



Article
scientifique

Revue de la
littérature

2023

Accepted
version

Public
access

This is an author manuscript post-peer-reviewing (accepted version) of the original publication. The layout of the published version may differ .

The role of orphan crops in the transition to nutritional quality-oriented crop improvement

Verbeecke, Vincent; Custódio, Laura; Strobbe, Simon; Van Der Straeten, Dominique

How to cite

VERBEECKE, Vincent et al. The role of orphan crops in the transition to nutritional quality-oriented crop improvement. In: *Biotechnology advances*, 2023, vol. 68, p. 2045–2322. doi: 10.1016/j.biotechadv.2023.108242

This publication URL: <https://archive-ouverte.unige.ch/unige:173258>

Publication DOI: [10.1016/j.biotechadv.2023.108242](https://doi.org/10.1016/j.biotechadv.2023.108242)

© This document is protected by copyright. Please refer to copyright holder(s) for terms of use.

Last deposit update in Archive ouverte UNIGE on 22.11.2023 14:11

Journal Pre-proof

The role of orphan crops in the transition to nutritional quality-oriented crop improvement

Vincent Verbeeke, Laura Custódio, Simon Strobbe, Dominique Van Der Straeten



PII: S0734-9750(23)00149-0

DOI: <https://doi.org/10.1016/j.biotechadv.2023.108242>

Reference: JBA 108242

To appear in: *Biotechnology Advances*

Received date: 25 June 2023

Revised date: 9 August 2023

Accepted date: 25 August 2023

Please cite this article as: V. Verbeeke, L. Custódio, S. Strobbe, et al., The role of orphan crops in the transition to nutritional quality-oriented crop improvement, *Biotechnology Advances* (2023), <https://doi.org/10.1016/j.biotechadv.2023.108242>

This is a PDF file of an article that has undergone enhancements after acceptance, such as the addition of a cover page and metadata, and formatting for readability, but it is not yet the definitive version of record. This version will undergo additional copyediting, typesetting and review before it is published in its final form, but we are providing this version to give early visibility of the article. Please note that, during the production process, errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

© 2023 Published by Elsevier Inc.

The role of orphan crops in the transition to nutritional quality-oriented crop improvement

Vincent Verbeecke^{1,2}, Laura Custódio^{1,2}, Simon Strobbe¹ and Dominique Van Der Straeten^{1,*}

1-Laboratory of Functional Plant Biology, Department of Biology, Ghent University, K.L. Ledeganckstraat 35, 9000 Ghent, Belgium.

2-These authors contributed to the work equally.

*Author for correspondence: Dominique.VanDerStraeten@UGent.be

Abstract

Micronutrient malnutrition is a persisting problem threatening global human health. Biofortification via metabolic engineering has been proposed as a cost-effective and short-term means to alleviate this burden. There has been a recent rise in the recognition of potential that underutilized, orphan crops can hold in decreasing malnutrition concerns. Here, we illustrate how orphan crops can serve as a medium to provide micronutrients to populations in need, whilst promoting and maintaining dietary diversity. We provide a roadmap, illustrating which aspects to be taken into consideration when evaluating orphan crops. Recent developments have shown successful biofortification via metabolic engineering in staple crops. This review provides guidance in the implementation of these successes to relevant orphan crop species, with a specific focus on the relevant micronutrients iron, zinc, provitamin A and folates.

Keywords: Micronutrient malnutrition, Biofortification, Orphan crops, Iron, Zinc, Provitamin A, Folates

Highlights

- Micronutrient malnutrition is a persisting global public health problem, particularly affecting poor rural populations.
- Recent developments in biofortification demonstrate that a shift towards nutritional quality improvement in highly consumed crops can alleviate this problem.
- Orphan crops can play a prominent role in feeding specific regional populations, typically burdened by several micronutrient deficiencies.
- Translation of validated biofortification approaches to a variety of orphan crops can be a tremendous asset in the fight against hidden hunger.
- Guidance is provided towards orphan crop biofortification, focusing on iron/zinc, provitamin A and folates.

The global problem of hidden hunger

Malnutrition is a problem that affects more than 2 billion people worldwide (Lowe 2021). The predominant cause for malnutrition is a lack of access to nutritious food (Muthayya 2013; Research Institute (IFPRI) 2014). Micronutrient deficiencies (MNDs) are characterized by an insufficient intake and/or absorption of vitamins and minerals. This often goes unnoticed at the onset, since it initially is manifested in mild to moderate symptoms, and only becomes visible when the deficiency status becomes severe. For this reason, this condition is regarded as “hidden hunger”. It is widespread, affecting almost one in every three humans (Muthayya 2013; Ruel-Bergeron 2015). The most affected groups are women at reproductive age and young children, yet MNDs can impair human health at any moment regardless of gender and/or age group (Muthayya 2013).

The overwhelming majority of people affected by MNDs come from lower-income households (Darnton-Hill and Mkparu 2015). People in such conditions most often cannot afford to consume a varied and nutrient-rich diet, such as animal-derived products or fruits and vegetables (Chaudhary, Gustafson, and Mathys 2018). Hence, these diets amount to the occurrence of multiple MNDs in a more frequent way than single MNDs (Darnton-Hill 2012). In other words, MNDs often do not come alone, making the issue even more problematic. Currently, among the most pressing micronutrient deficiencies within the general population are iron, zinc, and iodine, as well as vitamins A and B9 (folate) (Van Der Straeten 2020; Lowe 2021). Countries that have large shares of the population affected by this condition have a direct negative impact on their economic development given that MNDs can severely reduce cognitive function as well as physical performance (Darnton-Hill 2005; Steur 2012; 2015; Win 2016). The associated long-term health impacts are typically felt by the most vulnerable people in a society, particularly the rural poor and marginalized (Muthayya 2013; Research Institute (IFPRI) 2014).

At first glance, the problem of undernutrition seems a paradox, as we are living in a time of high agricultural productivity. Attaining adequate nutrition, however, is far more complicated than just having access to food to satisfy hunger. Hidden hunger is often the result of monotonous diets that rely heavily on starchy staple crops, such as maize (*Zea mays*), wheat (*Triticum aestivum*) and rice (*Oryza sativa*) (Burchi, Fanzo, and Frison 2011). These provide a high share of daily caloric needs (McKevith 2004), yet often fail to supply adequate amounts of micronutrients (Ruel-Bergeron 2015; S. Strobbe and Van Der Straeten 2018; Titcomb and Tanumihardjo 2019). It is well established that crop breeding has traditionally focused on maximizing yield-associated traits, at the cost of significant genetic diversity (Flint-García 2013; Gillespie and van den Bold 2017). This has resulted in neglect of crop nutritional quality and biodiversity, which negatively impacts human health. Indeed, across different regions and populations dietary diversity was found to positively correlate with nutrient adequacy scores (Nair, Augustine, and Konapur 2016). Additionally, MNDs can be the result of inadequate absorption of micronutrients, which can have a genetic basis or be the result of repetitive exposure to infection and/or inflammation (Bailey, West Jr., and Black 2015). Yet, most agriculture-based solutions haven't focused on nutritional quality gains, as it had been previously understood that malnutrition was simply a consequence of insufficient food production (Thompson, Cohen, and Meerman 2012; El Bilali 2019). It is now clear that tackling malnutrition will require a reform of the current global food system (Branca et al. 2020). This holistic and systematic change will have to be driven through advances in agricultural research and science-based policy making (Pinstrup-Andersen 2007). The current approaches to achieve food and nutrition security include both crop diversification and biofortification (McMullin 2021). These approaches are inherently complementary as they aim to increase the diversity of available foods, whilst also focusing on making them more nutritious (N. Kumar, Harris, and Rawat 2015; de Brauw et al. 2018). In this sense, the inclusion and promotion of

orphan crops for consumption, and as an object of biofortification efforts can be an effective path to combat malnutrition (Mabhaudhi 2019; Jamnadass 2020).

The necessity for scientific and agricultural development on orphan crops

Orphan crops (or minor crops) can be described as underutilized crop species that exhibit the following characteristics: they are not (or only limited) the object of research and development; they have marginal to low importance in global food production systems; they are often part of ancient cultural traditions; and they have emergent value due to their unrecognized traits (Gregory 2019; Tadele 2019). These minor crops are almost exclusively produced by small-scale farmers, and they represent a technological opportunity for innovation as they have diverse nutrition profiles. Furthermore, many have been utilized as medicinal plants, which points to their overall benefits for human health (Kamenya et al. 2021). Recently, many orphan crops have been marketed as healthy foods, or superfoods, as was the case for quinoa (*Chenopodium quinoa*). This increase in market interest might represent a shift in perception and the transition from marginal crop to a more widely consumed crop (Assogbadjo et al. 2021).

Orphan crops encompass e.g., cereal, root, fruit, and vegetable crops. These underutilized plant species hold an important role in the support of food security and agriculture in rural locations worldwide (Jamnadass 2020). Enset (*Ensete ventricosum*), for instance is crucial in preventing hunger in Ethiopia by its function as a subsistence crop (McCabe 1950), meaning that it serves as a food source for the farmers and thus the local community. As such, it stands to reason that they are an important part of any attempt to transform diets and agro-ecosystems. Targeting these orphan crops in future nutrition-focused policies seems favorable, especially because they are already positively viewed and enjoyed by local populations. Traditionally, these have been grown by marginal farmers mainly because they serve as subsistence crops. This means that the primary drive for their cultivation is directly related to survival of a small number of people (Bisht 2014), resulting in a lack of coordinated efforts to improve their productivity, reflected by their low yields. Most orphan crops have been cultivated in unfavorable conditions, which has promoted an enhanced overall resilience and tolerance for varied stress conditions due to evolutionary pressure (Mabhaudhi 2019; Kamenya et al. 2021). Additionally, in comparison to typical staples, these orphan crops were not the object of extensive breeding, being beneficial for the witnessed genetic diversity as well as micronutrient composition in most of them (Burchi, Fanzo, and Frison 2011).

There is a diverse array of underutilized crops that can have a clearly beneficial effect in tackling malnutrition. Yet, the genetic improvement of these crops has been lagging, despite their immense potential and cost-effectiveness (Steur 2010; 2012; 2015; Steur et al. 2017). Below, we illustrate the notion that extending current metabolic engineering approaches to orphan crops can serve to reduce MNDs. We focus on iron, zinc, provitamin A (carotenoids) and folates, as these are micronutrients for which deficiencies are widespread and have been successfully addressed in staple crops.

The role of genetic engineering for crop improvement

Biofortification, an intervention which involves increasing the natural micronutrient content of crops, has been presented as a cost-effective, sustainable method to help eradicate micronutrient deficiencies (Howarth E. Bouis and Saltzman 2017; Hay 2017; Garg 2018; Van Der Straeten 2020). Biofortified crops, obtained via breeding, particularly as part of the Harvest Plus initiative (CAST 2020), have already shown a beneficial impact on micronutrient intake and thus the health of the populations consuming them (Bouis and Saltzman 2017; Garg 2018; Birol and Bouis 2023). Next to breeding, biofortification of crops can be achieved via genetic engineering (GE). Indeed, many efforts have been

put into the development of biofortified plants through GE technology over the last decades, and this has mainly been focusing on highly consumed staple crops (Darnton-Hill 2012; Steur 2015; Garg 2018). However, the expected beneficial impact of GE biofortified crops on human health remains in the (very near) future, as the first GE biofortified crop, provitamin A-enriched Golden Rice (Ye and Beyer 1979; Paine 2005; Swamy 2019), was only approved for cultivation in the Philippines in July of 2021. In contrast to GE biofortified crops, biofortification by means of breeding has expanded into orphan crops as well. Examples of these include iron enriched pearl millet (*Pennisetum glaucum*) (K. N. Rai, Govindaraj, and Rao 2012; Finkelstein 2015) and orange-fleshed sweet potato (*Ipomoea batatas*) (Low and Thiele 2020). Biofortification via breeding has the advantage that it suffers less from obstruction by regulation, as compared to GE. GE has the advantage that it is not reliant on adequate genetic variations present in the specific crop and addition of micronutrients can not only go faster but also reach the necessary levels more easily than for regular breeding (Van Der Straeten 2020). If variation is present in the germplasm of a particular crop, meaning that certain sexually compatible accessions depict satisfactory micronutrient levels, breeding could be considered a favorable biofortification method (Van Der Straeten 2020). GE interventions, notwithstanding, hold the potential to tackle multiple (nutritional) target traits at once, thereby tremendously speeding up the process of crop nutritional enhancement (Van Der Straeten 2020). Using genetic engineering for crop improvement also provides the opportunity to incorporate other traits of agronomic interest (Van Der Straeten 2020), such as high-salinity or cold stress tolerance (Ribeiro et al. 2022). This exemplifies how the choice of breeding, GE or their combination is largely determined by the specific crop-micronutrient(s) combination in mind. This in turn is determined by the targeted population, burdened by one or multiple micronutrient deficiencies, as well as their dietary preferences.

Here, we illustrate the different parameters to be taken into account when considering the use of orphan crops for nutritional quality engineering, by scoring 20 orphan crops using radar graphs.

Case study – Tackling micronutrient deficiencies in Western Africa

To introduce a possible mode of action we have chosen Western Africa as an example region. This region has a high prevalence of micronutrient deficiencies (Figure 1) and a considerable diversity of locally consumed orphan crops such as finger millet (*Eleusine coracana*), cassava (*Manihot esculenta*, though often no longer considered an orphan crop), fonio (*Digitaria exilis*), and okra (*Abelmoschus esculentus*). A high percentage of the Western African population strongly suffers from iron, zinc, and/or vitamin A deficiency (Figure 1). Vitamin A deficiency even reaches up to 70% and 75% in Benin and Ghana, respectively. Taking neural tube defect (NTD) prevalence in children under 5 years as a proxy for vitamin B9 deficiency, no severe vitamin B9 insufficiencies seem present in most Western African countries apart from Nigeria (Figure 1). The limited data that exist on exact folate levels in women, however, indicate very high levels of folate deficiency in Sierra Leone (79%), Ghana (53,8%) and Côte d'Ivoire (86%) (Rogers et al. 2018). It is, therefore, reasonable to assume that deficiencies in all four micronutrients are very abundant in Western Africa, though the actual occurrence is subject to intra-national variability.

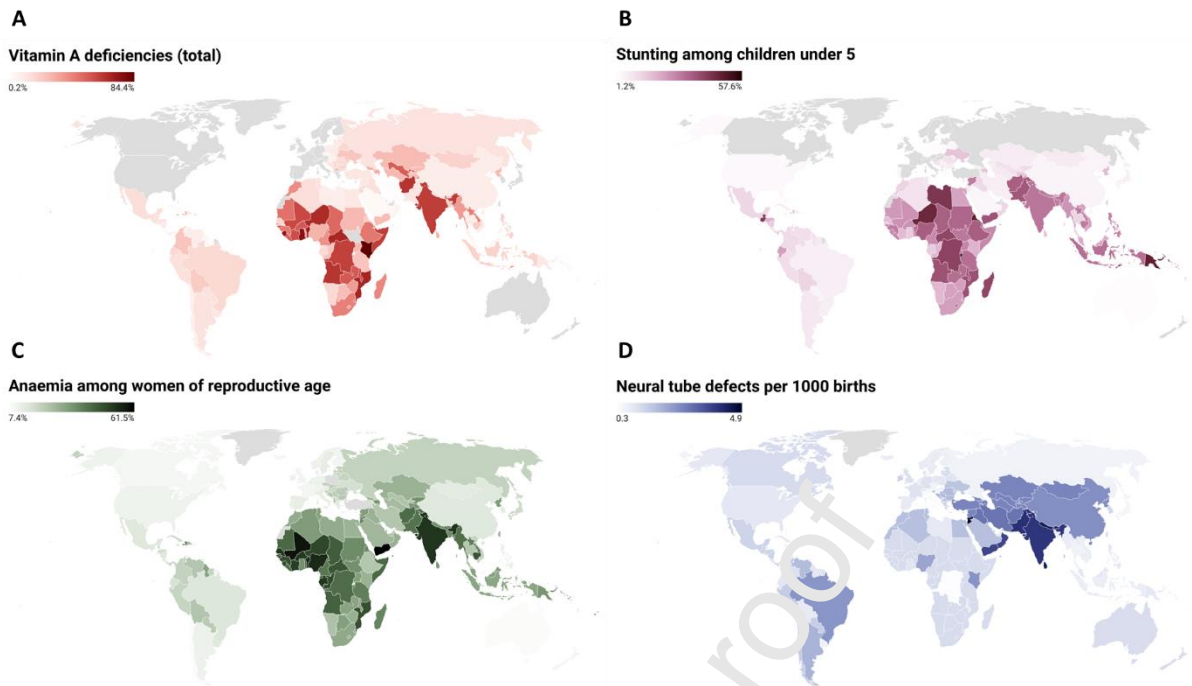


Figure 1: Geographical distribution of micronutrient deficiencies represented through related disease prevalence or micronutrient deficiency itself. A: Prevalence of vitamin A deficiency. Data from WHO, 2005; updated with data from Stevens et al., 2015 (Stevens 2015). B: Zinc deficiency prevalence visualized by stunting among children under five years old (FAO, 2020). Stunting is a common indicator for zinc deficiencies (Stammers 2015). C: Iron deficiency prevalence visualized by anaemia (FAO, 2020). Anaemia is one of the most common symptoms of iron deficiency and iron-deficiency Anaemia (IDA) represents nearly 50% of all anaemia cases (Stoltzfus 2003; Kassabaum 2016). D: Folate deficiency visualized by prevalence of neural tube defects (NTD's) per 1000 births. Folate deficiency is the primary cause of NTD's (The Modell Global Database of Congenital Disorders (Blencowe et al. 2018)). The maps presented here were created with Datawrapper online software.

A list of the most important orphan crops in Western Africa was composed and their nutritional profile was compared with that of important staple crops, including rice, potato, maize, wheat, and sweet potato (Table 1). Next, these crops were divided into four broad categories: fruits/vegetables, roots, cereals and legumes, and scored for nine distinct parameters, namely, crop robustness, yield, biotechnological potential, nutritional completeness, vitamin A content, vitamin B9 content, iron content, zinc content and health benefits. Within each parameter, different factors were utilized in order to score each crop: yield was assessed through crop productivity (in kg per hectare) and estimated post-harvest losses; crop robustness was represented through genetic diversity, breeding potential and stress (both biotic and abiotic) tolerance; biotechnological potential correlated with the availability of a transformation protocol and a genome sequence; nutritional completeness represented 50% of the daily recommended intake in terms of protein, fiber and fat; and, finally, health benefits accounted for the micronutrient density of the crop, the presence of anti-nutritional factors, gluten content, antioxidant content, and proven usage as medicinal plants. This thorough analysis allowed for the construction of the radar graphs presented in figure 2. At a glance, it immediately appears that low yield is a major drawback for all selected orphans, except cassava. Additionally, all orphans score remarkably high on health factors such as high antioxidant content and micronutrient content.

From each category one example crop is taken, for which we addressed which crop-specific interventions are needed to tackle hidden hunger. Broadly speaking, two approaches can be taken. On one hand, biofortification strategies can aim to create more complete crops by enhancing micronutrients that are specifically low in that particular crop. On the other hand, micronutrients that are already acceptably high could be increased to provide enough micronutrients for a person

independent of other components of the diet. Millets generally contain acceptable iron levels (Govindaraj et al. 2016; Krishnamurthy et al. 2016; Anitha et al. 2021; Hassan, Sebola, and Mabelebele 2021), but inhabitants of Niger, where millet consumption is high, still suffer from severe iron deficiency anemia, since it is not complemented by other iron rich dietary components (Figure 1). Iron biofortification of millet can thus be advisable. Another example is plantain, for which some cultivars exist that do contain sufficient pro-vitamin A. VAD in regions consuming plantain; however, remains high since these cultivars are not the preferred type for local farmers and consumers (Figure 1, Figure 3) (Norgrove and Hauser 2014). Here as well, breeding programs and GE to develop biofortified plantain could dampen the impact of VAD.

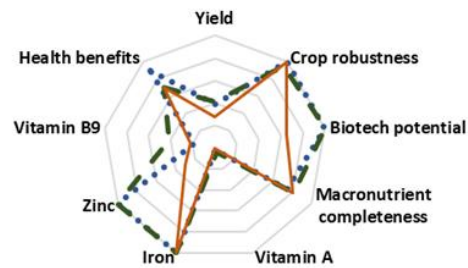
Within the following sections, we will comprehensively discuss specific metabolic engineering strategies for enhancement of micronutrients, as well as current breeding approaches, using plantain, cassava, millets, and cowpea as the chosen examples for the different categories of crops (Figure 4).

