iron status in Rwandan women after 128 Days in a randomized controlled feeding trial. *J. Nutr.* <https://doi.org/10.3945/jn.115.224741>.
- Harika, R., Faber, M., Samuel, F., Kimiywe, J., Mulugeta, A., Eilander, A., 2017. Micronutrient Status and Dietary Intake of Iron, Vitamin A, Iodine, Folate and Zinc in Women of Reproductive Age and Pregnant Women in Ethiopia, Kenya, Nigeria and South Africa: A Systematic Review of Data from 2005 to 2015. <https://doi.org/10.3390/nu9101096>. *Nutrients*.
- Harris, F.M., Mohammed, S., 2003. Relying on nature: wild foods in northern Nigeria. *AMBIO A J. Hum. Environ.* 32 (1), 24–29.
- Herrington, C., Lividini, K., Angel, M.D., Biro, E., 2018. Prioritizing Countries for Biofortification Interventions: Biofortification Priority Index, second ed. BPI 2.0) Harvest Plus working paper.
- Ibeanu, V., Okeke, E., Onyechi, U., Ejiofor, Uju, 2012. Assessment of anthropometric indices, iron and zinc status of preschoolers in a peri-urban community in south east Nigeria. *Int. J. Basic Appl. Sci. (IJBAS-IJENS IJENS I J E N S)*.
- Ibeawuchi, A.N.E., Onyiriuka, A.N., Abiodun, P.O., 2017. High prevalence of zinc deficiency in rural Nigerian preschool children: a community-based cross-sectional study. *Rom. J. Diabetes, Nutr. Metab. Dis.* <https://doi.org/10.1515/rjndm-2017-0004>.
- ICRISAT, 2016. New Sorghum Varieties Released in Nigeria with Higher Iron, Yields and Drought Resilience – ICRISAT [WWW Document]. URL, accessed 4.22.19. <https://www.icrisat.org/new-sorghum-varieties-will-fight-malnutrition-and-climate-change-in-sudan-and-sahel-region-of-nigeria/>.
- IFPRI (International Food Policy Research Institute), 2016. Global Food Policy Report. <https://doi.org/10.2499/9780896295827>. Washington, D.C.
- Ihemere Narayanan, N., Sayre Richard, T., Narayana, U.E., 2012. Iron biofortification and homeostasis in transgenic cassava roots expressing the algal iron assimilatory gene, FE1A. [Erratum appears in *Front Plant Sci.* 2013;4:492]. *Front. Plant Sci.*
- Ishimaru, Y., Masuda, H., Bashir, K., Inoue, H., Tsukamoto, T., Takahashi, M., Nakanishi, H., Aoki, N., Hirose, T., Ohsugi, R., Nishizawa, N.K., 2010. Rice metal-nicotianamine transporter, OsYSL2, is required for the long-distance transport of iron and manganese. *Plant J.* <https://doi.org/10.1111/j.1365-313X.2010.04158.x>.
- Johnson, A.A.T., Kyriacou, B., Callahan, D.L., Carruthers, L., Stangoulis, J., Lombi, E., Tester, M., 2011. Constitutive overexpression of the OsNAS gene family reveals single-gene strategies for effective iron- and zinc-biofortification of rice endosperm. *PLoS One.* <https://doi.org/10.1371/journal.pone.0024476>.
- Kennedy, E., Meyers, L., 2005. Dietary Reference Intakes: development and uses for assessment of micronutrient status of women - a global perspective. *American Journal of Clinical Nutrition.* <https://doi.org/10.1093/ajcn/81.5.1194>.
- Kruger, J., Taylor, J.R.N., Oelofse, A., 2012. Effects of reducing phytate content in sorghum through genetic modification and fermentation on in vitro iron availability in whole grain porridges. *Food Chem.* <https://doi.org/10.1016/j.foodchem.2011.08.063>.
- Merson, M.H., Black, R., Mills, A., 2012. Global Health: Diseases, Programs, Systems, and Policies, Health Policy and Planning.