*Table 1: Daily dietary intake recommendations (DIR, range representative of age and gender specifications) and nutritional content (per 100g of fresh weight) of four staple crops - potato (*Solanum tuberosum*), rice (*Oryza sativa*), wheat (*Triticum aestivum*), maize (*Zea mays*), soybean (*Glycine max*) – and twenty-eight orphan crops – Cereals: finger millet (*Eleusine coracana*), pearl millet (*Cenchrus americanus* – *Penisetum glaucum*; nono, hyletic clade), kodo millet (*Paspalum scrobiculatum*), teff (*Eragrostis tef*), Fonio (*Digitaria exilis*), sorghum (*Sorghum bicolor*, *Sorghum ssp.*); fruits: plantain (*Musa × paradisiaca*), okra (*Abelmoschus esculentus*); pseudocereals: quinoa (*Chenopodium quinoa*), amaranth (*Amaranthus caudatus* L., *Amaranthus cruentus* L., *Amaranthus hypochondriacus* L.), buckwheat (*Fagopyrum esculentum*), chia (*Salvia hispanica*); root crops: yams (*Dioscorea spp.*, *Dioscoreaceae*), enset (*Eleusine ventricosum*), cassava (*Manihot esculenta*), jicama (*Pachyrhizus erosus*); pulses: chickpea (*Cicer arietinum*), pigeon pea (*Cajanus cajan*), grass pea (*Lathyrus sativus*), cowpea (*Vigna unguiculata*), Bambara groundnut (*Vigna subterranea*). All nutritional data corresponds to the amount in 100g of fresh weight (FW) of each crop before any processing or transformation procedure. Sources: USDA (2016). US Department of Agriculture, Agricultural Research Service, Nutrient Data Laboratory. USDA National Nutrient Database for Standard Reference, Release 28. Institute of Medicine. 2006. Dietary Reference Intakes: The Essential Guide to Nutrient Requirements. Washington, DC: The National Academies Press.*

	DIR	Potato	Rice	Maize	Wheat	Soybean	finger millet	pearl millet	koala millet	fenugreek	horse gram	moth bean	okra	
kcal	2000	77	370	365	327	446	328.00	362	346	367	378	329	122	33
Carbohydrate (g)	60-210	17.5	81.7	74.3	71.2	30.2	72.00	67	59.2	73.1	82.67	72.1	31.9	7.45
Protein (g)	9.1-71	2.05	6.81	9.42	12.61	36.5	7.30	11.7	9.8	13.3	8.5	10.6	1.3	1.93
Vitamin A, RAE (mg)	0.3-1.3	0	0	0.011	0	1	0.00	0.0055	0	0	0	0	0.056	0.036
Vitamin B1 (mg)	0.2-1.4	0.081	0.18	0.385	0.383	0.874	0.48	0.38	0.32	0.39	0.3	0.332	0.062	0.2
Vitamin B6 (mg)	0.1-2.0	0.298	0.107	0.622	0.3	0.377	/	/	/	0.482	/	0.443	0.242	0.215
Vitamin B9 (mg)	0.065-0.6	0.015	0.007	0.019	0.038	0.375	0.02	0.0455	0.0231	0.036	0.0045	0.02	0.022	0.06
Vitamin C (mg)	15-120	19.7	0	0	0	6	0.00	0	0	88	38.5	0	18.4	23
Vitamin E (mg)	4-19	0.01	0	0.49	1.01	0.85	22.00	/	/	0.08	/	0.5	0	0.27
Iron (mg)	0.27-27	0.81	1.6	2.71	3.19	15.7	3.00	8	2.3	7.63	8.48	3.36	0.55	0.62
Zinc (mg)	2-13	0.3	1.2	2.21	2.65	4.89	2.79	3.1	0.7	3.63	3.615	1.67	0.19	0.58
Magnesium (mg)	30-400	23	23	127	126	280	166.35	137	180	184	70	165	36	57
Phosphorous (mg)	100-1250	57	71	210	288	704	229.09	96	188	429	170	289	32	61
Calcium (mg)	200-1300	12	11	7	29	277	281.98	42	27	180	18	13	3	82
Potassium (mg)	400-3400	425	77	287	363	1800	405.00	307	144	427	140	363	487	299

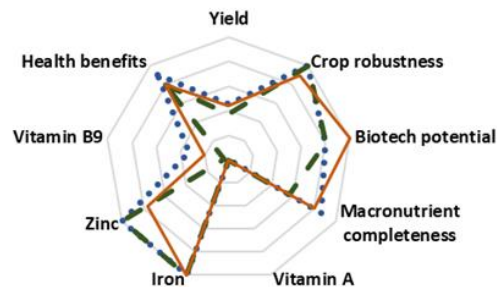
	DIR	Quinoa	Amaranth	Buckwheat	Chickpea	Ense	Cassava	Jicama	Chickpea	Pigeon pea	Grass pea	Cowpea	Bambara groundnut
kcal	2000	368	371	343	415	333	160	38	378	343	335	343	335
Carbohydrate (g)	60-210	64.2	65.2	71.5	12.1	64.8	38.1	8.82	63	62.8	50	59.6	65
Protein (g)	9.1-71	14.1	13.6	13.1	16.5	8.3	1.36	0.72	20.5	21.7	27	23.8	20.5
Vitamin A, RAE (mg)	0.3-1.3	0.014	0	0.021	0.054	0	0.001	0.001	0.003	0.001	0.00969	0.002	/
Vitamin B1 (mg)	0.2-1.4	0.36	0.115	0.4	0.62	0.28	0.087	0.02	0.477	0.643	0.46	0.68	0.47
Vitamin B6 (mg)	0.1-2.0	0.47	0.501	0.73	/	/	0.088	0.042	0.535	0.283	0.58	0.361	0.35
Vitamin B9 (mg)	0.065-0.6	0.184	0.082	0.03	0.049	/	0.027	0.012	0.557	0.456	0.54	0.639	0.21
Vitamin C (mg)	15-120	/	4.2	0	1.6	/	20.6	20.2	4	0	5.2	1.5	0.22
Vitamin E (mg)	4-19	2.44	1.19	5.46	0.5	/	0.19	0.46	0.82	0.39	/	/	8.33
Iron (mg)	0.27-27	4.57	7.61	2.2	7.72	8.6	0.27	0.6	4.31	5.23	6.21	9.95	6.5
Zinc (mg)	2-13	3.1	2.87	2.4	4.58	14.1	0.34	0.16	2.76	2.76	4.4	6.11	0.81
Magnesium (mg)	30-400	197	248	231	335	110	21	12	79	183	123	333	7.02
Phosphorous (mg)	100-1250	457	557	347	860	120	27	18	252	367	47	438	308
Calcium (mg)	200-1300	47	159	18	631	130	16	12	57	130	31	85	10.65
Potassium (mg)	400-3400	563	508	460	407	1790	271	150	718	1390	982	1380	1275

A Cereals: millets



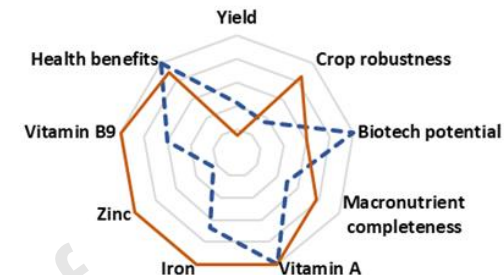
..... Finger Millet - - - Pearl millet - - - Kodo millet

B Cereals: other



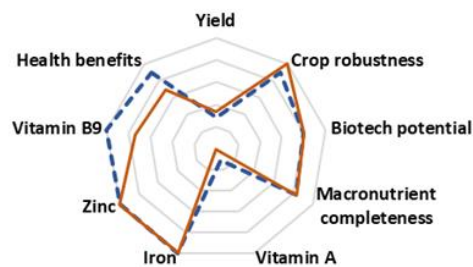
..... Teff - - - Fonio - - - Sorghum

C FRUITS



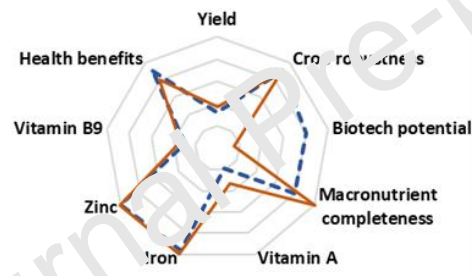
- - - Plantain - - - Okra

D Pseudocereals 1



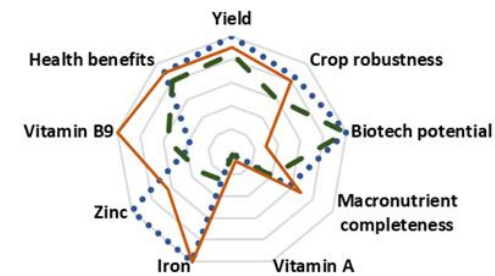
- - - Quinoa - - - Amaranth (grain)

E Pseudocereals 2



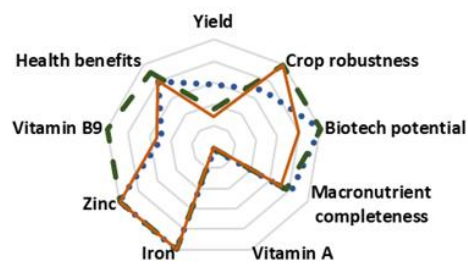
- - - Buckwheat (Common) - - - Chia

F Root crops



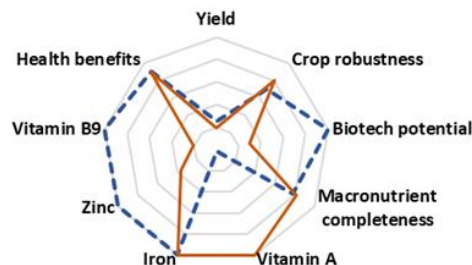
..... Enset - - - Cassava - - - Jicama (Mexican jam bean)

G Pulses 1



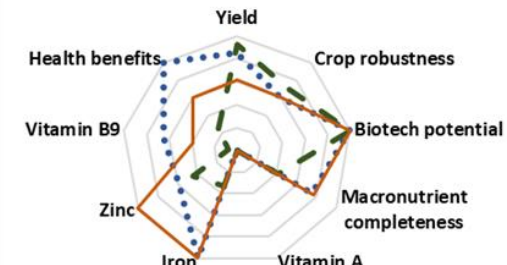
..... Chickpea - - - Pigeon pea - - - Grass pea

H Pulses 2



- - - Cowpea - - - Bamabara groundnut

I Potato, rice and wheat



..... Potato - - - Rice - - - Wheat

Figure 2: Radar graphs of twenty important orphan crops across the world and three major staples. A: Cereals: millets, finger millet (*Eleusine coracana*), pearl millet (*Cenchrus americanus*), kodo millet (*Paspalum scrobiculatum*); B: Cereals: others, teff (*Eragrostis tef*), Fonio (*Digitaria exilis*), sorghum (*Sorghum bicolor*, *Sorghum ssp.*); C: Fruits, plantain (*Musa × paradisiaca*), okra (*Abelmoschus esculentus*); D: Pseudocereals 1: quinoa (*Chenopodium quinoa*), amaranth (*Amaranthus caudatus* L., *Amaranthus cruentus* L., *Amaranthus hypochondriacus* L.); E: Pseudocereals 2, buckwheat (*Fagopyrum esculentum*), chia (*Salvia hispanica*); F: Root crops, enset (*Ensete ventricosum*), cassava (*Manihot esculenta*), jicama (*Pachyrhizus erosus*); G: Pulses 1, chickpea (*Cicer arietinum*), pigeon pea (*Cajanus cajan*), grass pea (*Lathyrus sativus*); H: pulses 2, cowpea (*Vigna unguiculata*), Bambara groundnut (*Vigna subterranea*). I: Major staples, potato (*Solanum tuberosum*), rice (*Oryza sativa*), wheat (*Triticum aestivum*). Crop robustness gives an indication how well the crop copes in high stress environments such as under high temperature and growth on marginal soils. The different parameters on which the crops are scored are yield: average yield + storage robustness; crop robustness: genetic diversity, breeding potential, (a)biotic stress resistance; biotech potential: existence transformation protocol, genome sequenced; completeness: protein, fiber, fat content.; Vitamin A, vitamin B9, iron, zinc: calculation for 50% of recommended daily intake (RDA) in 750 kcal; health benefits: other micronutrient content (Vitamin B1, B6, C and E, Mg, P, Ca), anti-nutritional factors, gluten, anti-oxidant properties, use as medicinal plant.

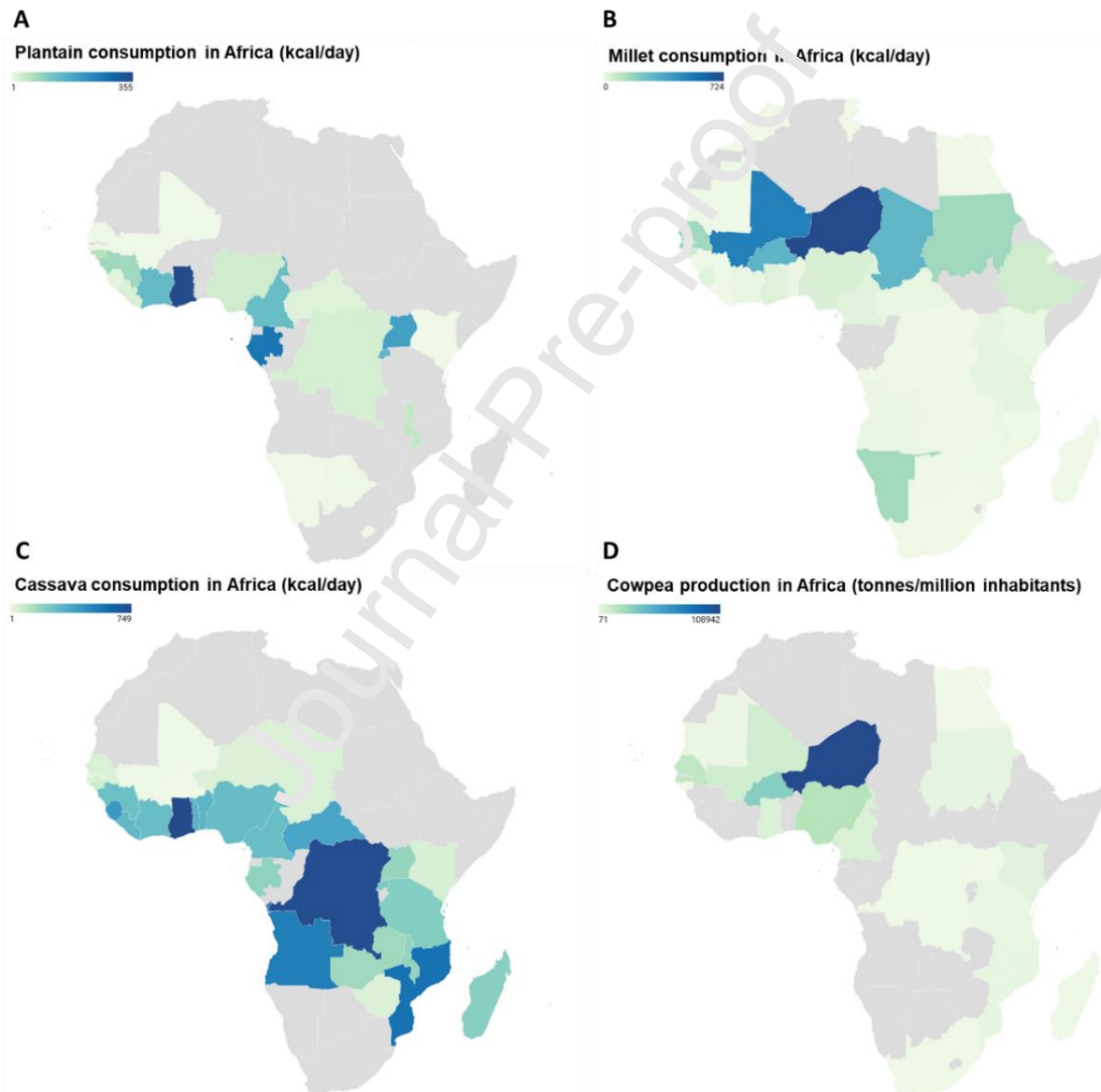


Figure 3: Consumption or production of selected orphan crops in Africa. A, B, C: per capita average daily consumption of plantain (*Musa ssp.*), millet (*Eleusine coracana*, *Cenchrus americanus*) and cassava (*Manihot esculenta*), respectively in kcal/day. D: for cowpea (*Vigna unguiculata*) the average production per capita (tonnes/million inhabitants) is shown since no consumption data are available. Source: FAO, 2020. The maps presented here were created with Datawrapper online software.

Biofortification of orphan crops through breeding

Enhancing iron and zinc

Plantain is recalcitrant to conventional breeding since most cultivars are triploid, have low fertility, and high heterozygosity (Jain and Priyadarshan 2009; D'hont 2012). Using genetic engineering (GE) as an alternative route to enhance the iron and zinc content of plantain (Table 2) is therefore of interest, as it would also allow the biofortification of several micronutrients at once (Naqvi 2009; Simrat Pal Singh, Gruissem, and Bhullar 2017). Nonetheless, some reports indicate that certain plantain varieties possess an iron and zinc content ranging from 0,78-2,53 mg iron and 0,22-3,74 mg zinc 100g⁻¹ fresh weight (FW) (Honfo, Tenkouano, and Coulibaly 2011; Adepoju 2012). Therefore it is needed to perform a broader screening on iron and zinc content in different plantain cultivars, which could then be used for future breeding programs (H.E. Bouis et al. 2011). Biofortification of cassava for higher mineral content through classical breeding has not been pursued because this root crop lacks adequate biodiversity in minerals such as iron and zinc (Chavez et al. 2000; Nanovanan et al. 2019). As such, again, GE has a clear advantage over breeding since it allows introduction of new characteristics into the crop. With respect to millets, high iron content is not consistent in different varieties. In a screening effort of Indian pearl millet varieties, it was shown that iron content ranged from 3,1-10,2mg Fe 100g⁻¹ FW (K. Rai 2013). Due to this biodiversity within millets, breeding seems suitable for biofortification, as for instance done by Harvest Plus (H.E. Bouis et al. 2011; K. Rai 2013; Govindaraj et al. 2016). Finally, in cowpea, the iron and zinc content are exceptionally high compared with most of the other orphan crops discussed in this review and relative to the RDA for both micronutrients (Figure 2; Table 2). Notwithstanding, the anti-nutrient content is high as well, reducing bioavailability (Gonçalves 2016). The concentration of phytate in cowpea varies from 0,26–1,5 per 100g; additionally, cowpea also contains high saponin levels (0,25-3,7g per 100g) which further reduces nutrient bioavailability (Gonçalves 2016). There is nonetheless a clear diversity in anti-nutrient levels in different cultivars. Breeding or engineering for lower phytate levels in different varieties would be a relevant attempt in biofortification efforts.

Enhancing folate

To the best of our knowledge, studies reporting diversity of folate levels of plantain have not been published so far. A limited number of studies in banana shows folate content ranging from 13 to 30 µg 100g⁻¹ FW (Englberger 2009, Ashokkumar et al. 2018; Ningsih and Megia 2019). Whereas breeding thus seems challenging, GE can step in for folate biofortification of plantain. In contrast, both cassava and millet contain a low amount of vitamin B9 (Table 2). To our knowledge, screening efforts to search cassava or millet cultivars with high folate levels are scant. Recently, limited variation has been reported in the high folate containing foxtail millet (*Setaria italica*) (Sandhya et al. 2020). In the absence thereof, as was the case for iron and zinc, GE becomes the favored method. In cowpea on the other hand, a great variation 177.2 to 780.7 µg 100g⁻¹ FW was observed, thereby nicely demonstrating the potential of using high folate germplasm in breeding efforts to increase folate levels of different cowpea genotypes (Nascimento, Cipriano, and Aragão 2022). This is also apparent in other legumes, such as soybean (*Glycine max*) and chickpea (*Cicer arietinum*), where the high folate contents of several cultivars positively point towards the promotion of future breeding efforts (Agyenim-Boateng et al. 2023).

Enhancing provitamin A

Many plantain varieties are rich in pro-vitamin A, yet many highly consumed cultivars contain low amounts of the micronutrient (Kozicka 2021). Consumption of pro-vitamin A rich plantain would

nonetheless bring positive health effects in regions struck by VAD (Figure 1; Figure 2; Figure 3) (Amah 2019; Blomme 2020; Kozicka 2021). To address this issue, Harvest Plus is targeting plantain with specific breeding programs to deliver pro-vitamin A rich plantain in sub-Saharan Africa (www.alliancebioversityciat.org/publications-data/promoting-pro-vitamin-rich-bananas-chronology). Both breeding and GE strategies can be followed for this goal. Since extensive literature (Howarth E. Bouis and Saltzman 2017; HarvestPlus, International Center for Tropical Agriculture (CIAT), Cali, Colombia and Andersson 2017; Amah 2019; CAST 2020) has been produced on the topic of plantain breeding for pro-vitamin A, we will mainly focus on GE here. For cassava, contrary to iron, zinc and folate, several cultivars contain significant amounts of pro-vitamin A (Carvalho 2012; Howarth E. Bouis and Saltzman 2017; Ceballos et al. 2017). Indeed, an elaborate screening effort showed β -carotene content in cassava up to $20,1 \mu\text{g gFW}^{-1}$ which is equal to $168 \mu\text{g RAE } 100\text{g}^{-1}$ (retinol activity equivalents; 12:1 ratio) (H.E. Bouis et al. 2011). Under a Harvest Plus breeding project (www.harvestplus.org/crop/vitamin-a-cassava/), different yellow root cultivars are grown in Nigeria (Howarth E. Bouis and Saltzman 2017). Pro-vitamin A biofortified cassava was shown to be a good source of vitamin A, lowering vitamin A insufficiency upon consumption (Carvalho 2012; Howarth E. Bouis and Saltzman 2017; Ceballos et al. 2017; Afolami 2021b; 2021a). Breeding is thus a very potent and successful approach to enhance pro-vitamin A levels in cassava. Nonetheless, several transgenic approaches have been followed since breeding cassava is time-consuming and difficult (Ceballos et al. 2017), given that genes responsible for high carotenoid content are spread over different loci in the genome which challenges breeders in retaining certain characteristics (including other micronutrients) of local varieties (Ceballos et al. 2017). Additionally, farmers often prefer local varieties. Here, GE clearly has an advantage (Adenle et al. 2012). Finally, for both millets and cowpea, the concentration of pro-vitamin A is very low (Table 2). Thus far, there have been limited efforts for screening carotenoid diversity in different cowpea (Soundar et al. 2022) or millet cultivars (C. B. Yadav et al. 2021).

*Table 2: The (in)ability of plantain, pearl millet, cassava, and cowpea to supply the recommended daily intake (RDA) for vitamin A, folate, iron, and zinc. The micronutrient concentration of plantain is given in relation to the RDA for non-pregnant women of reproductive age for a reference consumption of 423g or 516kcal plantain per day (Honfo, Tenkouano, and Coulibaly 2011). The micronutrient concentration of pearl millet is given in relation to the RDA for non-pregnant women of reproductive age for a reference consumption of 243g or 516 kcal pearl millet per day. The micronutrient concentration of cassava is given in relation to the RDA for non-pregnant women of reproductive age for a reference consumption of 323g or 516 kcal cassava per day (Moura 2015). The micronutrient concentration of cowpea is given in relation to the RDA for non-pregnant women of reproductive age for a reference consumption of 150g or 516 kcal cowpea per day. Reference RDA (Institute of Medicine (U.S.) and Panel on Micronutrients 2002): iron: 18mg; zinc: 9mg; vitamin B9: 400 μg ; vitamin A: 700 μg retinol activity equivalents (RAE). *Note, since molar phytate:iron and phytate:zinc ratios are above bioavailability thresholds (molar ratio > 10) (Honfo, Tenkouano, and Coulibaly 2011), calculations for contribution to RDA for iron and zinc are therefore overestimated (Honfo, Tenkouano, and Coulibaly 2011). **Note, phytate:iron and phytate:zinc ratios are very high in pearl millet, lowering the actual bioavailability of these micronutrients (Krishnan and Meera 2018). FW: fresh weight.*

	Pro-vitamin A (RAE)	Vitamin B9	Iron	Zinc
Plantain (<i>Musa ssp.</i>)				
Current % of RDA	34	23	13*	9*
Desired % of RDA	50	50	50	50
Current concentration (μg per 100g FW)	56	22	550	190
Desired concentration (μg per 100g FW)	81	48	2110	1060
Pearl Millet (<i>Cenchrus americanus</i> or <i>Pennisetum glaucum</i>)				
Current % of RDA	1	16	64**	49**
Desired % of RDA	50	50	/	/
Current concentration (μg per 100g FW)	5	45	8000	3100
Desired concentration	250	142	/	/

(µg per 100g FW)				
Cassava (<i>Manihot esculenta</i>)				
Current % of RDA	0	22	5	12
Desired % of RDA	50	50	50	50
Current concentration (µg per 100g FW)	1	27	270	340
Desired concentration (µg per 100g FW)	109	62	2790	1400
Cowpea (<i>Vigna unguiculata</i>)				
Current % of RDA	0	240	83	102
Desired % of RDA	50	/	/	/
Current concentration (µg per 100g FW)	2	64	9950	6110
Desired concentration (µg per 100g FW)	230	/	/	/

Biofortification through genetic engineering

Enhancing iron and zinc

Biofortification of iron and zinc can be accomplished with several strategies. Interesting methods discussed in our analysis include targeting the plantain fruit to enhance iron storage; facilitating iron transport from the iron rich peel (Arun 2015) to the plantain fruit and lowering the high phytate levels to enhance iron bioavailability. The latter methodology is discussed at length since it is transferable between the different crops.

GE-biofortification strategies for improving mineral content typically rely on improving storage capacity of a given target tissue. The common approach is over-expression of the iron binding protein ferritin (Borg 2012; Trijatmiko 2016). Indeed, fruit specific overexpression of soybean (*Glycine max*) ferritin (*GmFER*) in banana cultivar *Rasthali* led to 6.3-fold and 4.6-fold increase in iron and zinc respectively (G. B. S. Kumar, Srinivas, and Ganapathi 2011). Another approach for iron biofortification is by increasing iron translocation in the plant towards the fruits combined with fruit specific *Ferritin* gene expression. In the banana cultivar *Rasthali* the iron content in the leaves and roots reaches up to 15mg and 30 mg 100g⁻¹ FW respectively (K. Yadav et al. 2017), meaning that a minimal enhancement of translocation could indeed have a desired effect if combined with ferritin expression in the fruits. This can be achieved by altering the endogenous concentration of iron chelating molecules (Trijatmiko 2016). A major class of iron chelating molecules are the phytosiderophores of the mugineic acid (MA) family (

Figure 4A). Of these, nicotianamine (NA) is an Fe²⁺-chelator (Higuchi 1999; Inoue 2003). Overexpression of *NA-synthase* (*NAS*) successfully led to enhanced Fe²⁺-translocation in several crops such as rice and wheat (Trijatmiko 2016; S.P. Singh et al. 2017; Nozoye 2018). Plantain, as nongraminaceous plant, mainly uptakes Fe²⁺ from the soil. Enhancement of nicotianamine concentration by constitutive nicotianamine synthase (*NAS*) overexpression should thus enhance iron translocation in plantain and help in iron biofortification (

Figure 4A).

An alternative strategy is to redirect iron from the peel to the fruit. A proof of concept of this idea was performed in rice grains, where iron was successfully translocated from the surrounding aleurone

tissue to the rice endosperm (Wu, Gruissem, and Bhullar 2019). This was done by expressing *Arabidopsis NATURAL RESISTANCE ASSOCIATED MACROPHAGE PROTEIN (AtNRAMP3)* in an aleurone specific manner (Wu, Gruissem, and Bhullar 2019). This transporter exports iron from vacuoles to the cytosol (Wu, Gruissem, and Bhullar 2019). The strategy proved very successful combined with tissue specific ferritin expression. Plantain peel is rich in iron, accumulating up to 6.96 mg iron 100g⁻¹ FW (Arun 2015), translocation of iron could thus enhance plantain iron content. A challenge lies in identifying peel specific promoters, although this will most likely only require a minimal effort since several elaborate transcriptomic analyses have been performed on banana peels (Mbéguié-A-Mbéguié 2009; Yun 2019).

Mineral bioavailability is highly dependent on anti-nutrient composition in the given crop. The most relevant anti-nutrient is arguably phytic acid or phytate. This compound forms an insoluble complex with iron and zinc at the physiological pH of human intestines, drastically reducing bioavailability of the minerals (Coulibaly, Kouakou, and Chen 2010; Perera, Seneweera, and Hirotsu 2018; Krishnan and Meera 2018). Plantain contains between 0,17-1,23g phytate or phytic acid 100g⁻¹ dry weight (DW), with phytate:iron and phytate:zinc ratios above bioavailability thresholds, lowering the bioavailability of zinc and iron. Phytic acid content in pearl millet, for instance, ranges from 0,5 – 2g 100g⁻¹ FW (Coulibaly, Kouakou, and Chen 2010; Krishnan and Meera 2018).

Phytate is needed for proper plant growth as it is the principal storage form of phosphorus in plants. During seed germination, phytases are upregulated and release a usable form of phosphorous from phytate. Targeting phytate in a crop therefore often leads to reduced seed germination (Coulibaly, Kouakou, and Chen 2010; Perera, Seneweera, and Hirotsu 2018). Attempts in other crops to lower phytate have shown a tendency for yield penalties upon complete degradation of phytate (C. S. Reddy, Kim, and Kaul 2017). Several strategies exist, ranging from expressing phytase that breaks down phytic acid to knocking down enzymes in the biosynthetic pathway (Perera, Seneweera, and Hirotsu 2018; Raboy 2020), but no strategy seems to represent a silver bullet. As brought forward by *Raboy (2020)*, perhaps a yield penalty of 5-10% could be acceptable if bioavailability of several micronutrients drastically increases due to lower phytate levels (Raboy 2020). An important remark is that clonally propagated crops, as is the case for plantain, are not necessarily of concern in respect to reduced germination; for these only the effects on yield are of major consideration. Nonetheless, there are reasons to assume that a targeted approach will allow reduction of phytic acid with only a minimal yield penalty and no germination loss.

One such approach consists of lowering phytic acid content in crops by overexpressing phytase in a tissue-specific manner in the seeds (Raboy 2020). A very potent type of phytase for this purpose is the one found in the bacterium *Bacillus subtilis* as it only dephosphorylates until tri-myo-inositol, rather than full degradation as is the case with most plant based phytases (C. S. Reddy, Kim, and Kaul 2017). This causes a small increase of free inorganic phosphate and of lower inositol phosphates that are less potent mineral chelators (Sparvoli and Cominelli 2015). Indeed, engineering of phytases must be performed with the necessary care, as further dephosphorylation would generate an even lower inositol content and more free inorganic phosphate which play a role in many signaling pathways and thus can have a negative impact (C. S. Reddy, Kim, and Kaul 2017). *BsPhytase* expression in a tissue-specific manner should lower phytate levels while limiting a potential yield penalty. Given that public perception favors utilization of plant-based genetic elements, it might be favorable to use a cisgenic approach and identify or engineer several plant-based phytases with a similar function and express them in the target tissue instead.

Similar as to rice and wheat, phytate in millets is mainly localized in the nutrient dense aleurone layer (Jha, Krishnan, and Meera 2015; Krishnan and Meera 2018; Perera, Seneweera, and Hirotsu 2018).

Several studies in rice successfully silenced genes in the phytic acid biosynthetic pathway in an aleurone specific manner (Kuwano et al. 2009; N. Ali 2013; Karmakar 2020). However, the promoter used in the different studies was also active in the embryo (Kuwano et al. 2009), not exclusively in the aleurone layer. One good approach in millet could therefore be to specifically inhibit phytic acid biosynthesis exclusively in the aleurone layer by RNAi-technology. Ideally, to reduce a negative yield or germination impact, the biosynthetic gene responsible for conversion of tri-myoinositol to phytic acid is targeted rather than silencing the pathway upstream. As such tri-myoinositol can still serve as a phosphorus source for the germinating seedlings while it has a lower impact on bioavailability (Punjabi et al. 2018; Raboy 2020). It is important to consider that not only phytate poses a problem for mineral bioavailability. For instance, pearl millet (and other millets) contains high amounts of polyphenols which also form complexes with iron and further reduce bioavailability (Krishnan and Meera 2018; Hassan, Sebola, and Mabelebele 2021). GE could be used to mitigate this issue by targeting genes that influence the biosynthesis of polyphenolic compounds in millets; however, this could partly lower the positive health impact of millets (Hassan, Sebola, and Mabelebele 2021).

As for cowpea, most phytic acid in seeds is in the cotyledons (Ungara, Morton, and Daniel 1990; Gonçalves 2016). In an innovative study a late-biosynthesis gene of the lipid-dependent-pathway (*inositol polyphosphate 6-/3-/5-kinase (GmIPK2)*) was specifically targeted through the expression of a cotyledon-specific RNAi, thereby still allowing biosynthesis of myoinositol-triphosphate but not phytic acid in the seed (Punjabi et al. 2018). Translating this approach to cowpea could obtain similar successful results with limited side-effects, as this still provides a potent phosphorous source for the emerging seedlings while enhancing micronutrient bioavailability. Since this approach did not result in an observable yield penalty or germination loss in soybean, it is reasonable to expect a similar result in cowpea.

It becomes clear that for all crops, lowering phytic acid content is walking on a tightrope between potential germination loss and yield penalties. Yet, it still holds great potential for the future of iron biofortification. The impact of different processing techniques to prepare food was not considered when accounting for bioavailability. It is important to note that some standard processing techniques such as boiling, soaking and fermentation can have a positive impact on enhancing bioavailability, by for instance breaking down the antinutrients (Devi et al. 2014). Notwithstanding, the actual amount of nutrients available often is negatively impacted when boiling and milling are employed, as they result in breakdown of many vitamins (M. B. Reddy and Love 1999). This is best illustrated by the 10-64% loss in folate content upon boiling leafy vegetables (Maharaj et al. 2015). On the other hand, many orphan crops are rich in enhancers, such as citric acid, which have a positive influence on nutrient bioavailability, examples of this include amaranth and sweet potato (Uusiku et al. 2010).

Enhancing folate

Several studies in crops such as rice (Storozhenko 2007; Blancquaert et al. 2015), potato (*Solanum tuberosum*) (De Lepeleire et al. 2018), tomato (*Solanum lycopersicum*) (Garza, Gregory, and Hanson 2007), lettuce (*Lactuca sativa*) (Nunes, Kalkmann, and Aragão 2009) and others (Simon Strobbe and Van Der Straeten 2017) have shown successful folate enhancement by GE. In rice, folate levels were increased up to 2500µg per 100g FW, making the biofortified cultivar one of the most folate dense crops (Blancquaert et al. 2015). For plantain, the main goal should be folate enhancement rather than enhancing stability, since, in contrast to most other staples, plantain is not stored for long periods, making folate stability less of a concern. Strategies on folate stability are, however, still applicable for millets, cassava, and cowpea, which can be stored for a long period of time.

The final step of folate biosynthesis takes place in mitochondria, while the precursors for biosynthesis, *para*-aminobenzoate (p-ABA) and 6-hydroxymethyl-dihydropterin (HMDHP), are produced in the plastids and cytosol respectively (

Figure 4B) (Simon Strobbe and Van Der Straeten 2017). One very successful GE strategy consists of overexpressing the genes responsible for biosynthesis of the precursors in the targeted tissue. Indeed, the high folate levels in rice were achieved by overexpressing *aminodeoxychorismate synthase (ADCS)* for p-ABA and *GTP cyclohydrolase I (GTPCHI)* for HMDHP accumulation. This strategy also proved successful in tomato (Garza, Gregory, and Hanson 2007). Based on the current scientific knowledge, we put forward the hypothesis that this strategy should be sufficient to obtain the desired folate levels in plantain (Table 2).

In rice the folate content of kernels was enhanced with more than 50-fold compared to wild-type (Storozhenko 2007; Blancquaert et al. 2015). A similar approach has been applied in wheat and maize, albeit the levels reached were by far not as high as in rice (Liang 2019). It stands to reason that an analogous strategy can be used in millets. Presumably the most efficient approach is by overexpressing Arabidopsis *GTPCHI* and *ADCS* in an endosperm specific manner as was applied in rice rather than using the soybean orthologs as has been done in wheat and maize (Storozhenko 2007; Blancquaert et al. 2015; Liang 2019). Since both pearl, finger and kodo millet already contain between 18-45µg vitamin B9 per 100g FW, a 3-8-fold enhancement is sufficient to achieve the targeted goal (Table 2).

Another interesting strategy is to combine *ADCS* and *GTPCH* overexpression with the expression of the bifunctional enzyme *HMDHP pyrophosphokinase/ dihydropteroate synthase (HPPK/DHPS)*, which performs the coupling of both precursors in mitochondria, and *folylpolyglutamate synthase (FPGS)*, which turns folates into a poly-glutamylated form that is retained within the mitochondria (Blancquaert et al. 2015; De Lepeleire et al. 2018). Successful inclusion of *HPPK/DHPS* and *FPGS* expression was demonstrated in potato tubers (Simon Strobbe and Van Der Straeten 2017; De Lepeleire et al. 2018). This led to a 12-fold enhancement of stable folate content. Since cassava already contains folate to some degree, this strategy is specifically interesting as the necessary fold-enhancement is moderate (Table 2). Lastly, folate stability could be enhanced by introducing a folate binding protein (FBP) in the targeted tissue to shield folates from breakdown (Blancquaert et al. 2015; Simon Strobbe and Van Der Straeten 2017). The major challenge here, however, taking public perception into account, is to find a potent FBP of plant origin (Puthusseri 2018), rather than the FBP of mammalian origin that was used as proof of concept, albeit very successful (Blancquaert et al. 2015).

Enhancing pro-vitamin A

The main objective in pro-vitamin A biofortification is the enhancement of β-carotene in a crop, the most potent form of pro-vitamin A. One of the enzymes targeted in almost all strategies is phytoene synthase (*PSY*), a key regulatory enzyme in carotenoid biosynthesis which is often the rate limiting step (Ye and Beyer 1979; Fraser 2002; Paine 2005; Giuliano 2017).

Germplasm screening in different Indian banana cultivars showed a positive correlation between *Musa acuminata (Ma)PSY1* expression and β-carotene content (Kaur 2017), making *PSY* an interesting target for plantain as well. Indeed, expression of a banana-derived *phytoene synthase (MtPsy2a)* gene led to a significant enhancement in the 'Cavendish' dessert banana, generating a so called 'Golden Banana' (Paul 2017). So far, no reports have been published repeating this strategy in plantain,

although overexpressing *PSY* in plantain is promising. The main challenge currently lies in finding promoters that are exclusively active in plantain fruit, and not in any other tissues as was the case for the promoters used for creation of Golden Banana (Paul 2017). This problem needs to be solved for all other biofortification strategies in plantain as well. In addition to *PSY* expression, expression of the *Arabidopsis Orange^{His}* gain of function mutant might be interesting to further enhance β -carotene in plantain as well (L. Li 2012; Yuan 2015; Bai 2016). This gene has a role in chromoplast development and increases the storage stability of β -carotene, next to stabilizing *PSY* (Osorio 2019). Induction of a single nucleotide polymorphism (SNP) resulting in conversion of a conserved Arginine to Histidine in this *Orange* gene made it a powerful chromoplast inducer (Tzuri 2015; Yuan 2015). Overexpression of the “golden-SNP” containing *AtOrange^{His}* or wild type (WT) *AtOrange* gene already proved successful in rice, tomato, sweet potato and other plants (Yuan 2015; Bai 2016; Endo et al. 2019; Yazdani 2019; Kim et al. 2021). In conclusion, although breeding towards provitamin A enriched plantains is needed, our assessment highlights that additional enrichment using GE can be promising. On the other hand, combined micronutrient enhancement is a rational way forward (Van Der Straeten 2020).

For cassava, the most successful strategy thus far generated β -carotene content up to 35 $\mu\text{g g}^{-1}$ DW which also showed enhanced shelf life (Beyene 2018). This was achieved through the root-specific expression of two biosynthesis-related genes, *AtDXS* (deoxy-D-xylulose-5-phosphate synthase) from *A. thaliana* and *PacrB* (phytoene synthase) from the bacterium *Pantoea ananatis*. The high carotenoid content, however, was offset by a drastic reduction in dry matter content and thus a severe yield penalty (Beyene 2018). It was shown that a single nucleotide polymorphism (SNP) in the cassava *PSY2* gene is responsible for β -carotene accumulation in the roots due to increased enzymatic activity (Welsch 2010). Additionally, significant Orange protein accumulation was observed in yellow cassava roots compared to white cassava roots (Jaramillo 2022). Overexpression of cassava *MeOR_X1* together with *ZmPSY* led to very high β -carotene levels in cassava callus (Jaramillo 2022). The cassava *MeOr_X1* gene does not contain the SNP mutation (*Orange^{His}*) referenced previously (Jaramillo 2022). It is thus highly promising to target both the *PSY* and *MeOr_X1* gene of cassava using a gene editing intervention such as CRISPR/Cas to induce the described “golden SNP’s” in both genes simultaneously (Welsch 2010; Endo et al. 2019). This should strongly increase carotenoid biosynthesis and create a sink for carotenoids in the root at the same time.

In the case of pulses and legumes, genetic engineering to successfully increase the β -carotene content has been limited to soybean thus far (Schmidt 2015). This knowledge gap poses a challenge to develop strategies for genetic enhancement of β -carotene in pulses. Nonetheless, several reasonable approaches can be thought out based on current knowledge. In soybean, β -carotene levels up to 845 $\mu\text{g g}^{-1}$ DW were obtained by overexpression of a bacterial phytoene synthase (*PaCrtB*). However, due to the very strong lectin promoter used in this approach, the protein content of the transgenic beans increased drastically. In chickpea it was found that amongst others, *zeta-carotene desaturase* (*ZDS*) and on a minor level *phytoene synthase* (*PSY*) were positively correlated with the carotenoid concentration across different cultivars (Rezaei, Deokar, and Tar’an 2016). Projecting these data on legumes in general, it is reasonable to expect that a similar approach as followed in other crops, such as rice and potato, will be successful in legumes like cowpea as well (Paine 2005; Diretto 2007; Giuliano 2017). This approach consists of tissue specific *PSY/CrtB* and *CrtI* expression. Bacterial *CrtI* can perform the four desaturase steps in carotenoid biosynthesis of which *ZDS* is a part. As for all biofortification strategies, the choice of a suitable promoter will be crucial for cowpea as well. Usage of a too strong promoter might bring negative side effects as observed in soybean by increasing the protein content of the seeds drastically, while a too weak promoter might not result in the desired β -carotene accumulation (Ye and Beyer 1979; Schaub 2012).

In cereal crops, pro-vitamin A biofortification has been successfully performed (Giuliano 2017). In maize, β -carotene levels up to $60\mu\text{g}$ per 100g DW were obtained, while in rice levels up to $37\mu\text{g}$ β -carotene per 100g DW were obtained (Paine 2005; Naqvi 2009). In both crops the same strategy was used, namely endosperm specific overexpression of *ZmPSY* and *Pantoea ananatis* (*Pa*)*CRTI*. Additional to boosting biosynthesis of pro-vitamin A, there is the issue of stability. Beta-carotene, the main plant derived source for vitamin A, is notoriously unstable in certain food matrices as for instance in the rice endosperm (Pénicaud et al. 2011; Bollinedi 2019). It is unclear whether β -carotene would be stable in millets, thus expressing the “golden SNP”-containing *Orange* gene from *Arabidopsis* in the biofortified tissue to stabilize β -carotene could be a valuable strategy, (Lopez et al. 2008; L. Li 2012; Yuan 2015; Bai 2016; Endo et al. 2019). Overall, it is reasonable to assume that tissue specific expression of *ZmPSY*, *PaCRTI* and optionally *AtOr^{His}* will lead to pro-vitamin A enhanced millet.