- Narayanan, N., Beyene, G., Chauhan, R.D., Gaitán-Solís, E., Gehan, J., Butts, P., Siritunga, D., Okwuonu, I., Woll, A., Jiménez-Aguilar, D.M., Boy, E., Grusak, M.A., Anderson, P., Taylor, N.J., 2019. Biofortification of field-grown cassava by engineering expression of an iron transporter and ferritin. *Nat. Biotechnol.* 37, 144–151. <https://doi.org/10.1038/s41587-018-0002-1>.
- Narayanan, N., Beyene, G., Chauhan, R.D., Gaitán-Solís, E., Grusak, M.A., Taylor, N., Anderson, P., 2015. Overexpression of Arabidopsis VIT1 increases accumulation of iron in cassava roots and stems. *Plant Sci.* <https://doi.org/10.1016/j.plantsci.2015.09.007>.
- National Agricultural Extension and Research Liaison Services (Nigeria) (NAERLS), n.d. Agricultural Performance Survey of 2013 Wet Season in Nigeria : (national report).
- Ogundari, K., Arifalo, S.F., 2013. Determinants of household demand for fresh fruit and vegetable in Nigeria: a double hurdle approach. *Q. J. Int. Agric.*
- Oluwatoyin B O, Adebukola A A. Dietary iron intake in Nigeria. *Nov Tech Nutri Food Sci.* 1(4).
- Onyemaobi, G.A., Onimawo, I.A., 2011. Zinc status of under-five children in rural and urban Imo state, Nigeria. *J. Basic. Appl. Sci. Res.*
- Oparinde, A., Abdoulaye, T., Mignouna, D.B., Bamire, A.S., 2017. Will farmers intend to cultivate Provitamin A genetically modified (GM) cassava in Nigeria? Evidence from a k-means segmentation analysis of beliefs and attitudes. *PLoS One* 12 (7), e0179427.
- Otekinrin, Oluotosin, Sawicka, Barbara, 2019. Cassava, a 21st century staple crop: how can Nigeria harness its enormous trade potentials? *Acta Agric. Slov.* 3, 194–202. <https://doi.org/10.31080/ASAG.2019.03.0586>.
- Potrykus, I., 2010. Lessons from the “Humanitarian Golden Rice” project: regulation prevents development of public good genetically engineered crop products. *Nat. Biotechnol.* <https://doi.org/10.1016/j.nbt.2010.07.012>.
- Prasad, A.S., 2013. Discovery of human zinc deficiency: its impact on human health and disease. *Adv. Nutr.: Int. Rev. J.* <https://doi.org/10.3945/an.112.003210>.
- Read, Scott, Obeid, Stephanie, Ahlenstiel, Chantelle, Ahlenstiel, Golo, 2019. The role of zinc in antiviral immunity. *Adv. Nutr.* 10, 696–710. <https://doi.org/10.1093/advances/nmz013>. Bethesda, Md.).
- Reed, H.E., Mberu, B.U., 2013. Capitalizing on Nigeria's demographic dividend: reaping the benefits and diminishing the burdens. *Etude la Popul. Africaine.* <https://doi.org/10.11564/27-2-477>.
- Roohani, N., Hurrell, R., Kelishadi, R., Schulin, R., 2013. Zinc and its importance for human health: an integrative review. *J. Res. Med. Sci.* 18 (2), 144–157.
- Sandstrom, B., 2001. Micronutrient interactions: effects on absorption and bioavailability. *Br. J. Nutr.* 85 (2), S181–185.
- Sanginga, N., Mbabu, A., 2015. Root and Tuber Crops (Cassava, Yam, Potato and Sweet Potato), Background Paper - Feeding Africa Conference. October 2015.
- Sayre, R., Beeching, J.R., Cahoon, E.B., Egesi, C., Fauquet, C., Fellman, J., Fregene, M., Gruissem, W., Mallowa, S., Manary, M., Maziya-Dixon, B., Mbanaso, A., Schachtman, D.P., Siritunga, D., Taylor, N., Vanderschuren, H., Zhang, P., 2011. The BioCassava plus program: biofortification of cassava for sub-Saharan Africa. *Annu. Rev. Plant Biol.* 62, 251–272. <https://doi.org/10.1146/annurev-arplant-042110-103751>.