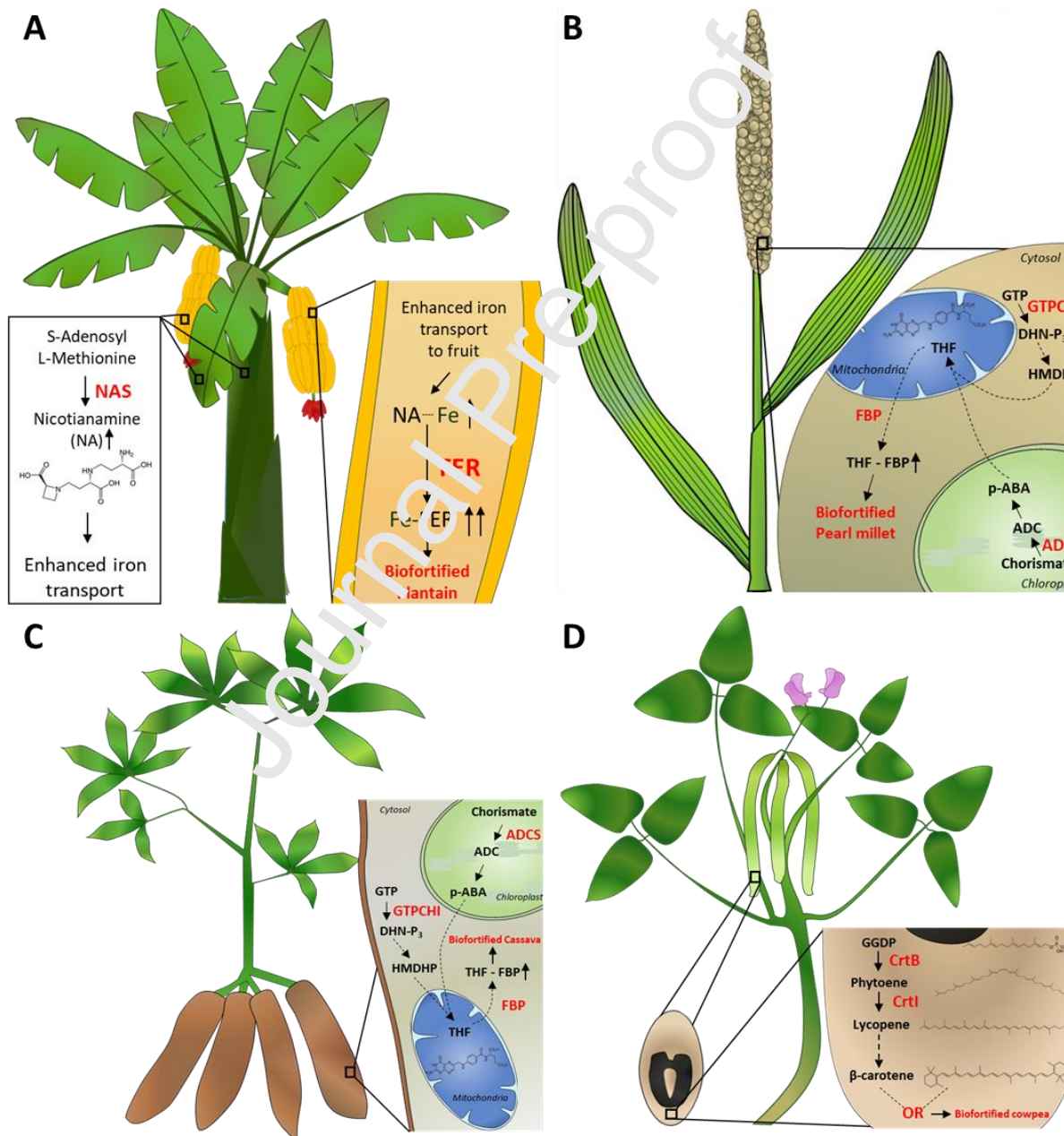


Figure 4: Biofortification strategies for different orphan crops using genetic engineering. A: Development of iron biofortified plantain by constitutive overexpression of *nicotianamine synthase* (*NAS*) and fruit specific expression of *Ferritin* (*FER*). Nicotianamine (NA) is synthesized by from S-Adenosyl L-methionine by action of *NAS*. NA enhances iron

transport in the plant through chelation of Fe^{2+} . In the fruit *Ferritin* binds the iron mobilized inwards by NA-Fe chelates creating iron bound ferritin and biofortified plantain. B: Development of folate biofortified millet by endosperm specific overexpression of *GTP cyclohydrolase I (GTPCH)*, *aminodeoxychorismate synthase (ADCS)* and *folate binding protein (FBP)*. In the pterin branch, 6-hydroxymethylidihydropterin (HMDHP) is synthesized from GTP with the first committed step performed by GTPCHI. In the p-ABA branch the first committed step is performed by ADCS, converting chorismate into aminodeoxychorismate (ADC). Both are transported into the mitochondrion by active/passive transport (dotted line) where they are condensed into tetrahydrofolate (THF). THF is transported to the cytosol by active/passive transport and stabilized by FBP, generating folate biofortified pearl millet. C: Development of biofortified cassava by root specific expression of *GTP cyclohydrolase I (GTPCH)*, *aminodeoxychorismate synthase (ADCS)* and *folate binding protein (FBP)*. See above for description of pathway. D: Development of pro-vitamin A biofortified cowpea by pea specific overexpression of bacterial *Phytoene synthase (CrtB)*, *Phytoene desaturase (CrtI)* and *Arabidopsis Orange (OR)*. CrtB performs the first committed step in carotenoid biosynthesis by condensing two geranylgeranyl diphosphate (GGDP) molecules into phytoene. Phytoene is subsequently converted into all-trans-lycopene by CrtI and converted into β -carotene by endogenous enzymes (dotted arrow). OR induces plastoglobuli and chromoplast formation which stabilizes β -carotene. NAS: Nicotianamine synthase; NA: nicotianamine; Fer: Ferritin; GTP: guanosine triphosphate; GTPCHI: GTP cyclohydrolase I; DHN-P: dihydroneopterin monophosphate; HMDHP: 6-hydroxymethylidihydropterin; THF: tetrahydrofolate; p-ABA: para-aminobenzoate; ADC: aminodeoxychorismate; ADCS: ADC synthase; FBP: folate binding protein; GGDP: geranylgeranyl diphosphate, CrtB: bacterial phytoene synthase; CrtI: bacterial phytoene desaturase; Or: *Orange*. Figure adapted from Strobbe *et al.*, (2017) for folate biosynthesis

Ending hunger in all its forms

Eliminating hunger and malnutrition has been a primary goal of the international community for more than 40 years (*The State of Food Security and Nutrition in the World 2020* 2020). Since around the year 2000, there has been a concerted global effort to present solutions for mitigating hidden hunger (Allen, World Health Organization, and Food and Agriculture Organization of the United Nations 2006). This has resulted in a multi-level approach targeting several fronts. Micronutrient supplementation as well as fortification of food products were deployed on a massive scale, thereby severely reducing the occurrence of deficiencies in the population and addressed (Rautiainen *et al.* 2016; Imdad *et al.* 2017; Keats *et al.* 2019; Tam *et al.* 2020). Though these interventions have helped relieving hidden hunger for millions of people, their reliance on continuous efforts (e.g., distribution) and specialized infrastructure hampers their sustainability and resilience (Rao 2016). An example of the latter includes the currently witnessed exacerbation of micronutrient malnutrition during the COVID-19 pandemic (Headey 2020; Carducci *et al.* 2021; Menisi and Udenigwe 2021; Osendarp 2021). On top of the disruption of supplementation programs (Carducci *et al.* 2021), the COVID-19 pandemic has been reported to have a negative influence on dietary patterns (Fleming and Luo 2021; Rodriguez-Ieyva and Pierce 2021). This confirms the dire need for additional efforts against micronutrient malnutrition by allowing sustainable, local acquisition of micronutrient-dense food.

Biofortification can provide a sustainable, cost-effective additional strategy in the fight against malnutrition (Howarth E. Louis and Saltzman 2017; Steur *et al.* 2017; CAST 2020). These efforts have generally focused on highly consumed staples such as rice (Steur 2015; Tiozon, Fernie, and Sreenivasulu 2021), wheat (M. W. Ali and Borrill 2020; Saini, Devi, and Kaushik 2020; Gupta *et al.* 2021), maize (Maqbool and Beshir 2019; Goredema-matongera 2021) and potato (Diretto 2007; B. Singh *et al.* 2021). Yet, for many communities biofortification of their local crops would prove more beneficial since they are often specifically adapted to the environment in which they grow and are part of local diets.

The hypothesized strategies in this review are based on the current scientific knowledge as proven in different species, so their success would need to be experimentally validated. Here, we propose a mode-of-action that demonstrates how biofortification efforts should be deployed. The selected crops are ideal examples for metabolic engineering. Plantain, pearl millet, cassava and cowpea all have a sequenced genome and several transformation protocols have been published for each crop (Figure 1). It is noteworthy to point out that cassava is technically no orphan crop anymore thanks to recent research attention (Amelework *et al.* 2021). Since cassava is a crop of major importance for sub-

Saharan Africa and research in the crop is still in development, we chose cassava as root crop example, nonetheless. For most other orphan crops this is, unfortunately, not the case. Bambara groundnut for instance has its genome sequenced but is very recalcitrant for genetic transformation, making biofortification through GE very difficult. This immediately shows the biggest weakness of GE, being that applications in less researched crops often are very difficult due to a lack of knowledge about the crop on a molecular level. It is therefore of utmost importance to make use of genetic engineering in combination with conventional breeding to create biofortified crops (Van Der Straeten 2020). Both tools have their inherent limitations, but their combination can have synergistic effects.

Whereas multi-biofortification is very challenging using conventional breeding, it is very feasible through metabolic engineering (Van Der Straeten 2020). The only -but possibly significant- challenge lies in transforming a larger DNA-construct into the crop of interest to target several micronutrients in a single transformation event. Multi-biofortification of specific orphan crops would also boost their value to be implemented in dietary diversification campaigns, advocating for consumption of such nutrient-dense locally produced crops (Amelework et al. 2021).

The role of orphan crops in our agricultural system

The role of orphan crops in the wake of the Covid-19 pandemic

The fight against malnutrition has recently gained an added challenge due to the spread and global impact that COVID-19 has had on human development (Udmale et al. 2020; Osendarp 2021; *The State of Food Security and Nutrition in the World 2021* 2021). Before this pandemic, large areas of the world were already struggling with hunger and poverty; now the pressure has increased and spread to new regions (Udmale et al. 2020). This has proved to be a notable challenge to the resilience of our current food systems, and a reminder that they are not sturdy (Béné 2020). Orphan crops and biofortification can be part of a long-term solution to this systematic fragility. Diversification of our agricultural outputs should be a priority (Hertel et al. 2021), as it minimizes the negative effects of possible future disruptions of staple food production. Thus, the promotion of underutilized, local crops is beneficial. This could aid in reducing our over-reliance on specific crops whilst also contributing to rural economic development (Borelli et al. 2020). Furthermore, because these crops are typically cultivated by marginal populations, it can make them more resilient against possible market fluctuations and improve the substance of their small-scale farmers (Borelli et al. 2020). Improving the nutritional content via biofortification only brings more benefit to orphan crop implementation, as this would help reduce current nutritional gaps of the populations consuming these crops (S. Strobbe and Van Der Straeten 2018). Also, if biofortified orphan crops are widely produced and consumed, diet quality across different regions could improve. As there is a direct link between diet quality and immune function in humans (Childs, Calder, and Miles 2019), this type of intervention would have a positive effect on the health status in targeted regions, and consequently, on productivity and economic performance.

Orphan crops and climate change

Climate change is forcibly driving change within our agricultural system (Kummu et al. 2021). The increasing weather variability witnessed at the moment is expected to worsen in coming decades (Shukla et al. 2019). This will have a significant effect on crop yield (Challinor 2014), and as such calls for adaptation mechanisms that enhance the resilience of our food system. Orphan crops can be key to this problem, as most are adapted to grow in unfavorable conditions (Kamenya et al. 2021). Underutilized plant species have been grown in marginal land, forced to develop stress-coping mechanisms, in order to adapt to the high climate variability that these crops have endured (X. Li,

Yadav, and Siddique 2020). This is well illustrated in the high tolerance to drought stress that has been identified in many orphan crops, including finger millet (Krishnamurthy et al. 2016), grass pea (*Lathyrus sativus*) (Campbell 1997), tef (*Eragrostis tef*) (Assefa et al. 2015), Bambara groundnut (Majola, Gerrano, and Shimelis 2021) and quinoa (Hinojosa et al. 2018). Furthermore, orphan crops have also been shown to be able to cope with low-fertility and/or contaminated soil. Examples of this are finger millet (Thilakarathna and Raizada 2015), amaranth (Chinmayee et al. 2012) and African yam bean (*Sphenostylis stenocarpa*) (Nnamani et al. 2021). Hence, it stands to reason that orphan crops can be deployed as part of climate-adaptation agricultural plans.

Concluding remarks

Here, we have examined different separate biofortification strategies for orphan crops. It should be noted that given the typical small-scale consumption of these crops as compared to the major staples as rice, potato, and wheat, such biofortification strategies would be applicable to more confined populations. This would inevitably result in a lower cost-effectiveness of the biofortification interventions. To counteract this issue, multi-biofortification, the simultaneous increase of several micronutrients at once, seems very promising. Indeed, multi-biofortification appears relevant as almost all researched orphan crops have unsatisfactory levels of several micronutrients. The biofortification strategies described here serve as a blueprint in the fight against the most widespread micronutrient deficiencies.

Biofortified orphan crops are the ideal candidates to be included in dietary diversification programs. Their potential applicability in rural regions makes them a more beneficial strategy, as these are often difficult to reach with supplementation programs (Gebremedhin et al. 2014). It is important to assess whether there is an opportunity to enhance micronutrient content by breeding programs as well. Ideally, combination of breeding and genetic engineering (potentially making use of genome editing tools) can be employed to create elite, micronutrient rich (multi-biofortified) staples.

Acknowledgements

DVDS is grateful to Ghent University for financial support (GOA 01G00409; Bijzonder Onderzoeksfonds UGent). SS was indebted to the Agency for Innovation by Science and Technology in Flanders (IWT) for a predoctoral fellowship. SS gratefully acknowledges the Bijzonder Onderzoeksfonds (BOF-UGent) for a postdoctoral fellowship (BOF.P-DO.2019.0008.01). VV is thankful to FWO for receiving predoctoral grant (1SD0522N). LC would like to thank FWO for predoctoral fellowship 1127322N.

Competing interests statement

The authors declare no financial or commercial conflict of interest.

References

- A. Obisesan, Idowu, Ayobola M. A. Sakpere, Bamidele J. Amujoyegbe, Efere M. Obuotor, and Gbenga E. Ogundepo. 2020. 'Drought Response of a Tropical Legume (*Pachyrhizus erosus* L.): Physio-Biochemical Adjustments at Seedling Stage'. *Journal of Plant Sciences* 16 (1): 1–9. <https://doi.org/10.3923/jps.2021.1.9>.
- Abrouk, Michael, Hanin Ibrahim Ahmed, Philippe Cubry, Denisa Šimoníková, Stéphane Cauet, Yveline Pailles, Jan Bettgenhaeuser, et al. 2020. 'Fonio Millet Genome Unlocks African Orphan Crop Diversity for Agriculture in a Changing Climate'. *Nature Communications* 11 (1): 4488. <https://doi.org/10.1038/s41467-020-18329-4>.
- Adenle, A.A., O.C. Aworh, R. Akromah, and G. Parayil. 2012. 'Developing GM Super Cassava for Improved Health and Food Security: Future Challenges in Africa'. *Agriculture & Food Security* 1.

- Adepoju, Oladejo Thomas. 2012. 'Nutrient Composition and Contribution of Plantain (*Musa Paradisiacea*) Products to Dietary Diversity of Nigerian Consumers'. *African Journal of Biotechnology* 11 (71): 13601–5. <https://doi.org/10.5897/AJB11.3046>.
- Adewale, Babasola Daniel, and Catherine Veronica Nnamani. 2022. 'Introduction to Food, Feed, and Health Wealth in African Yam Bean, a Locked-in African Indigenous Tuberous Legume'. *Frontiers in Sustainable Food Systems* 6. <https://www.frontiersin.org/articles/10.3389/fsufs.2022.726458>.
- Adheka, J. G., D. B. Dhed'a, D. Karamura, G. Blomme, R. Swennen, and E. De Langhe. 2018. 'The Morphological Diversity of Plantain in the Democratic Republic of Congo'. *Scientia Horticulturae* 234 (April): 126–33. <https://doi.org/10.1016/j.scienta.2018.02.034>.
- Afolami, I. 2021a. 'Daily consumption of pro-vitamin A biofortified (yellow) cassava improves serum retinol concentrations in preschool children in Nigeria: a randomized controlled trial'. *American Journal of Clinical Nutrition* 113: 221–31.
- Afolami, I. 2021b. 'The Contribution of Provitamin A Biofortified Cassava to Vitamin A Intake in Nigerian Pre-Schoolchildren'. *British Journal of Nutrition* 126: 1364–72.
- Agyenim-Boateng, Kwadwo Gyapong, Shengrui Zhang, Md Jahidul Islam Shohag, Abdulwahab S. Shaibu, Jing Li, Bin Li, and Junming Sun. 2023. 'Folate Biofortification in Soybean: Challenges and Prospects'. *Agronomy* 13 (1): 241. <https://doi.org/10.3390/agronomy13010241>.
- Ali, Muhammad Waqas, and Philippa Borrill. 2020. 'Applying Genomic Resources to Accelerate Wheat Biofortification'. *Heredity* 125 (6): 386–95. <https://doi.org/10.1038/s41437-020-0326-8>.
- Ali, N. 2013. 'Development of Low Phytate Rice by RNAi Mediated Seed-Specific Silencing of Inositol 1,3,4,5,6-Pentakisphosphate 2-Kinase Gene (IPK1)'. *PLoS One* 8.
- Aliyu, Siise, Festo Massawe, and Sean Mayes. 2016. 'Genetic Diversity and Population Structure of Bambara Groundnut (*Vigna Subterranea* (L.) Verdc.): Synopsis of the Past Two Decades of Analysis and Implications for Crop Improvement Programmes'. *Genetic Resources and Crop Evolution* 63 (6): 925–43. <https://doi.org/10.1007/s10722-016-0406-z>.
- Allen, Lindsay, World Health Organization, and Food and Agriculture Organization of the United Nations. 2006. *Guidelines on Food Fortification with Micro-nutrients*. Geneva; Rome: World Health Organization; Food and Agriculture Organization of the United Nations. <http://catalog.hathitrust.org/api/volumes/oclc/152582146.html>.
- Amah, D. 2019. 'Recent Advances in Banana (*Musa* Spp.) Biofortification to Alleviate Vitamin A Deficiency'. *Critical Reviews in Food Science and Nutrition* 59. <https://doi.org/10.1080/10408398.2018.1495175>.
- Amelework, Assefa B., Michael W. Bairu, Obakeng Maema, Sonja L. Venter, and Mark Laing. 2021. 'Adoption and Promotion of Resilient Crops for Climate Risk Mitigation and Import Substitution: A Case Analysis of Cassava for South African Agriculture'. *Frontiers in Sustainable Food Systems* 5. <https://www.frontiersin.org/article/10.3389/fsufs.2021.617783>.
- Anitha, Seetha, Joanna Kane-Potaka, Rosemary Botha, D. Ian Givens, Nur Liana Binti Sulaiman, Shweta Upadhyay, Mani Vetriventhan, et al. 2021. 'Millets Can Have a Major Impact on Improving Iron Status, Hemoglobin Level, and in Reducing Iron Deficiency Anemia—A Systematic Review and Meta-Analysis'. *Frontiers in Nutrition* 8 (October): 725529. <https://doi.org/10.3389/fnut.2021.725529>.
- Arun, K.B. 2015. 'Plantain Peel - a Potential Source of Antioxidant Dietary Fibre for Developing Functional Cookies'. *J Food Sci Technology* 52: 6355–64.
- Ashokkumar, K., S. Elayabalan, V. Shobana, P. Kumar, and M. Pandiyan. 2018. 'Nutritional Value of Banana (*Musa* Spp.) Cultivars and Its Future Prospects: A Review'. *Current Advances in Agricultural Sciences (An International Journal)* 10: 73.
- Assefa, K., J.-K. Yu, M. Zeid, G. Belay, H. Tefera, and M. E. Sorrells. 2011. 'Breeding Tef [*Eragrostis Tef* (Zucc.) Trotter]: Conventional and Molecular Approaches'. *Plant Breeding* 130 (1): 1–9. <https://doi.org/10.1111/j.1439-0523.2010.01782.x>.

- Assefa, Kebebew, Gina Cannarozzi, Dejene Girma, Rizqah Kamies, Solomon Chanyalew, Sonia Plaza-Wüthrich, Regula Blösch, Abiel Rindisbacher, Suhail Rafudeen, and Zerihun Tadele. 2015. 'Genetic Diversity in Tef [*Eragrostis Tef* (Zucc.) Trotter]'. *Frontiers in Plant Science* 6. <https://www.frontiersin.org/articles/10.3389/fpls.2015.00177>.
- Assefa, Kebebew, Gina Cannarozzi, Dejene Girma, Rizqah Kamies, Solomon Chanyalew, Sonia Plaza-Wüthrich, Regula Blösch, Abiel Rindisbacher, Suhail Rafudeen, and Zerihun Tadele. 2015. 'Genetic Diversity in Tef [*Eragrostis Tef* (Zucc.) Trotter]'. *Frontiers in Plant Science* 6. <https://www.frontiersin.org/article/10.3389/fpls.2015.00177>.
- Assogbadjo, Achille E., Flora Josiane Chadare, Leonard Manda, and Brice Sinsin. 2021. 'A 20-Year Journey Through an Orphan African Baobab (*Adansonia Digitata* L.) Towards Improved Food and Nutrition Security in Africa'. *Frontiers in Sustainable Food Systems* 5 (December): 675382. <https://doi.org/10.3389/fsufs.2021.675382>.
- Aubert, Lauranne, and Muriel Quinet. 2021. 'Temperature Rise and Water Stress Effects on Two Buckwheat Species (*Fagopyrum Esculentum* and *Fagopyrum Tataricum*)'. In <https://dial.uclouvain.be/pr/boreal/object/boreal:250740>.
- Ayenon, Mathieu Anatole Tele, Agyemang Danquah, Léonard Essehoun Aboton, and Kwadwo Ofori. 2017. 'Utilization and Farmers' Knowledge on Pigeon pea Diversity in Benin, West Africa'. *Journal of Ethnobiology and Ethnomedicine* 13 (1): 37. <https://doi.org/10.1186/s13002-017-0144-9>.
- Ayinde, Opeyemi, and Matthew Olaniyi Adewumi. 2009. 'Analysis of Post-Harvest Losses among Plantain/Banana (*Musa* Spp. L.) Marketers in Lagos State, Nigeria'. *Nigerian Journal of Agriculture, Food and Environment* 5 (2-4): 35-38.
- Backiyalakshmi, C., Mani Vetriventhan, Santosh Deshpande, C. Babu, V. Allan, D. Naresh, Rajeev Gupta, and Vania C. R. Azevedo. 2021. 'Genome-Wide Assessment of Population Structure and Genetic Diversity of the Global Finger Millet Germplasm Panel Conserved at the ICRISAT Genebank'. *Frontiers in Plant Science* 12. <https://www.frontiersin.org/articles/10.3389/fpls.2021.692463>.
- Bai, C. 2016. 'Bottlenecks in Carotenoid Biosynthesis and Accumulation in Rice Endosperm Are Influenced by the Precursor-Product Balance'. *Plant Biotechnology Journal* 14: 195-205.
- Bailey, Regan L., Keith P. West Jr., and Robert E. Black. 2015. 'The Epidemiology of Global Micronutrient Deficiencies'. *Annals of Nutrition and Metabolism* 66 (Suppl. 2): 22-33. <https://doi.org/10.1159/000371618>.
- Baoua, I. B., L. Amadou, V. Margam, and L. L. Murdock. 2012. 'Comparative Evaluation of Six Storage Methods for Postharvest Preservation of Cowpea Grain'. *Journal of Stored Products Research* 49 (April): 171-75. <https://doi.org/10.1016/j.jspr.2012.01.003>.
- Barik, D. P., U. Mohapatra, and P. K. Chand. 2005. 'Transgenic Grass pea (*Lathyrus Sativus* L.): Factors Influencing Agrobacterium-Mediated Transformation and Regeneration'. *Plant Cell Reports* 24 (9): 523-31. <https://doi.org/10.1007/s00299-005-0957-5>.
- Bazile, Didier, Sven-Erik Jacobsen, and Alexis Verniau. 2016. 'The Global Expansion of Quinoa: Trends and Limits'. *Frontiers in Plant Science* 7: 622. <https://doi.org/10.3389/fpls.2016.00622>.
- Béné, Christophe. 2020. 'Resilience of Local Food Systems and Links to Food Security – A Review of Some Important Concepts in the Context of COVID-19 and Other Shocks'. *Food Security* 12 (4): 805-22. <https://doi.org/10.1007/s12571-020-01076-1>.
- Berchie, J.N. 2012. 'Evaluation of Five Bambara Groundnut (*Vigna Subterranea* (L.) Verdc.) Landraces to Heat and Drought Stress at Tono-Navrongo, Upper East Region of Ghana'. *AFRICAN JOURNAL OF AGRICULTURAL RESEARCH* 7 (January). <https://doi.org/10.5897/AJAR11.817>.
- Bejene, G. 2018. 'Provitamin A Biofortification of Cassava Enhances Shelf Life but Reduces Dry Matter Content of Storage Roots Due to Altered Carbon Partitioning into Starch'. *Plant Biotechnology Journal* 16: 1186-1200.

- Bhatt, Ritika, Prem Prakash Asopa, Rohit Jain, Aditi Kothari-Chajer, Shanker Lal Kothari, and Sumita Kachhwaha. 2021. 'Optimization of Agrobacterium Mediated Genetic Transformation in Paspalum Scrobiculatum L. (Kodo Millet)'. *Agronomy* 11 (6): 1104. <https://doi.org/10.3390/agronomy11061104>.
- Birhanu, Asaye, Yismaw Degenet, and Zeyinu Tahir. 2020. 'Yield and Agronomic Performance of Released Tef [*Eragrostis Tef* (Zucc.) Trotter] Varieties under Irrigation at Dembia, North-western, Ethiopia'. Edited by Manuel Tejada Moral. *Cogent Food & Agriculture* 6 (1): 1762979. <https://doi.org/10.1080/23311932.2020.1762979>.
- Birmeta, Genet. 2004. 'Genetic Variability and Biotechnological Studies for the Conservation and Improvement of Ensete Ventricosum'. Doctoral Thesis, Alnarp: Swedish University of Agricultural Sciences.
- Biol, Ekin, and Howarth E. Bouis. 2023. 'Role of Socio-Economic Research in Developing, Delivering and Scaling New Crop Varieties: The Case of Staple Crop Biofortification'. *Frontiers in Plant Science* 14. <https://www.frontiersin.org/articles/10.3389/fpls.2023.1099496>.
- Bisht, I.S. 2014. 'Subsistence Farming, Agrobiodiversity, and Sustainable Agriculture: A Case Study'. *Agroecology and Sustainable Food Systems* 38: 890–912.
- Blancquaert, Dieter, Jeroen Van Daele, Simon Strobbe, Filip Kiekens, Sergei Prozhenko, Hans De Steur, Xavier Gellynck, Willy Lambert, Christophe Stove, and Dominique Van Der Straeten. 2015. 'Improving Folate (Vitamin B9) Stability in Biofortified Rice through Metabolic Engineering'. *Nature Biotechnology* 33 (10): 1076–78. <https://doi.org/10.1038/nbt.3358>.
- Blomme, G. 2020. 'Pro-vitamin A Carotenoid Content of 48 Plantain (Musa AAB Genome) Cultivars Sourced from Eastern Democratic Republic of Congo'. *Journal of the Science of Food and Agriculture* 100: 634–47.
- Bochicchio, Rocco, Tim D. Philips, Stella Lovelli, Rosanna Leotta, Fernanda Galgano, Antonio Di Marisco, Michele Perniola, and Mariana Amato. 2015. 'Innovative Crop Productions for Healthy Food: The Case of Chia (*Salvia Hispanica* L.)'. In *The Sustainability of Agro-Food and Natural Resource Systems in the Mediterranean Basin*, edited by Antonella Vastola, 29–45. Cham: Springer International Publishing. https://doi.org/10.1007/978-3-319-16357-4_3.
- Bollinedi, H. 2019. 'Kinetics of β -Carotene Degradation under Different Storage Conditions in Transgenic Golden Rice[®] Lines'. *Food Chemistry* 278: 773–79.
- Borelli, Teresa, Danny Hunter, Stefano Pasculli, Nadezda Amaya, Gennifer Meldrum, Daniela Moura de Oliveira Beltrame, Gamini Samarasinghe, et al. 2020. 'Local Solutions for Sustainable Food Systems: The Contribution of Orphan Crops and Wild Edible Species'. *Agronomy* 10 (2): 231. <https://doi.org/10.3390/agronomy10020231>.
- Borg, S. 2012. 'Wheat Ferritins: Improving the Iron Content of the Wheat Grain'. *J Cereal Sci* 56: 204–13.
- Borrell, James S., Mark Godwin, Guy Blomme, Kim Jacobsen, Abebe M. Wendawek, Dawd Gashu, Ermias Lulekal, Zemed Asfaw, Sebebe Demissew, and Paul Wilkin. 2020. 'Enset-Based Agricultural Systems in Ethiopia: A Systematic Review of Production Trends, Agronomy, Processing and the Wider Food Security Applications of a Neglected Banana Relative'. *Plants, People, Planet* 2 (3): 212–28. <https://doi.org/10.1002/ppp3.10084>.
- Borrell, James S., Zelalem Gebremariam, and Wendawek M. Abebe. 2021. 'Utilize Existing Genetic Diversity before Genetic Modification in Indigenous Crops'. *Nature Biotechnology* 39 (9): 1064–65. <https://doi.org/10.1038/s41587-021-01048-6>.
- Bouis, H.E., C. Hotz, B. McClafferty, J.v Meenakshi, and W.H. Pfeiffer. 2011. 'Biofortification: A New Tool to Reduce Micronutrient Malnutrition'. *Food Nutrition Bulletin* 32: 31–40.
- Bouis, Howarth E., and Amy Saltzman. 2017. 'Improving Nutrition through Biofortification: A Review of Evidence from Harvest Plus, 2003 through 2016'. *Global Food Security* 12 (March): 49–58. <https://doi.org/10.1016/j.gfs.2017.01.009>.
- Branca, Francesco, Alessandro Demaio, Emorn Udomkesmalee, Phillip Baker, Victor M Aguayo, Simon Barquera, Katie Dain, et al. 2020. 'A New Nutrition Manifesto for a New Nutrition Reality'. *The Lancet* 395 (10217): 8–10. [https://doi.org/10.1016/S0140-6736\(19\)32690-X](https://doi.org/10.1016/S0140-6736(19)32690-X).

- Brauw, Alan de, Patrick Eozenou, Daniel O. Gilligan, Christine Hotz, Neha Kumar, and J.v. Meenakshi. 2018. 'Biofortification, Crop Adoption and Health Information: Impact Pathways in Mozambique and Uganda'. *American Journal of Agricultural Economics* 100 (3): 906–30. <https://doi.org/10.1093/ajae/aay005>.
- Bredeson, Jessen V., Jessica B. Lyons, Simon E. Prochnik, G. Albert Wu, Cindy M. Ha, Eric Edsinger-Gonzales, Jane Grimwood, et al. 2016. 'Sequencing Wild and Cultivated Cassava and Related Species Reveals Extensive Interspecific Hybridization and Genetic Diversity'. *Nature Biotechnology* 34 (5): 562–70. <https://doi.org/10.1038/nbt.3535>.
- Bruce, Myron A., and Jessica L. Shoup Rupp. 2019. 'Agrobacterium-Mediated Transformation of *Solanum Tuberosum* L., Potato'. *Methods in Molecular Biology (Clifton, N.J.)* 1864: 203–23. https://doi.org/10.1007/978-1-4939-8778-8_15.
- Burchi, Francesco, Jessica Fanzo, and Emile Frison. 2011. 'The Role of Food and Nutrition System Approaches in Tackling Hidden Hunger'. *International Journal of Environmental Research and Public Health* 8 (2): 358–73. <https://doi.org/10.3390/ijerph8020358>.
- Campbell, C. G. 1997. Grass Pea, *Lathyrus Sativus* L. Promoting the Conservation and Use of Underutilized and Neglected Crops, 18. <https://www.bioversityinternational.org/e-library/publications/detail/grass-pea-lathyrus-sativus-l/>.
- Cannarozzi, Gina, Sonia Plaza-Wüthrich, Korinna Esfeld, Stéphanie Harari, Yi Song Wilson, Dejene Girma, Edouard de Castro, et al. 2014. 'Genome and Transcriptome Sequencing Identifies Breeding Targets in the Orphan Crop Tef (*Eragrostis Tef*)'. *BMC Genomics* 15 (1): 581. <https://doi.org/10.1186/1471-2164-15-581>.
- Cantwell, M., W. Orozco, V. Rubatzky, and L. Hernández. 1992. 'Postharvest Handling and Storage of Jicama Roots'. *Acta Horticulturae*, no. 318 (November): 333–44. <https://doi.org/10.17660/ActaHortic.1992.318.46>.
- Carducci, B., E. C. Keats, M. Ruel, L. Haddad, S. J. M. Osendarp, and Z. A. Bhutta. 2021. 'Food Systems, Diets and Nutrition in the Wake of COVID-19'. *Nature Food* 2 (2): 68–70. <https://doi.org/10.1038/s43016-021-00233-9>.
- Carvalho, Lucia M J. 2012. 'Retention of Total Carotenoid and β -Carotene in Yellow Sweet Cassava (*Manihot Esculenta* Crantz) after Domestic Cooking'. *Food Nutrition Research* 56: 15788.
- CAST, S.O. 2020. 'Issue Paper Food Biofortification—Reaping the Benefits of Science to Overcome Hidden Hunger A Paper in the Series on The Need for Agricultural Innovation to Sustainably Feed the World by 2050 Chapter 1. Justification for Biofortification'.
- Castellanos-Arévalo, Andrea P., Andrés A. Estrada-Luna, José L. Cabrera-Ponce, Eliana Valencia-Lozano, Humberto Herrera-Ubaldo, Stefan de Folter, Alejandro Blanco-Labra, and John P. Délano-Frier. 2020. 'Agrobacterium Rhizogenes-Mediated Transformation of Grain (*Amaranthus Hypochondriacus*) and Leafy (*A. Hybridus*) Amaranths'. *Plant Cell Reports* 39 (9): 1143–60. <https://doi.org/10.1007/s00299-020-02553-9>.
- Castrillón-Arbeláez, Paula Andrea, John Paul Délano Frier, Paula Andrea Castrillón-Arbeláez, and John Paul Délano Frier. 2016. Secondary Metabolism in *Amaranthus* Spp. — A Genomic Approach to Understand Its Diversity and Responsiveness to Stress in Marginally Studied Crops with High Agronomic Potential. *Abiotic and Biotic Stress in Plants - Recent Advances and Future Perspectives*. IntechOpen. <https://doi.org/10.5772/61820>.
- Ceballos, Hernán, Carlos A. Iglesias, Juan C. Pérez, and Alfred G.O. Dixon. 2004. 'Cassava Breeding: Opportunities and Challenges'. *Plant Molecular Biology* 56 (4): 503–16. <https://doi.org/10.1007/s11103-004-5010-5>.
- Ceballos, Hernán, Fabrice Davrieux, Elise F. Talsma, John Belalcazar, Paul Chavarriaga, and Meike S. Andersson. 2017. 'Carotenoids in Cassava Roots'. In *Carotenoids*, edited by Dragan J. Cvetkovic and Goran S. Nikolic. InTech. <https://doi.org/10.5772/intechopen.68279>.
- Chadare, F. J., A. R. Linnemann, J. D. Hounhouigan, M. J. R. Nout, and M. A. J. S. Van Boekel. 2008. 'Baobab Food Products: A Review on Their Composition and Nutritional Value'. *Critical Reviews in Food Science and Nutrition* 49 (3): 254–74. <https://doi.org/10.1080/10408390701856330>.
- Challinor, A.J. 2014. 'A Meta-Analysis of Crop Yield under Climate Change and Adaptation'. *Nat Clim Chang* 4: 287–91.

- Chang, Yue, Huan Liu, Min Liu, Xuezhu Liao, Sunil Kumar Sahu, Yuan Fu, Bo Song, et al. 2019. 'The Draft Genomes of Five Agriculturally Important African Orphan Crops'. *GigaScience* 8 (3): giy152. <https://doi.org/10.1093/gigascience/giy152>.
- Chaudhary, A., D. Gustafson, and A. Mathys. 2018. 'Multi-indicator sustainability assessment of global food systems'. *Nature Communications* 9: 1–13.
- Chavez, A. L., J. M. Bedoya, T. Sánchez, C. Iglesias, H. Ceballos, and W. Roca. 2000. 'Iron, Carotene, and Ascorbic Acid in Cassava Roots and Leaves'. *Food and Nutrition Bulletin* 21 (4): 410–13. <https://doi.org/10.1177/156482650002100413>.
- Che, Ping, Shujun Chang, Marissa K. Simon, Zhifen Zhang, Ahmed Shaharyar, Jesse Ourada, Dennis O'Neill, et al. 2021. 'Developing a Rapid and Highly Efficient Cowpea Regeneration, Transformation and Genome Editing System Using Embryonic Axis Explants'. *The Plant Journal: For Cell and Molecular Biology* 106 (3): 817–30. <https://doi.org/10.1111/tpj.15202>.
- Childs, Caroline E., Philip C. Calder, and Elizabeth A. Miles. 2019. 'Diet and Immune Function'. *Nutrients* 11 (8): 1933. <https://doi.org/10.3390/nu11081933>.
- Chinmayee, M. Devi, B. Mahesh, S. Pradesh, I. Mini, and T. S. Swarna. 2012. 'The Assessment of Phytoremediation Potential of Invasive Weed *Amaranthus Spinosus* L'. *Applied Biochemistry and Biotechnology* 167 (6): 1550–59. <https://doi.org/10.1007/s12010-012-9657-0>.
- Chowdhury, Mahboob A., and Alfred E. Slinkard. 2000. 'Genetic Diversity in Grass pea (*Lathyrus Sativus* L.)'. *Genetic Resources and Crop Evolution* 47 (2): 163–69. <https://doi.org/10.1023/A:1008760604990>.
- Coulibaly, A., B. Kouakou, and J. Chen. 2010. 'Phytic Acid in Cereal Grains: Structure, Healthy or Harmful Ways to Reduce Phytic Acid in Cereal Grains and Their Effect on Nutritional Quality'. *American Journal of Plant Nutrition and Fertilization Technology* 1: 1–22.
- D'hont, A. 2012. 'The Banana (*Musa Acuminata*) Genome and the Evolution of Monocotyledonous Plants'. *Nature* 488 : 213–17.
- D'Hont, Angélique, France Denoeud, Jean-Marc Aury, Franc-Christophe Baurens, Françoise Carreel, Olivier Garsmeur, Benjamin Noel, et al. 2012. 'The Banana (*Musa Acuminata*) Genome and the Evolution of Monocotyledonous Plants'. *Nature* 488 (7410): 213–17. <https://doi.org/10.1038/nature11241>.
- Danielsen, Solveig, Alejandro Bonifacio, and Teresa Ames. 2003. 'Diseases of Quinoa (*Chenopodium Quinoa*)'. *Food Reviews International* 19 (1–2): 43–59. <https://doi.org/10.1081/FRI-120018867>.
- Darnton-Hill, I. 2005. 'Micronutrient Deficiencies and Gender: Social and Economic Costs'. *American Journal of Clinical Nutrition* 81.
- Darnton-Hill, I. 2012. 'Global Burden and Significance of Multiple Micronutrient Deficiencies in Pregnancy'. *Nestle Nutrition Inst Works Ser* 70: 49–60.
- Darnton-Hill, I., and U.C. Mkparu. 2015. 'Micronutrients in Pregnancy in Low- and Middle-Income Countries'. *Nutrients* 7: 1744.
- De Lepeleire, Jolien, Simon Strobbe, Jana Verstraete, Dieter Blancquaert, Lars Ambach, Richard G.F. Visser, Christophe Stove, and Dominique Van Der Straeten. 2018. 'Folate Biofortification of Potato by Tuber-Specific Expression of Four Folate Biosynthesis Genes'. *Molecular Plant* 11 (1): 175–88. <https://doi.org/10.1016/j.molp.2017.12.008>.
- DEBNATH, N., Md Rasul, M. SARKER, M. RAHMAN, and A. PAUL. 2008. 'Genetic Divergence in Buckwheat (*Fagopyrum Esculentum* Moench.)'. *International Journal of Sustainable Crop Production* 3 (2): 60–68.
- Dépigny, Sylvain, Elodie Delrieu Wils, Philippe Tixier, Michel Ndoumbé Keng, Christian Cilas, Thierry Lescot, and Patrick Jagoret. 2019. 'Plantain Productivity: Insights from Cameroonian Cropping Systems'. *Agricultural Systems* 168 (January): 1–10. <https://doi.org/10.1016/j.agsy.2018.10.001>.
- Devi, Palanisamy Bruntha, Rajendran Vijayabharathi, Sathyaseelan Sathyabama, Nagappa Gurusiddappa Malleshi, and Venkatesan Brindha Priyadarisini. 2014. 'Health Benefits of Finger Millet (*Eleusine Coracana* L.)

- Polyphenols and Dietary Fiber: A Review'. *Journal of Food Science and Technology* 51 (6): 1021–40. <https://doi.org/10.1007/s13197-011-0584-9>.
- Dhankhar, S. K., and A. V. V. Koundinya. 2020. 'Accelerated Breeding in Okra'. In *Accelerated Plant Breeding, Volume 2: Vegetable Crops*, edited by Satbir Singh Gosal and Shabir Hussain Wani, 337–53. Cham: Springer International Publishing. https://doi.org/10.1007/978-3-030-47298-6_12.
- Diack, Oumar, Ndjido A. Kane, Cecile Berthouly-Salazar, Mame C. Gueye, Baye M. Diop, Amadou Fofana, Ousmane Sy, et al. 2017. 'New Genetic Insights into Pearl Millet Diversity As Revealed by Characterization of Early- and Late-Flowering Landraces from Senegal'. *Frontiers in Plant Science* 8. <https://www.frontiersin.org/articles/10.3389/fpls.2017.00818>.
- Díaz-Valderrama, Jorge R., Anastasia W. Njoroge, Dennis Macedo-Valdivia, Nancy Orihuela-Ordóñez, Bradley W. Smith, Victor Casa-Coila, Nelly Ramírez-Calderón, Jackeline Zanabria-Gálvez, Charles Woloshuk, and Dieudonne Baributsa. 2020. 'Postharvest Practices, Challenges and Opportunities for Grain Producers in Arequipa, Peru'. *PLOS ONE* 15 (11): e0240857. <https://doi.org/10.1371/journal.pone.0240857>.
- Diretto, G. 2007. 'Metabolic Engineering of Potato Carotenoid Content through Tuber-Specific Overexpression of a Bacterial Mini-Pathway'. *PLoS One* 2: 350.
- Dixit, Girish Prasad, Ashok Kumar Parihar, Abhishek Bohra, and Narenra Pratap Singh. 2016. 'Achievements and Prospects of Grass Pea (*Lathyrus Sativus* L.) Improvement for Sustainable Food Production'. *The Crop Journal, Grain Legume Pulses*, 4 (5): 407–16. <https://doi.org/10.1016/j.cj.2016.06.008>.
- El Bilali, Hamid. 2019. 'Research on Agro-Food Sustainability Transitions: Where Are Food Security and Nutrition?' *Food Security* 11 (3): 559–77. <https://doi.org/10.1007/s12571-019-00922-1>.
- Elegba, Wilfred, Emily McCallum, Wilhelm Gruissem, and Hervé Vanderschuren. 2021. 'Efficient Genetic Transformation and Regeneration of a Farmer-Preferred Cassava Cultivar from Ghana'. *Frontiers in Plant Science* 12: 668042. <https://doi.org/10.3389/fpls.2021.668042>.
- Emmrich, Peter M. F., Abhimanyu Sarkar, Isaac Njau, Gemy George Kaithakottil, Noel Ellis, Christopher Moore, Anne Edwards, et al. 2020. 'A Draft Genome of Grass Pea (*Lathyrus Sativus*), a Resilient Diploid Legume'. *bioRxiv*. <https://doi.org/10.1101/2020.04.24.058167>.
- Endo, A., H. Saika, M. Takemura, N. Misawa, and S. Toki. 2019. 'A Novel Approach to Carotenoid Accumulation in Rice Callus by Mimicking the Cauliflower Orange Mutation via Genome Editing'. *Rice* 12.
- Englberger, L. 2009. 'Carotenoid and Vitamin Content of Karat and Other Micronesian Banana Cultivars'. <https://doi.org/10.1080/09637420600372010>.
- Fakrudin, B., T. N. Lakshmidharmna, J. Ugalat, Raghavendra Gunnaiah, J. Khan, S. P. Gautham Suresh, K. A. Apoorva, et al. 2021. 'Genomic Designing for Biotic Stress Resistance in Sorghum'. In *Genomic Designing for Biotic Stress Resistant Cereal Crops*, edited by Chittaranjan Kole, 213–55. Cham: Springer International Publishing. https://doi.org/10.1007/978-3-030-75879-0_5.
- Fang, Jinggui, Chih-Cheng T. Chao, Philip A. Roberts, and Jeffrey D. Ehlers. 2007. 'Genetic Diversity of Cowpea [*Vigna Unguiculata* (L.) Walp.] in Four West African and USA Breeding Programs as Determined by AFLP Analysis'. *Genetic Resources and Crop Evolution* 54 (6): 1197–1209. <https://doi.org/10.1007/s10722-006-9101-9>.
- Fanta, Solomon Workneh, and Satheesh Neela. 2019. 'A Review on Nutritional Profile of the Food from Enset: A Staple Diet for More than 25 per Cent Population in Ethiopia'. *Nutrition & Food Science* 49 (5): 824–43. <https://doi.org/10.1108/NFS-11-2018-0306>.
- FAO. n.d. 'Yam Bean'. Food and Agriculture Organization of the United Nations. Accessed 19 April 2023. <http://www.fao.org/traditional-crops/yambean/en/>.
- Ferguson, Morag E., Trushar Shah, Peter Kulakow, and Hernan Ceballos. 2019. 'A Global Overview of Cassava Genetic Diversity'. *PLOS ONE* 14 (11): e0224763. <https://doi.org/10.1371/journal.pone.0224763>.