- Scott, S.P., Murray-Kolb, L.E., Wenger, M.J., Udipi, S.A., Ghugre, P.S., Boy, E., Haas, J.D., 2018. Cognitive performance in Indian school-going adolescents is positively affected by consumption of iron-biofortified pearl millet: a 6-month randomized controlled efficacy trial. *J. Nutr.* <https://doi.org/10.1093/jn/nxy113>.
- Singh, S.P., Keller, B., Gruissem, W., Bhullar, N.K., 2017. Rice NICOTIANAMINE SYNTHASE 2 expression improves dietary iron and zinc levels in wheat. *Theor. Appl. Genet.* <https://doi.org/10.1007/s00122-016-2808-x>.
- Stephenson, K., et al., 2010. Consuming cassava as a staple food places children 2-5 years old at risk for inadequate protein intake, an observational study in Kenya and Nigeria. *Nutr. J.* 9, 9.
- Taylor, N.J., Halsey, M., Gaitán-Solís, E., Anderson, P., Gichuki, S., Miano, D., Bua, A., Alicai, T., Fauquet, C.M., 2012. The VIRCA project. *GM Crops Food* 3, 93–103. <https://doi.org/10.4161/gmcr.19144>.
- Taylor, N.J., Narayanan, N., Beyene, G., Chauhan, R.D., Gaitán-Solís, E., Siritunga, D., Grusak, M.A., Anderson, P., 2016. Iron and zinc biofortification of cassava storage roots to nutritionally significant levels. *Vitro Cell Dev. Biol.* 52, S38–S38.
- Thompson, B., Amoroso, L., 2011. Combating Micronutrient Deficiencies, Food-Based Approaches. <https://doi.org/10.1079/9781845937140.0000>.
- Tohme, J., Beyer, P., 2014. Transgenic Biofortified Crops.
- Trijatmiko, K.R., Duenäs, C., Tsakirpaloglou, N., Torrizo, L., Arines, F.M., Adeva, C., Balindong, J., Oliva, N., Sapasap, M.V., Borrero, J., Rey, J., Francisco, P., Nelson, A., Nakanishi, H., Lombi, E., Tako, E., Glahn, R.P., Stangoulis, J., Chadha-Mohanty, P., Johnson, A.A.T., Tohme, J., Barry, G., Slamet-Loedin, I.H., 2016. Biofortified indica rice attains iron and zinc nutrition dietary targets in the field. *Sci. Rep.* <https://doi.org/10.1038/srep19792>.

- Ugwuaja, E.I., Ejikeme, B.N., Ugwu, N.C., Obidoa, O., 2011. A Comparative study of plasma trace elements (copper, iron and zinc) status in anaemic and non-anaemic pregnant women in Abakaliki, Nigeria. *Online J. Health Allied Sci.*
- UNICEF, 2016. Nigeria - the Children - Maternal and Child Health.
- Wessels, I., Maywald, M., Rink, L., 2017. Zinc as a gatekeeper of immune function. *Nutrients* 9 (12), 1286. <https://doi.org/10.3390/nu9121286>.
- WHO, 2014. Global Nutrition Targets 2025 Policy Brief Series. WHO Int. <https://doi.org/10.2165/00024677-200302060-00002>.
- WHO - World Health Organization, 2017. World Health Statistics 2017 : Monitoring Health for the SDGs. World Health Organization. <https://doi.org/10.1017/CBO9781107415324.004>.
- Zhu, C., Naqvi, S., Gomez-Galera, S., Pelacho, A.M., Capell, T., Christou, P., 2007. Transgenic strategies for the nutritional enhancement of plants. *Trends Plant Sci.* <https://doi.org/10.1016/j.tplants.2007.09.007>.