- Ferris, Alex C., Richard O. J. H. Stutt, David Godding, and Christopher A. Gilligan. 2020. 'Computational Models to Improve Surveillance for Cassava Brown Streak Disease and Minimize Yield Loss'. *PLOS Computational Biology* 16 (7): e1007823. <https://doi.org/10.1371/journal.pcbi.1007823>.
- Finkelstein, J.L. 2015. 'A Randomized Trial of Iron-Biofortified Pearl Millet in School Children in India'. *Journal of Nutrition* 145: 1576–81.
- Firoz, Za, Ma Islam, M Mohiuddin, and Mm Rahman. 2014. 'Yield and Yield Attributes of Okra as Influenced by Planting Time and Plant Spacing in Hill Slope Condition'. *Progressive Agriculture* 18 (2): 67–73. <https://doi.org/10.3329/pa.v18i2.18161>.
- Fleming, E., and Y. Luo. 2021. 'Dietary Patterns During National Lockdowns Are at Odds with Recommendations for Preventing Morbidity and Mortality of COVID-19'. *ES Food & Agroforestry*. <https://doi.org/10.30919/esfaf472>.
- Flint-Garcia, Sherry A. 2013. 'Genetics and Consequences of Crop Domestication'. Review-article. ACS Publications. American Chemical Society. World. 14 June 2013. <https://doi.org/10.1021/jf305511d>.
- Fraser, P.D. 2002. 'Evaluation of Transgenic Tomato Plants Expressing an Additional Phytoene Synthase in a Fruit-Specific Manner'. *Proc Natl Acad Sci U S A* 99: 1092–97.
- Garg, M. 2018. 'Biofortified Crops Generated by Breeding, Agronomy, and Transgenic Approaches Are Improving Lives of Millions of People around the World'. *Front Nutr* 5: 12.
- Garza, R.I.D., J.F. Gregory, and A.D. Hanson. 2007. 'Folate Biofortification of Tomato Fruit'. *Proc Natl Acad Sci U S A* 104: 4218–22.
- Gebre, Endale, Likyelesh Gugsa, Urte Schlüter, and Karl Kurzer. 2013. 'Transformation of Tef (*Eragrostis Tef*) by *Agrobacterium* through Immature Embryo Regeneration System for Inducing Semi-Dwarfism'. *South African Journal of Botany* 87 (July): 9–17. <https://doi.org/10.1016/j.sajb.2013.03.004>.
- Gebremedhin, S., A. Samuel, G. Mamo, T. Moge, and T. Assefa. 2014. 'Coverage, Compliance and Factors Associated with Utilization of Iron Supplementation during Pregnancy in Eight Rural Districts of Ethiopia: A Cross-Sectional Study'. *BMC Public Health* 14: 1–8.
- Gerura, Fetta Negash, Beira Hailu Meressa, Ivaldo Martina, Abush Tesfaye, Temesgen Magule Olango, and Yao Nasser. 2019. 'Genetic Diversity and Population Structure of Enset (*Ensete Ventricosum* Welw Cheesman) Landraces of Gurage Zone, Ethiopia'. *Genetic Resources and Crop Evolution* 66 (8): 1813–24. <https://doi.org/10.1007/s10722-019-00325-2>.
- Gillespie, Stuart, and Mara van der Bold. 2017. 'Agriculture, Food Systems, and Nutrition: Meeting the Challenge'. *Global Challenges* 1 (3): 1600002. <https://doi.org/10.1002/gch2.201600002>.
- Giuliano, Giovanni. 2017. 'Provitamin A Biofortification of Crop Plants: A Gold Rush with Many Miners'. *Current Opinion in Biotechnology* 44 (April): 169–80. <https://doi.org/10.1016/j.copbio.2017.02.001>.
- Gonçalves, A. 2016. 'Cowpea (*Vigna Unguiculata* L. Walp), a Renewed Multipurpose Crop for a More Sustainable Agri-Food System: Nutritional Advantages and Constraints'. *J Sci Food Agric* 96: 2941–51.
- Gonçalves-Dias, José, and Markus G. Stetter. 2021. 'PopAmaranth: A Population Genetic Genome Browser for Grain Amaranths and Their Wild Relatives'. *G3 (Bethesda, Md.)* 11 (7): jkab103. <https://doi.org/10.1093/g3journal/jkab103>.
- Goredema-matongera, N. 2021. 'Multinutrient Biofortification of Maize (*Zea Mays* L.) in Africa: Current Status, Opportunities and Limitations'. *Nutrients* 13: 1039 13, 1039.
- Goron, Travis L., and Manish N. Raizada. 2015. 'Genetic Diversity and Genomic Resources Available for the Small Millet Crops to Accelerate a New Green Revolution'. *Frontiers in Plant Science* 6. <https://www.frontiersin.org/articles/10.3389/fpls.2015.00157>.
- Govindaraj, M., K.N. Rai, A. Kanatti, G. Velu, and H. Shivade. 2016. 'Breeding High-Iron Pearl Millet Cultivars: Present Status and Future Prospects'. In *2nd International Conference on Global Food Security 2015*.

- Govindaraj, Mahalingam, Kedar N. Rai, Anand Kanatti, Hari D. Upadhyaya, Harshad Shivade, and Aluri S. Rao. 2020. 'Exploring the Genetic Variability and Diversity of Pearl Millet Core Collection Germplasm for Grain Nutritional Traits Improvement'. *Scientific Reports* 10 (1): 21177. <https://doi.org/10.1038/s41598-020-77818-0>.
- Gregory, P.J. 2019. 'Crops For the Future (CFF): An Overview of Research Efforts in the Adoption of Underutilised Species'. *Planta* 250: 979–88.
- Gupta, P.K., H.S. Balyan, S. Sharma, and R. Kumar. 2021. 'Biofortification and Bioavailability of Zn, Fe and Se in Wheat: Present Status and Future Prospects'. *Theoretical and Applied Genetics* 134: 1–35.
- Gupta, Sanjay Mohan, Sandeep Arora, Neelofar Mirza, Anjali Pande, Charu Lata, Swati Puranik, J. Kumar, and Anil Kumar. 2017. 'Finger Millet: A "Certain" Crop for an "Uncertain" Future and a Solution to Food Insecurity and Hidden Hunger under Stressful Environments'. *Frontiers in Plant Science* 8. <https://www.frontiersin.org/articles/10.3389/fpls.2017.00643>.
- Hardigan, Michael A., F. Parker E. Laimbeer, Linsey Newton, Emily Crisovan, John P. Hamilton, Brieanne Vaillancourt, Krystle Wiegert-Rininger, et al. 2017. 'Genome Diversity of Tuber-Bearing Solanum Uncovers Complex Evolutionary History and Targets of Domestication in the Cultivated Potato'. *Proceedings of the National Academy of Sciences* 114 (46): E9999–10008. <https://doi.org/10.1073/pnas.1714380114>.
- Harrison, James, Karen A. Moore, Konrad Paszkiewicz, Thomas Jones, Murray R. Grant, Daniel Ambacheew, Sadik Muzemil, and David J. Studholme. 2014. 'A Draft Genome Sequence for *Ensete ventricosum*, the Drought-Tolerant "Tree Against Hunger"'. *Agronomy* 4 (1): 13–33. <https://doi.org/10.3390/agronomy4010013>.
- Harvest Plus, International Centre for Tropical Agriculture (CIAT), Cali, Colombia, and Meike Andersson. 2017. 'Progress Update: Crop Development of Biofortified Staple Food Crops under HarvestPlus'. *African Journal of Food, Agriculture, Nutrition and Development* 17 (02): 11905–35. <https://doi.org/10.18697/ajfand.78.HarvestPlus05>.
- Hassan, Z.M., N.A. Sebola, and M. Mabelebele. 2021. 'The Nutritional Use of Millet Grain for Food and Feed: A Review'. *Agric Food Secur* 10: 1–14.
- Hay, S.I. 2017. 'Global, Regional, and National Disability-Adjusted Life-Years (DALYs) for 333 Diseases and Injuries and Healthy Life Expectancy (HALE) for 195 Countries and Territories, 1990–2016: A Systematic Analysis for the Global Burden of Disease Study 2016'. *The Lancet* 390: 1260–1344.
- Headey, D. 2020. 'Impacts of COVID-19 on Childhood Malnutrition and Nutrition-Related Mortality'. *The Lancet* 396: 519–21.
- Hertel, Thomas, Ismahane Elouadi, Mchakot Tanticharoen, and Frank Ewert. 2021. 'Diversification for Enhanced Food Systems Resilience'. *Nature Food* 2 (11): 832–34. <https://doi.org/10.1038/s43016-021-00403-9>.
- Heuzé, V., G. Tran, and C.urger-Reverdin. 2015. 'Scrobic (*Paspalum Scrobiculatum*) Forage and Grain.' *Feedipedia, a Programme by INRAE, CIRAD, AFZ and FAO*. 6 October 2015. <https://www.feedipedia.org/node/401>.
- Higuchi, K. 1999. 'Cloning of Nicotianamine Synthase Genes, Novel Genes Involved in the Biosynthesis of Phytosiderophores'. *Plant Physiol* 119: 471–79.
- Hinojosa, Leonardo, Juan A. González, Felipe H. Barrios-Masias, Francisco Fuentes, and Kevin M. Murphy. 2018. 'Quinoa Abiotic Stress Responses: A Review'. *Plants* 7 (4): 106. <https://doi.org/10.3390/plants7040106>.
- Hittalmani, Shailaja, H. B. Mahesh, Meghana Deepak Shirke, Hanamareddy Biradar, Govindareddy Uday, Y. R. Aruna, H. C. Lohithaswa, and A. Mohanrao. 2017. 'Genome and Transcriptome Sequence of Finger Millet (*Eleusine Coracana* (L.) Gaertn.) Provides Insights into Drought Tolerance and Nutraceutical Properties'. *BMC Genomics* 18 (1): 465. <https://doi.org/10.1186/s12864-017-3850-z>.
- Hoidal, Natalie, Maria Díaz Gallardo, Sven-Erik Jacobsen, and Gabriela Alandia. 2019. 'Amaranth as a Dual-Use Crop for Leafy Greens and Seeds: Stable Responses to Leaf Harvest Across Genotypes and Environments'. *Frontiers in Plant Science* 10 (June): 817. <https://doi.org/10.3389/fpls.2019.00817>.

- Honfo, F.G., A. Tenkouano, and O. Coulibaly. 2011. 'Banana and Plantain-Based Foods Consumption by Children and Mothers in Cameroon and Southern Nigeria: A Comparative Study'. *African Journal of Food Science* 5: 287–291.
- Imdad, A., E. Mayo-Wilson, K. Herzer, and Z.A. Bhutta. 2017. 'Vitamin A Supplementation for Preventing Morbidity and Mortality in Children from Six Months to Five Years of Age'. *Cochrane Database of Systematic Reviews* 2017.
- Indurker, Shivani, Hari S. Misra, and Susan Eapen. 2007. 'Genetic Transformation of Chickpea (*Cicer Arietinum* L.) with Insecticidal Crystal Protein Gene Using Particle Gun Bombardment'. *Plant Cell Reports* 26 (6): 755–63. <https://doi.org/10.1007/s00299-006-0283-6>.
- Inoue, H. 2003. 'Three Rice Nicotianamine Synthase Genes, OsNAS1, OsNAS2, and OsNAS3 Are Expressed in Cells Involved in Long-Distance Transport of Iron and Differentially Regulated by Iron'. *Plant Journal* 36: 366–81.
- Institute of Medicine (U.S.) and Panel on Micronutrients. 2002. *DRI: Dietary Reference Intakes for Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium, and Zinc: A Report of the Panel on Micronutrients ... and the Standing Committee on the Scientific Evaluation of Dietary Reference Intakes, Food and Nutrition Board, Institute of Medicine*. Washington, D.C.: National Academy Press. <http://site.ebrary.com/id/10032471>.
- International Treaty on Plant Genetic Resources for Food and Agriculture and Food and Agriculture Organization of the United Nations. n.d. 'Characterization, Genetic Enhancement and Revitalization of Finger Millet in Western Kenya'. <https://www.fao.org/3/bb136e/bb136e.pdf>.
- Ivo, Nayche L., Cristina P. Nascimento, Livia S. Vieira, Francisco A. F. Campos, and Francisco J. L. Aragão. 2008. 'Biolytic-Mediated Genetic Transformation of Cowpea (*Vigna unguiculata*) and Stable Mendelian Inheritance of Transgenes'. *Plant Cell Reports* 27 (9): 1475–83. <https://doi.org/10.1007/s00299-008-0573-2>.
- Jain, S. Mohan, and P. M. Priyadarshan, eds. 2009. *Breeding Plantation Tree Crops: Tropical Species*. New York, NY: Springer New York. <https://doi.org/10.1007/978-0-387-71201-7>.
- Jamnadas, R. 2020. 'Enhancing African Orphan Crops with Genomics'. *Nat Genet* 52: 356–60.
- Jaramillo, A.M. 2022. 'Characterization of Casava ORANGE Proteins and Their Capability to Increase Provitamin A Carotenoids Accumulation'. *PLoS One* 17: 0252412.
- Jarvis, David E., Yung Shwen Ho, Damien J. Lightfoot, Sandra M. Schmöckel, Bo Li, Theo J. A. Borm, Hajime Ohyanagi, et al. 2017. 'The Genome of *Chenopodium Quinoa*'. *Nature* 542 (7641): 307–12. <https://doi.org/10.1038/nature21370>.
- Jha, N., R. Krishnan, and M.S. Meera. 2015. 'Effect of Different Soaking Conditions on Inhibitory Factors and Bioaccessibility of Iron and Zinc in Pearl Millet'. *J Cereal Sci* 66: 46–52.
- Johnson, Matthew, Santosh Deshpande, Mani Vetriventhan, Hari D. Upadhyaya, and Jason G. Wallace. 2019. 'Genome-Wide Population Structure Analyses of Three Minor Millets: Kodo Millet, Little Millet, and Proso Millet'. *The Plant Genome* 12 (3): 1–9. <https://doi.org/10.3835/plantgenome2019.03.0021>.
- Kamenya, Sandra Ndagire, Erick Owuor Mikwa, Bo Song, and Damaris Achieng Odeny. 2021. 'Genetics and Breeding for Climate Change in Orphan Crops'. *Theoretical and Applied Genetics* 134 (6): 1787–1815. <https://doi.org/10.1007/s00122-020-03755-1>.
- Kang, Minjeong, Keunsub Lee, Todd Finley, Hal Chappell, Veena Veena, and Kan Wang. 2022. 'An Improved Agrobacterium-Mediated Transformation and Genome-Editing Method for Maize Inbred B104 Using a Ternary Vector System and Immature Embryos'. *Frontiers in Plant Science* 13. <https://www.frontiersin.org/articles/10.3389/fpls.2022.860971>.
- Karmakar, A. 2020. 'RNAi-Mediated Silencing of ITPK Gene Reduces Phytic Acid Content, Alters Transcripts of Phytic Acid Biosynthetic Genes, and Modulates Mineral Distribution in Rice Seeds'. *Rice Sci* 27: 315–28.
- Karmakar, Subhasis, Kutubuddin Ali Molla, Dipak Gayen, Aritra Karmakar, Kaushik Das, Sailendra Nath Sarkar, Karabi Datta, and Swapan K. Datta. 2019. 'Development of a Rapid and Highly Efficient Agrobacterium-Mediated

Transformation System for Pigeon Pea [*Cajanus cajan* (L.) Millsp.]. *GM Crops & Food* 10 (2): 115–38. <https://doi.org/10.1080/21645698.2019.1625653>.

Kaur, A. 2017. 'Genetic Transformation of Pigeon pea Through Particle Gun And Agrobacterium Using Cry 1 Ac Transgene'. In <https://www.semanticscholar.org/paper/Genetic-Transformation-Of-Pigeonpea-Through-Gun-And-Kaur/2459eda98d65d65d838287d6b34b89ab4c0f5f35>.

Kaur, N. 2017. 'Regulation of Banana Phytoene Synthase (MaPSY) Expression, Characterization and Their Modulation under Various Abiotic Stress Conditions'. *Front Plant Sci* 8.

Kawuki, R. S., L. Herselman, M. T. Labuschagne, I. Nzuki, I. Ralimanana, M. Bidiaka, M. C. Kanyange, et al. 2013. 'Genetic Diversity of Cassava (*Manihot Esculenta* Crantz) Landraces and Cultivars from Southern, Eastern and Central Africa'. *Plant Genetic Resources* 11 (2): 170–81. <https://doi.org/10.1017/S1479262113000014>.

Keats, E.C., L.M. Neufeld, G.S. Garrett, M.N.N. Mbuya, and Z.A. Bhutta. 2019. 'Improved Micronutrient Status and Health Outcomes in Low-and Middle-Income Countries Following Large-Scale Fortification: Evidence from a Systematic Review and Meta-Analysis'. *American Journal of Clinical Nutrition* 109: 1696–1708.

Khan, Md Mahmudul Hasan, Mohd Y. Rafii, Shairul Izan Ramlee, Mash'arun Yusoh, and Md Al Mamun. 2021. 'Genetic Analysis and Selection of Bambara Groundnut (*Vigna Subterranea* (L.) Verdc.) Landraces for High Yield Revealed by Qualitative and Quantitative Traits'. *Scientific Reports* 11 (1): 7597. <https://doi.org/10.1038/s41598-021-87039-8>.

Kim, So-Eun, Chan-Ju Lee, Sul-U Park, Ye-Hoon Lim, Woo Sung Park, Hye-Jin Kim, Mi-Jeong Ahn, Sang-Soo Kwak, and Ho Soo Kim. 2021. 'Overexpression of the Golden SNF Carrying Orange Gene Enhances Carotenoid Accumulation and Heat Stress Tolerance in Sweet potato Plants'. *Antioxidants* 10 (1): 51. <https://doi.org/10.3390/antiox10010051>.

Kojima, M., Y. Arai, N. Iwase, K. Shirotori, H. Shioiri, and M. Nozue. 2000. 'Development of a Simple and Efficient Method for Transformation of Buckwheat Plants (*Fagopyrum Esculentum*) Using Agrobacterium Tumefaciens'. *Bioscience, Biotechnology, and Biochemistry* 64 (4): 845–47. <https://doi.org/10.1271/bbb.64.845>.

Kozicka, M. 2021. 'Reassessing the Cost-Effectiveness of High-Provitamin A Bananas to Reduce Vitamin A Deficiency in Uganda'. *Front Sustain Food Sys* 5: 61.

Krishnamurthy, L., Hari D. Upadhyaya, J. Kishorwagi, R. Purushothaman, Sangam L. Dwivedi, and V. Vadez. 2016. 'Variation in Drought-Tolerance Components and Their Interrelationships in the Minicore Collection of Finger Millet Germplasm'. *Crop Science* 56 (1): 1914–26. <https://doi.org/10.2135/cropsci2016.03.0191>.

Krishnan, Rateesh, and M. S. Meera. 2018. 'Pearl Millet Minerals: Effect of Processing on Bioaccessibility'. *Journal of Food Science and Technology* 55 (2): 3362–72. <https://doi.org/10.1007/s13197-018-3305-9>.

Kumar, G.B.S., L. Srinivas, and V.R. Ganapathi. 2011. 'Iron Fortification of Banana by the Expression of Soybean Ferritin'. *Biol Trace Elem Res* 132: 232–41.

Kumar, Neha, Jody Harris, and Rahul Rawat. 2015. 'If They Grow It, Will They Eat and Grow? Evidence from Zambia on Agricultural Diversity and Child Undernutrition'. *The Journal of Development Studies* 51 (8): 1060–77. <https://doi.org/10.1080/00220388.2015.1018901>.

Kummu, Matti, Matias Heino, Maija Taka, Olli Varis, and Daniel Viviroli. 2021. 'Climate Change Risks Pushing One-Third of Global Food Production Outside the Safe Climatic Space'. *One Earth* 4 (5): 720–29. <https://doi.org/10.1016/j.oneear.2021.04.017>.

Kundy, Aloyce, Omary Mponda, Charles Mkandawile, and Geoffrey Mkamilo. 2015. 'Yield Evaluation of Eighteen Pigeon Pea (*Cajanus cajan* (L.) Millsp.) Genotypes in Southeastern Tanzania'. *European Journal of Physical and Agricultural Sciences* 3 (January): 9–15.

Kusvuran, Sebnem. 2012. 'Influence of Drought Stress on Growth, Ion Accumulation and Antioxidative Enzymes in Okra Genotypes'. *International Journal of Agriculture and Biology* 14 (January): 401–6.

- Kuwano, M., T. Mimura, F. Takaiwa, and K.T. Yoshida. 2009. 'Generation of Stable "Low Phytic Acid" Transgenic Rice through Antisense Repression of the 1D-Myo-Inositol 3-Phosphate Synthase Gene (RINO1) Using the 18-KDa Oleosin Promoter'. *Plant Biotechnology Journal* 7: 96–105.
- Kyei-Boahen, Stephen, Canon E. N. Savala, David Chikoye, and Robert Abaidoo. 2017. 'Growth and Yield Responses of Cowpea to Inoculation and Phosphorus Fertilization in Different Environments'. *Frontiers in Plant Science* 8 (May): 646. <https://doi.org/10.3389/fpls.2017.00646>.
- Lambein, Fernand, Silvia Travella, Yu-Haey Kuo, Marc Van Montagu, and Marc Heijde. 2019. 'Grass Pea (*Lathyrus Sativus* L.): Orphan Crop, Nutraceutical or Just Plain Food?' *Planta* 250 (3): 821–38. <https://doi.org/10.1007/s00425-018-03084-0>.
- Li, Haobing, Matthew Rodda, Annathurai Gnanasambandam, Mohammad Aftab, Robert Redden, Kristy Hobson, Garry Rosewarne, Michael Materne, Sukhjiwan Kaur, and Anthony T. Slater. 2015. 'Breeding for Biotic Stress Resistance in Chickpea: Progress and Prospects'. *Euphytica* 204 (2): 257–88. <https://doi.org/10.1007/s10681-015-1462-8>.
- Li, L. 2012. 'The *OR* Gene Enhances Carotenoid Accumulation and Stability during Post-Harvest Storage of Potato Tubers'. *Molecular Plant* 5: 339–52.
- Li, Xuan, Rashmi Yadav, and Kadambot H. M. Siddique. 2020. 'Neglected and Underutilized Crop Species: The Key to Improving Dietary Diversity and Fighting Hunger and Malnutrition in Asia and the Pacific'. *Frontiers in Nutrition* 7. <https://www.frontiersin.org/article/10.3389/fnut.2020.003711>.
- Liang, Q. 2019. 'Improved Folate Accumulation in Genetically Modified Maize and Wheat'. *J Exp Bot* 70: 1539–51.
- Liu, Guoquan, Karen Massel, Basam Tabet, and Ian D. Gowen. 2020. 'Biolytic DNA Delivery and Its Applications in *Sorghum Bicolor*'. *Methods in Molecular Biology (Clifton, N.J.)* 2124: 197–215. https://doi.org/10.1007/978-1-0716-0356-7_10.
- Lonardi, Stefano, María Muñoz-Amatriaín, Qihua Liang, Shengqiang Shu, Steve I. Wanamaker, Sassoum Lo, Jaakko Tanskanen, et al. 2019. 'The Genome of Cowpea (*Vigna Unguiculata* [L.] Walp.)'. *The Plant Journal* 98 (5): 767–82. <https://doi.org/10.1111/tbj.14349>
- Lopez, A. B., J. Van Eck, B. J. Conlin, D. J. Paulillo, J. O'Neill, and L. Li. 2008. 'Effect of the Cauliflower *OR* Transgene on Carotenoid Accumulation and Chromoplast Formation in Transgenic Potato Tubers'. *Journal of Experimental Botany* 59 (2): 213–23. <https://doi.org/10.1093/jxb/erm299>.
- López-Marqués, Rosa L., Anton F. Nørrevang, Peter Ache, Max Moog, Davide Visintainer, Toni Wendt, Jeppe T. Østerberg, et al. 2020. 'Prospects for the Accelerated Improvement of the Resilient Crop Quinoa'. *Journal of Experimental Botany* 71 (18): 5333–47. <https://doi.org/10.1093/jxb/eraa285>.
- Low, J.W., and G. Thiele. 2021. 'Understanding Innovation: The Development and Scaling of Orange-Fleshed Sweet potato in Major African Food Systems'. *Agric Syst* 179: 102770.
- Lowe, N.M. 2021. 'The Global Challenge of Hidden Hunger: Perspectives from the Field'. In *Proceedings of the Nutrition Society*, 80:283–89. Cambridge University Press.
- Mabhaudhi, T. 2019. 'Prospects of Orphan Crops in Climate Change'. *Planta* 250: 695–708.
- Mabhaudhi, Tafadzwanashe, Vimbayi Grace Petrova Chimonyo, Sithabile Hlahla, Festo Massawe, Sean Mayes, Luxon Nhamo, and Albert Thembinkosi Modi. 2019. 'Prospects of Orphan Crops in Climate Change'. *Planta* 250 (3): 695–708. <https://doi.org/10.1007/s00425-019-03129-y>.
- Mafakheri, Khosro, Mohammad Reza Bihamta, and Ali Reza Abbasi. 2017. 'Assessment of Genetic Diversity in Cowpea (*Vigna Unguiculata* L.) Germplasm Using Morphological and Molecular Characterisation'. Edited by Manuel Tejada Moral. *Cogent Food & Agriculture* 3 (1): 1327092. <https://doi.org/10.1080/23311932.2017.1327092>.

- Maharaj, Prayna P. P., Surendra Prasad, Riteshma Devi, and Romila Gopalan. 2015. 'Folate Content and Retention in Commonly Consumed Vegetables in the South Pacific'. *Food Chemistry* 182 (September): 327–32. <https://doi.org/10.1016/j.foodchem.2015.02.096>.
- Majola, Nomathemba Gloria, Abe Shegro Gerrano, and Hussein Shimelis. 2021. 'Bambara Groundnut (*Vigna Subterranea* [L.] Verdc.) Production, Utilisation and Genetic Improvement in Sub-Saharan Africa'. *Agronomy* 11 (7): 1345. <https://doi.org/10.3390/agronomy11071345>.
- Majola, Nomathemba Gloria, Abe Shegro Gerrano, and Hussein Shimelis. 2021. 'Bambara Groundnut (*Vigna Subterranea* [L.] Verdc.) Production, Utilisation and Genetic Improvement in Sub-Saharan Africa'. *Agronomy* 11 (7): 1345. <https://doi.org/10.3390/agronomy11071345>.
- Manyong, Victor M., Ranajit Bandyopadhyay, B. B. Maziya-Dixon, and R. Djouaka. 2012. 'The Agriculture and Health Program of the International Institute of Tropical Agriculture (IITA), a CGIAR Institution in Africa'. In <https://cgspace.cgiar.org/handle/10568/97286>.
- Maqbool, M.A., and A.R. Beshir. 2019. 'Zinc Biofortification of Maize (*Zea Mays* L.): Status and Challenges'. *Plant Breeding* 138: 1–28.
- Matheka, Jonathan, Jaindra Nath Tripathi, Ibsa Merga, Endale Gebre, and Leona Tripathi. 2019. 'A Simple and Rapid Protocol for the Genetic Transformation of *Ensete Ventrerosum*'. *Plant Methods* 15: 130. <https://doi.org/10.1186/s13007-019-0512-y>.
- Mbégué-A-Mbégué, D. 2009. 'Expression Patterns of Cell Wall-Modifying Genes from Banana during Fruit Ripening and in Relationship with Finger Drop'. *J Exp Bot* 60: 2021–24.
- Mbinda, Wilton, and Hosea Masaki. 2021. 'Breeding Strategies and Challenges in the Improvement of Blast Disease Resistance in Finger Millet. A Current Review'. *Frontiers in Plant Science* 11. <https://www.frontiersin.org/articles/10.3389/fpls.2020.602882>.
- Mccabe, J. 1998. 'The Tree Against Hunger: Enset Cultivation in Ethiopia'.
- McKevith, Brigid. 2004. 'Nutritional Aspects of Cereals'. *Nutrition Bulletin* 29 (2): 111–42. <https://doi.org/10.1111/j.1467-3010.2004.00412.x>.
- McMullin, S. 2021. 'Determining Appropriate Interventions to Mainstream Nutritious Orphan Crops into African Food Systems'. *Glob Food Sec* 28: 100435.
- Mensi, A., and C.C. Udenigwe. 2021. 'Emerging and Practical Food Innovations for Achieving the Sustainable Development Goals (SDG) Target 2.2'. *Trends Food Sci Technol* 111: 783–89.
- Miller, Kyle, Alan L. Eggenberger, Keunsub Lee, Fei Liu, Minjeong Kang, Madison Drent, Andrew Ruba, Tyler Kirscht, Kan Wang, and Simon Jiang. 2021. 'An Improved Biolistic Delivery and Analysis Method for Evaluation of DNA and CRISPR-Cas Delivery Efficacy in Plant Tissue'. *Scientific Reports* 11 (1): 7695. <https://doi.org/10.1038/s41598-021-86549-9>.
- Minten, Bart, Ermias Engida, and Seneshaw Tamru. 2016. 'How Big Are Post-Harvest Losses? Evidence from Teff'. Project Paper. ESSP II Research Note. Washington, D.C.: International Food Policy Research Institute (IFPRI) and Ethiopian Development Research Institute (EDRI). <https://www.ifpri.org/publication/synopsis-how-big-are-post-harvest-losses-evidence-teff>.
- Mishra, Gyan P., Bijendra Singh, Tania Seth, Achuit K. Singh, Jaydeep Halder, Nagendran Krishnan, Shailesh K. Tiwari, and Prabhakar M. Singh. 2017. 'Biotechnological Advancements and Begomovirus Management in Okra (*Abelmoschus Esculentus* L.): Status and Perspectives'. *Frontiers in Plant Science* 8. <https://www.frontiersin.org/articles/10.3389/fpls.2017.00360>.
- Molla, Mohammad, Kamrul Islam, Shaikh Uddin, and M.S Mawla. 2011. 'Assessment of Postharvest Practises and Losses of Cereal Crops in Selected Areas of Bangladesh'. *Eco-Friendly Agriculture Journal* 4 (September): 687–92.

- Morris, Kwami Justina, Nitty Hirawaty Kamarulzaman, and Kenobi Isima Morris. 2019. 'Small-Scale Postharvest Practices among Plantain Farmers and Traders: A Potential for Reducing Losses in Rivers State, Nigeria'. *Scientific African* 4 (July): e00086. <https://doi.org/10.1016/j.sciaf.2019.e00086>.
- Moura, F.F. 2015. 'Cassava intake and Vitamin A status among women and preschool children in Akwa- Ibom, Nigeria'. *PLoS One* 10.
- Mula, M. G., and K. B. Saxena. 2010. Lifting the Level of Awareness on Pigeon pea - A Global Perspective. Patancheru: International Crops Research Institute for the Semi-Arid Tropics. <http://oar.icrisat.org/193/>.
- Muthayya, S. 2013. 'The Global Hidden Hunger Indices and Maps: An Advocacy Tool for Action'. *PLoS One* 8: 67860.
- Nair, Madhavan K., Little Flower Augustine, and Archana Konapur. 2016. 'Food-Based Interventions to Modify Diet Quality and Diversity to Address Multiple Micronutrient Deficiency'. *Frontiers in Public Health* 3 (January). <https://doi.org/10.3389/fpubh.2015.00277>.
- Nansamba, Moureen, Julia Sibiya, Robooni Tumuhimbise, Deborah Karamura, Jerome Kubiriba, and Eldad Karamura. 2020. 'Breeding Banana (*Musa Spp.*) for Drought Tolerance: A Review'. *Plant Breeding* 139 (4): 685–96. <https://doi.org/10.1111/pbr.12812>.
- Naqvi, S. 2009. 'Transgenic Multivitamin Corn through Biofortification of Endosperm with Three Vitamins Representing Three Distinct Metabolic Pathways'. *Proc Natl Acad Sci U S A* 106: 7762–67.
- Narayanan, Narayanan, Getu Beyene, Raj Deepika Chauhan, Liliana Gaitán-Solís, Jackson Gehan, Paula Butts, Dimuth Siritunga, et al. 2019. 'Biofortification of Field-Grown Cassava by Engineering Expression of an Iron Transporter and Ferritin'. *Nature Biotechnology* 37 (2): 147–51. <https://doi.org/10.1038/s41587-018-0002-1>.
- Narendran, M., Satish G. Deole, Satish Harkude, Dattatraya Simale, Asaram Nanote, Pankaj Bihani, Srinivas Parimi, Bharat R. Char, and Usha B. Zehr. 2013. 'Efficient Genetic Transformation of Okra (*Abelmoschus Esculentus* (L.) Moench) and Generation of Insect-Resistant Transgenic Plants Expressing the Cry1Ac Gene'. *Plant Cell Reports* 32 (8): 1191–98. <https://doi.org/10.1007/s00299-013-1415-4>.
- Nascimento, Cristina P., Thaís M. Cipriano, and Francisco J. L. Aragão. 2022. 'Natural Variation of Folate Content in Cowpea (*Vigna Unguiculata*) Germplasm and Its Correlation with the Expression of the GTP Cyclohydrolase I Coding Gene'. *Journal of Food Composition and Analysis* 107 (April): 104357. <https://doi.org/10.1016/j.jfca.2021.104357>.
- Ningsih, Rita, and Rita Megia. 2019. 'Folic Acid Content and Fruit Characteristics of Five Indonesian Dessert Banana Cultivars'. *Biodiversitas: Journal of Biological Diversity* 20 (1): 144–51. <https://doi.org/10.13057/biodiv/d200117>.
- Nnamani, C. V., D. B. Adewale, O. J. Oselebe, and C. J. Atkinson. 2021. 'African Yam Bean the Choice for Climate Change Resilience: Need for Conservation and Policy'. In *African Handbook of Climate Change Adaptation*, edited by Nicholas Oguege, Desalegn Ayal, Lydia Adeleke, and Izael da Silva, 453–69. Cham: Springer International Publishing. https://doi.org/10.1007/978-3-030-45106-6_203.
- Norgrove, L., and S. Hauser. 2014. 'Improving Plantain (*Musa Spp.* AAB) Yields on Smallholder Farms in West and Central Africa'. *Food Security* 6: 501–14.
- Noyer, J. L., S. Causse, K. Tomekpe, A. Bouet, and F. C. Baurens. 2005. 'A New Image of Plantain Diversity Assessed by SSR, AFLP and MSAP Markers'. *Genetica* 124 (1): 61–69. <https://doi.org/10.1007/s10709-004-7319-z>.
- Nozoye, T. 2018. 'The Nicotianamine Synthase Gene Is a Useful Candidate for Improving the Nutritional Qualities and Fe-Deficiency Tolerance of Various Crops'. *Frontiers in Plant Science* 9: 340.
- Ntui, Valentine, Edak Uyoh, Ikuo Nakamura, and Masahiro Mii. 2017. 'Agrobacterium-Mediated Genetic Transformation of Fonio (*Digitaria Exilis* (L.) Stapf)'. *African Journal of Biotechnology* 16 (June): 1302–7. <https://doi.org/10.5897/AJB2017.15903>.
- Nunes, A.C.S., D.C. Kalkmann, and F.J.L. Aragão. 2009. 'Folate Biofortification of Lettuce by Expression of a Codon Optimized Chicken GTP Cyclohydrolase I Gene'. *Transgenic Research* 18: 661–67.

Nyambo, B. T. 1993. 'Post-harvest Maize and Sorghum Grain Losses in Traditional and Improved Stores in South Nyanza District, Kenya'. *International Journal of Pest Management* 39 (2): 181–87. <https://doi.org/10.1080/09670879309371787>.

O'Kennedy, Martha M., Hester C. Stark, and Nosisa Dube. 2011. 'Biostic-Mediated Transformation Protocols for Maize and Pearl Millet Using Pre-Cultured Immature Zygotic Embryos and Embryogenic Tissue'. *Methods in Molecular Biology (Clifton, N.J.)* 710: 343–54. https://doi.org/10.1007/978-1-61737-988-8_23.

Olango, Temesgen Magule, Bizuayehu Tesfaye, Mario Augusto Pagnotta, Mario Enrico Pè, and Marcello Catellani. 2015. 'Development of SSR Markers and Genetic Diversity Analysis in Enset (*Ensete Ventricosum* (Welw.) Cheesman), an Orphan Food Security Crop from Southern Ethiopia'. *BMC Genetics* 16 (1): 98. <https://doi.org/10.1186/s12863-015-0250-8>.

Olayemi, F. F., J. A. Adegbola, E. I. Bamishaiye, and E. F. Awagu. 2012. 'Assessment of Post Harvest Losses of Some Selected Crops in Eight Local Government Areas of Rivers State, Nigeria'. *Asian Journal of Rural Development* 2 (1): 13–23. <https://doi.org/10.3923/ajrd.2012.13.23>.

Opabode, Jelili. 2019. 'Assessment of Growth, Lipid Peroxidation and Reactive Oxygen Species Scavenging Capacity of Ten Elite Cassava Cultivars Subjected to Heat Stress'. *Agricultural Research & Technology: Open Access Journal* 21 (April). <https://doi.org/10.19080/ARTOAJ.2019.21.5561.1>

Osendarp, S. 2021. 'The COVID-19 Crisis Will Exacerbate Maternal and Child Undernutrition and Child Mortality in Low- and Middle-Income Countries'. *Nature Food* 2: 476–84.

Osorio, Claudia E. 2019. 'The Role of Orange Gene in Carotenoid Accumulation: Manipulating Chromoplasts Toward a Colored Future'. *Frontiers in Plant Science* 10 (October): 1235. <https://doi.org/10.3389/fpls.2019.01235>.

Ou, Wenjun, Xiang Mao, Chao Huang, Weiwei Tie, Yan Chen, Zehong Ding, Chunlai Wu, et al. 2018. 'Genome-Wide Identification and Expression Analysis of the KIP Family under Abiotic Stress in Cassava (*Manihot Esculenta* Crantz)'. *Frontiers in Physiology* 9: 17. <https://doi.org/10.3389/fphys.2018.00017>.

Paine, J.A. 2005. 'Improving the Nutritional Value of Golden Rice through Increased Pro-Vitamin A Content'. *Nature Biotechnology* 23: 482–87.

Pal, Saheb, and Solanki Bal. 2018. 'Postharvest Management, Processing and Value Addition of Okra (*Abelmoschus Esculentus* (L.) Moench)'. In *Advances in Post Harvest Management, Processing and Value Addition of Horticultural Crops - Part 2: Vegetables, Spices & Plantation Crops*, 159–80. Today and Tomorrow's Printers and Publishers.

Panda, Debabrata, N. Hema Sathia, Prafulla K. Behera, Kartik Lenka, Shyam S. Sharma, and Sangram K. Lenka. 2021. 'Genetic Diversity of Under Utilized Indigenous Finger Millet Genotypes from Koraput, India for Crop Improvement'. *Journal of Plant Biochemistry and Biotechnology* 30 (1): 99–116. <https://doi.org/10.1007/s13562-020-00557-w>.

Paterson, Andrew H., John E. Bowers, Rémy Bruggmann, Inna Dubchak, Jane Grimwood, Heidrun Gundlach, Georg Haberer, et al. 2009. 'The Sorghum Bicolor Genome and the Diversification of Grasses'. *Nature* 457 (7229): 551–56. <https://doi.org/10.1038/nature07723>.

Paul, J.Y. 2017. 'Golden Bananas in the Field: Elevated Fruit pro-Vitamin A from the Expression of a Single Banana Transgene'. *Plant Biotechnology Journal* 15: 520–32.

Peláez, Pablo, Domancar Orona-Tamayo, Salvador Montes-Hernández, María Elena Valverde, Octavio Paredes-López, and Angélica Cibrián-Jaramillo. 2019. 'Comparative Transcriptome Analysis of Cultivated and Wild Seeds of *Salvia Hispanica* (Chia)'. *Scientific Reports* 9 (1): 9761. <https://doi.org/10.1038/s41598-019-45895-5>.

Pénicaud, C., N. Achir, C. Dhuique-Mayer, M. Dornier, and P. Bohuon. 2011. 'Degradation of β -Carotene during Fruit and Vegetable Processing or Storage: Reaction Mechanisms and Kinetic Aspects: A Review'. *Fruits* 66: 417–40.

Perera, I., S. Seneweera, and N. Hirotsu. 2018. 'Manipulating the Phytic Acid Content of Rice Grain Toward Improving Micronutrient Bioavailability'. *Rice* 11.

- Pinstrup-Andersen, Per. 2007. 'Agricultural Research and Policy for Better Health and Nutrition in Developing Countries: A Food Systems Approach: Agricultural Research and Policy for Better Health and Nutrition in Developing Countries'. *Agricultural Economics* 37 (December): 187–98. <https://doi.org/10.1111/j.1574-0862.2007.00244.x>.
- Punjabi, M., N. Bharadvaja, M. Jolly, A. Dahuja, and A. Sachdev. 2018. 'Development and Evaluation of Low Phytic Acid Soybean by siRNA Triggered Seed Specific Silencing of Inositol Polyphosphate 6-/3-/5-Kinase Gene'. *Front Plant Sci* 9.
- Puthusseri, B. 2018. 'Novel Folate Binding Protein in Arabidopsis Expressed during Salicylic Acid-Induced Folate Accumulation'. *J Agric Food Chem* 66: 505–11.
- Raboy, V. 2020. 'Low Phytic Acid Crops: Observations Based on Four Decades of Research'. *Plants* 9: 140.
- Rai, K. 2013. 'Breeding Pearl Millet Cultivars for High Iron Density with Zinc Density as an Associated Trait'. *Journal of SAT Agricultural Research* 11: 1–7.
- Rai, K.N., M. Govindaraj, and A.S. Rao. 2012. 'Genetic Enhancement of Grain Iron and Zinc Content in Pearl Millet'. *Quality Assurance and Safety of Crops and Foods* 4: 119–25.
- Ramadevi, R., K. V. Rao, and V. D. Reddy. 2014. 'Agrobacterium Tumefaciens-Mediated Genetic Transformation and Production of Stable Transgenic Pearl Millet (*Pennisetum Glaucum* [L.] R. Br.)'. *In Vitro Cellular & Developmental Biology - Plant* 50 (4): 392–400. <https://doi.org/10.1007/s11627-013-9592-y>.
- Rani, Anju, Poonam Devi, Uday Chand Jha, Kamal Dev Sharma, Kacambot H. M. Siddique, and Harsh Nayyar. 2020. 'Developing Climate-Resilient Chickpea Involving Physiological and Molecular Approaches with a Focus on Temperature and Drought Stresses'. *Frontiers in Plant Science* 10. <https://www.frontiersin.org/articles/10.3389/fpls.2019.01159>.
- Rao, D. 2016. 'Micronutrient Deficiencies in the Developing World: An Evaluation of Delivery Methods'. In *GHTC 2016 - IEEE Global Humanitarian Technology Conference: Technology for the Benefit of Humanity, Conference Proceedings* 597–604. <https://doi.org/10.1109/GHTC.2016.7857340>.
- Rautiainen, S., J.E. Manson, A.H. Lichtenstein, and H.D. Sesso. 2016. 'Dietary Supplements and Disease Prevention—a Global Overview'. *Nat Rev Endocrinol* 12: 407–20.
- Reddy, C.S., S.-C. Kim, and T. Kaul. 2017. 'Genetically Modified Phytase Crops Role in Sustainable Plant and Animal Nutrition and Ecological Development: A Review'.
- Reddy, Manju B., and Mark Love. 1999. 'The Impact of Food Processing on the Nutritional Quality of Vitamins and Minerals'. In *Impact of Processing on Food Safety*, edited by Lauren S. Jackson, Mark G. Knize, and Jeffrey N. Morgan, 99–106. *Advances in Experimental Medicine and Biology*. Boston, MA: Springer US. https://doi.org/10.1007/978-1-4615-4853-9_7.
- Research Institute (IFPRI), International Food Policy. 2014. '2014 Global Hunger Index The Challenge of Hidden Hunger'. 0 ed. Washington, DC: International Food Policy Research Institute. <https://doi.org/10.2499/9780896299580>.
- Rezaei, M.K., A. Deokar, and B. Tar'an. 2016. 'Identification and expression analysis of candidate genes involved in carotenoid biosynthesis in chickpea seeds'. *Front Plant Sci* 7: 1867.
- Riggins, Chance W., Ana Paulina Barba de la Rosa, Matthew W. Blair, and Eduardo Espitia-Rangel. 2021. 'Editorial: Amaranthus: Naturally Stress-Resistant Resources for Improved Agriculture and Human Health'. *Frontiers in Plant Science* 12. <https://www.frontiersin.org/articles/10.3389/fpls.2021.726875>.
- Rivero, R.M., R. Mittler, E. Blumwald, and S.I. Zandalinas. 2022. 'Developing Climate-Resilient Crops: Improving Plant Tolerance to Stress Combination'. *The Plant Journal* 109: 373–89.
- Rodriguez, Juan Pablo, Sven-Erik Jacobsen, Christian Andreasen, and Marten Sørensen. 2020. 'Cañahua (*Chenopodium Pallidicaule*): A Promising New Crop for Arid Areas'. In *Emerging Research in Alternative Crops*, edited by Abdelaziz Hirich, Redouane Choukr-Allah, and Ragab Ragab, 221–43. *Environment & Policy*. Cham: Springer International Publishing. https://doi.org/10.1007/978-3-319-90472-6_9.

- Rodriguez-leyva, D., and G.N. Pierce. 2021. 'The Impact of Nutrition on the COVID-19 Pandemic and the Impact of the COVID-19 Pandemic on Nutrition'. *Nutrients* 13.
- Rogers, Lisa M., Amy M. Cordero, Christine M. Pfeiffer, Dorothy B. Hausman, Becky L. Tsang, Luz María De-Regil, Jorge Rosenthal, et al. 2018. 'Global Folate Status in Women of Reproductive Age: A Systematic Review with Emphasis on Methodological Issues: Folate Status in Women and Methodological Issues'. *Annals of the New York Academy of Sciences* 1431 (1): 35–57. <https://doi.org/10.1111/nyas.13963>.
- Ruel-Bergeron, J.C. 2015. 'Global update and trends of hidden hunger, 1995-2011: The hidden hunger Index'. *PLoS One* 10: 0143497.
- Sági, László, Bart Panis, Serge Remy, Hilde Schoofs, Kris De Smet, Rony Swennen, and Bruno P. A. Cammue. 1995. 'Genetic Transformation of Banana and Plantain (*Musa* Spp.) via Particle Bombardment'. *Bio/Technology* 13 (5): 481–85. <https://doi.org/10.1038/nbt0595-481>.
- Saini, D.K., P. Devi, and P. Kaushik. 2020. 'Advances in Genomic Interventions for Wheat Biofortification: A Review'. *Agronomy* 10, Page 62 10: 62.
- Sammour, Reda, Mohammed Mira, Safa Radwan, and Salwa Fahmey. 2020. 'Genetic Diversity and Phylogenetic Relationships among and within *Amaranthus* Spp. Using RAPD Markers'. *Revista Mexicana de Biodiversidad* 91 (October): 913254. <https://doi.org/10.22201/ib.20078706e.2020.91.1.254>
- Sandhya, M., J. V. Ramana, D. Ratna Babu, V. Padma, and K. Vijaya Gopal. 2020. 'Evaluation of Foxtail Millet [*Setaria Italica* (L.) Beauv.] Germplasm for Lysine Content'. *International Journal of Current Microbiology and Applied Sciences* 9 (11): 1910–15. <https://doi.org/10.20546/ijcmias.2020.911.226>.
- Sanjana Reddy, P., C. Tara Satyavathi, Vikas Khandelwal, M. T. Patil, P. C. Gupta, L. D. Sharma, K. D. Mungra, et al. 2021. 'Performance and Stability of Pearl Millet Varieties for Grain Yield and Micronutrients in Arid and Semi-Arid Regions of India'. *Frontiers in Plant Science* 12 (March): 670201. <https://doi.org/10.3389/fpls.2021.670201>.
- Saxena, Kul Bhushan, Arbind K. Choudhary, Rajat J. Saxena, and Rajeev K. Varshney. 2018. 'Breeding Pigeon pea Cultivars for Intercropping: Synthesis and Strategies'. *Breeding Science* 68 (2): 159–67. <https://doi.org/10.1270/jsbbs.17105>.
- Schaub, P. 2012. 'On the Structure and Function of the Phytoene Desaturase CRTI from *Pantoea Ananatis*, a Membrane-Peripheral and FAD-Dependent Condase/Isomerase'. *PLoS One* 7.
- Schmidt, M.A. 2015. 'Transgenic Soybean Seeds Accumulating β -Carotene Exhibit the Collateral Enhancements of Oleate and Protein Content Traits'. *Plant Biotechnology Journal* 13: 590–600.
- Serba, Desalegn D., and Rattan S. Yadav. 2016. 'Genomic Tools in Pearl Millet Breeding for Drought Tolerance: Status and Prospects'. *Frontiers in Plant Science* 7. <https://www.frontiersin.org/articles/10.3389/fpls.2016.01724>.
- Shah, Deepak. 2014. 'Pre and Post-Harvest Losses of Pigeon Pea in Maharashtra: An Empirical Assessment'. *Indian Journal of Economics and Development* Vol. 10 (March): 1–9. <https://doi.org/10.5958/j.2322-0430.10.1.001>.
- Shekara, B G, P Mahadevu, N M Chikkarugi, and N Manasa. 2019. 'Response of Pearl Millet (*Pennisetum Glaucum* L.) Varieties to Nitrogen Levels for Higher Green Forage Yield and Quality in Southern Dry Zone of Karnataka'. *Forage Research* 45 (3): 232–34.
- Shukla, A., Nidhi Srivastava, Poonam Suneja, Satish Yadav, Zakir Hussain, J.C. Rana, and Shiv Yadav. 2018. 'Genetic Diversity Analysis in Buckwheat Germplasm for Nutritional Traits'. *Indian Journal of Experimental Biology* 56 (November): 827–37.
- Shukla, P.R., J. Skea, E. Calvo Buendia, V. Masson-Delmotte, H. O. Portner, D.C. Roberts, P. Zhai, et al. 2019. 'IPCC, 2019: Climate Change and Land: An IPCC Special Report on Climate Change, Desertification, Land Degradation, Sustainable Land Management, Food Security, and Greenhouse Gas Fluxes in Terrestrial Ecosystems'. <https://www.ipcc.ch/srccl/>.

- Singh, B., Aakansha Goswami, and Vaishali Shami. 2017. 'Study of Genetic Diversity in Okra [*Abelmoschus Esculentus* (L.) Moench]'. *Vegetos- An International Journal of Plant Research* 30 (June): 109. <https://doi.org/10.5958/2229-4473.2017.00044.1>.
- Singh, Baljeet, Umesh Goutam, Sarvjeet Kukreja, Sundaresha Siddappa, Salej Sood, Jagdev Sharma, and Vinay Bhardwaj. 2021. 'Biofortification Strategies to Improve Iron Concentrations in Potato Tubers: Lessons and Future Opportunities'. *Potato Research*, June. <https://doi.org/10.1007/s11540-021-09508-x>.
- Singh, S.P., B. Keller, W. Gruissem, and N.K. Bhullar. 2017. 'Rice NICOTIANAMINE SYNTHASE 2 Expression Improves Dietary Iron and Zinc Levels in Wheat'. *Theoretical and Applied Genetics* 130: 283–92.
- Singh, Simrat Pal, Wilhelm Gruissem, and Navreet K. Bhullar. 2017. 'Single Genetic Locus Improvement of Iron, Zinc and β -Carotene Content in Rice Grains'. *Scientific Reports* 7 (1): 6883. <https://doi.org/10.1038/s41598-017-07198-5>.
- Sinha, Sweta, and N. Kumaravadevel. 2016. 'Understanding Genetic Diversity of Sorghum Using Quantitative Traits'. *Scientifica* 2016: 3075023. <https://doi.org/10.1155/2016/3075023>.
- Sodedji, Frejus Ariel Kpedetin, Dahye Ryu, Jaeyoung Choi, Symphonie Agbahoungba, Achille Ephrem Assogbadjo, Simon-Pierre Assanvo N'Guetta, Je Hyeong Jung, Chu Won Cho, and Ho-Youn Kim. 2022. 'Genetic Diversity and Association Analysis for Carotenoid Content among Proxymes of Cowpea (*Vigna Unguiculata* L. Walp)'. *International Journal of Molecular Sciences* 23 (7): 3696. <https://doi.org/10.3390/ijms23073696>.
- Sparvoli, F., and E. Cominelli. 2015. 'Seed Biofortification and Phytic Acid Reduction: A Conflict of Interest for the Plant?' *Plants* 4: 728–55.
- Steur, H. 2010. 'Health Impact in China of Folate-Biofortified Rice'. *Nature Biotechnology* 28: 554–56.
- Steur, H. 2012. 'Potential impact and cost-effectiveness of multi-biofortified rice in China'. *Nature Biotechnology* 29: 432–42.
- Steur, H. 2015. 'Status and Market Potential of Transgenic Biofortified Crops'. *Nature Biotechnology* 33: 25–29.
- Steur, H., M. Demont, X. Gellynck, and A.J. Stein. 2017. 'The Social and Economic Impact of Biofortification through Genetic Modification'. *Current Opinion in Biotechnology* 44: 161–68.
- Storozhenko, S. 2007. 'Folate Fortification of rice by Metabolic Engineering'. *Nature Biotechnology* 25: 1277–79.
- Strobbe, S. and Van Der Straeten, D. 2018. 'Toward Eradication of B-Vitamin Deficiencies: Considerations for Crop Biofortification'. *Frontiers in Plant Science* 9. <https://doi.org/10.3389/fpls.2018.00443>.
- Strobbe, S., and Van Der Straeten, D.. 2017. 'Folate Biofortification in Food Crops'. *Current Opinion in Biotechnology* 44 (April): 202–11. <https://doi.org/10.1016/j.copbio.2016.12.003>.
- Swamy, B.P.M. 2019. 'Compositional Analysis of Genetically Engineered GR2E "Golden Rice" in Comparison to That of Conventional Rice'. *J Agric Food Chem* 67: 7986–94.
- Tadele, Zerihun. 2019. 'Orphan Crops: Their Importance and the Urgency of Improvement'. *Planta* 250 (3): 677–94. <https://doi.org/10.1007/s00425-019-03210-6>.
- Talabi, Abidemi Olutayo, Prashant Vikram, Sumitha Thushar, Hifzur Rahman, Hayatullah Ahmadzai, Nhamo Nhamo, Mohammed Shahid, and Rakesh Kumar Singh. 2022. 'Orphan Crops: A Best Fit for Dietary Enrichment and Diversification in Highly Deteriorated Marginal Environments'. *Frontiers in Plant Science* 13. <https://www.frontiersin.org/articles/10.3389/fpls.2022.839704>.
- Tam, E., E.C. Keats, F. Rind, J.K. Das, and Z.A. Bhutta. 2020. 'Micronutrient Supplementation and Fortification Interventions on Health and Development Outcomes among Children Under-Five in Low- and Middle-Income Countries: A Systematic Review and Meta-Analysis'. *Nutrients* 12: 289.
- Tan, Xin Lin, Susan Azam-Ali, Ee Von Goh, Maysoun Mustafa, Hui Hui Chai, Wai Kuan Ho, Sean Mayes, Tafadzwanashe Mabhaudhi, Sayed Azam-Ali, and Festo Massawe. 2020. 'Bambara Groundnut: An Underutilized

Leguminous Crop for Global Food Security and Nutrition'. *Frontiers in Nutrition* 7. <https://www.frontiersin.org/articles/10.3389/fnut.2020.601496>.

Tara Satyavathi, C., Supriya Ambawat, Subaran Singh, Charu Lata, Shalini Tiwari, and Chandra Nayaka Siddaiah. 2021. 'Genomic Designing for Biotic Stress Resistance in Pearl Millet [*Pennisetum Glaucum* (L.) R. Br.]'. In *Genomic Designing for Biotic Stress Resistant Cereal Crops*, edited by Chittaranjan Kole, 257–94. Cham: Springer International Publishing. https://doi.org/10.1007/978-3-030-75879-0_6.

Tari, I., G. Laskay, Z. Takács, and P. Poór. 2013. 'Response of Sorghum to Abiotic Stresses: A Review'. *Journal of Agronomy and Crop Science* 199 (4): 264–74. <https://doi.org/10.1111/jac.12017>.

Tay Fernandez, Cassandria G., Kalidas Pati, Anita A. Severn-Ellis, Jacqueline Batley, and David Edwards. 2021. 'Studying the Genetic Diversity of Yam Bean Using a New Draft Genome Assembly'. *Agronomy* 11 (5): 953. <https://doi.org/10.3390/agronomy11050953>.

Taylor, John R.N., and M. Naushad Emmambux. 2008. 'Gluten-Free Foods and Beverages from Millets'. In *Gluten-Free Cereal Products and Beverages*, 119–48. Elsevier. <https://doi.org/10.1016/B978-012373739-7.50008-3>.

The State of Food Security and Nutrition in the World 2020. 2020. FAO, IFAD, UNICEF, WFP and WHO. <https://doi.org/10.4060/ca9692en>.

The State of Food Security and Nutrition in the World 2021. 2021. FAO, IFAD, UNICEF, WFP and WHO. <https://doi.org/10.4060/cb4474en>.

Thilakarathna, Malinda S., and Manish N. Raizada. 2015. 'A Review of Nutrient Management Studies Involving Finger Millet in the Semi-Arid Tropics of Asia and Africa'. *Agronomy* 5 (3): 262–90. <https://doi.org/10.3390/agronomy5030262>.

Thompson, Brian, Marc J. Cohen, and Janice Meerman. 2012. 'World Food Insecurity and Malnutrition: Scope, Trends, Causes and Consequences'. In *The Impact of Climate Change and Bioenergy on Nutrition*, edited by Brian Thompson and Marc J. Cohen, 21–41. Dordrecht: Springer Netherlands. https://doi.org/10.1007/978-94-007-0110-6_3.

Thresh, J. M., and R. J. Cooter. 2005. 'Strategies for Controlling Cassava Mosaic Virus Disease in Africa'. *Plant Pathology* 54 (5): 587–614. <https://doi.org/10.1111/j.1365-3059.2005.01282.x>.

Tiozon, R.J.N., A.R. Fernie, and N. Sreeshivasulu. 2021. 'Meeting Human Dietary Vitamin Requirements in the Staple Rice via Strategies of Biofortification and Post-Harvest Fortification'. *Trends Food Science Technology* 109: 65–82.

Titcomb, T.J., and S.A. Tanumihardjo. 2019. 'Global Concerns with B Vitamin Statuses: Biofortification, Fortification, Hidden Hunger Interactions, and Toxicity'. *Comprehensive Reviews in Food Science and Food Safety* 18: 1968–84.

Trijatmiko, K.R. 2016. 'Biofortified Indica Rice Attains Iron and Zinc Nutrition Dietary Targets in the Field'. *Sci Rep* 6: 19792.

Tzuri, G. 2015. 'A "Golden" SNP in CmOr Governs the Fruit Flesh Color of Melon (*Cucumis Melo*)'. *Plant Journal* 82: 267–79.

Ude, G., M. Pillay, E. Ogundiwin, and A. Tenkouano. 2003. 'Genetic Diversity in an African Plantain Core Collection Using AFLP and RAPD Markers'. *Theoretical and Applied Genetics* 107 (2): 248–55. <https://doi.org/10.1007/s00122-003-1246-8>.

Udmale, Parmeshwar, Indrajit Pal, Sylvia Szabo, Malay Pramanik, and Andy Large. 2020. 'Global Food Security in the Context of COVID-19: A Scenario-Based Exploratory Analysis'. *Progress in Disaster Science* 7 (October): 100120. <https://doi.org/10.1016/j.pdisas.2020.100120>.

Uusiku, Nangula P., André Oelofse, Kwaku G. Duodu, Megan J. Bester, and Mieke Faber. 2010. 'Nutritional Value of Leafy Vegetables of Sub-Saharan Africa and Their Potential Contribution to Human Health: A Review'. *Journal of Food Composition and Analysis, Horticulture, Biodiversity and Nutrition*, 23 (6): 499–509. <https://doi.org/10.1016/j.jfca.2010.05.002>.

- Uzogara, S.G., I.D. Morton, and J.W. Daniel. 1990. 'Changes in Some Antinutrients of Cowpeas (*Vigna Unguiculata*) Processed with "Kanwa" Alkaline Salt'. *Plant Foods for Human Nutrition* 40: 249–58.
- Van Der Straeten, D. 2020. 'Multiplying the Efficiency and Impact of Biofortification through Metabolic Engineering'. *Nature Communications* 11: 1–10.
- Varshney, Rajeev K., Chengcheng Shi, Mahendar Thudi, Cedric Mariac, Jason Wallace, Peng Qi, He Zhang, et al. 2017. 'Pearl Millet Genome Sequence Provides a Resource to Improve Agronomic Traits in Arid Environments'. *Nature Biotechnology* 35 (10): 969–76. <https://doi.org/10.1038/nbt.3943>.
- Varshney, Rajeev K., Chi Song, Rachit K. Saxena, Sarwar Azam, Sheng Yu, Andrew G. Sharpe, Steven Cannon, et al. 2013. 'Draft Genome Sequence of Chickpea (*Cicer Arietinum*) Provides a Resource for Trait Improvement'. *Nature Biotechnology* 31 (3): 240–46. <https://doi.org/10.1038/nbt.2491>.
- Varshney, Rajeev K., Mahendar Thudi, and Fred Muehlbauer, eds. 2017. *The Chickpea Genome*. *Compendium of Plant Genomes*. Cham: Springer International Publishing. <https://doi.org/10.1007/978-3-319-66117-9>.
- Varshney, Rajeev K., Manish Roorkiwal, Shuai Sun, Prasad Bajaj, Annapurna Chitikineni, Mahendar Thudi, Narendra P. Singh, et al. 2021. 'A Chickpea Genetic Variation Map Based on the Sequencing of 3,366 Genomes'. *Nature* 599 (7886): 622–27. <https://doi.org/10.1038/s41586-021-04066-1>.
- Varshney, Rajeev K., Wenbin Chen, Yupeng Li, Arvind K. Bharti, Rachit K. Saxena, Jessica A. Schlueter, Mark T. A. Donoghue, et al. 2012. 'Draft Genome Sequence of Pigeonpea (*Cajanus Cajan*), an Orphan Legume Crop of Resource-Poor Farmers'. *Nature Biotechnology* 30 (1): 83–89. <https://doi.org/10.1038/nbt.2022>.
- Vastola, Antonella, ed. 2015. *The Sustainability of Agro-Food and Natural Resource Systems in the Mediterranean Basin*. Cham: Springer International Publishing. <https://doi.org/10.1007/978-3-319-16357-4>.
- Vetriventhan, M., Vania C. R. Azevedo, H. D. Upadhyaya, A. Nirmalakumari, Joanna Kane-Potaka, S. Anitha, S. Antony Ceasar, et al. 2020. 'Genetic and Genomic Resources, and Breeding for Accelerating Improvement of Small Millets: Current Status and Future Interventions'. *The Nucleus* 63 (3): 217–39. <https://doi.org/10.1007/s13237-020-00322-3>.
- Wafula WN, Nicholas KK, Henry OF, Siambi M & Gweyi-Onyango JP (2016) Finger millet (*Eleusine coracana* L.) grain yield and yield components as influenced by phosphorus application and variety in Western Kenya. *Tropical Plant Research* 3(3): 673–680. <https://doi.org/10.22271/tp.2016.v3.i3.088>
- Wang, Fang, Tao Yang, Marina Burlyakova, Ling Li, Junye Jiang, Li Fang, Robert Redden, and Xuxiao Zong. 2015. 'Genetic Diversity of Grasspea and Its Relative Species Revealed by SSR Markers'. *PLoS One* 10 (3): e0118542. <https://doi.org/10.1371/journal.pone.0118542>.
- Wang, Wenquan, Bin Xiao Feng, Jinfu Xiao, Zhiqiang Xia, Xincheng Zhou, Pinghua Li, Weixiong Zhang, et al. 2014. 'Cassava Genome from a Wild Ancestor to Cultivated Varieties'. *Nature Communications* 5 (1): 5110. <https://doi.org/10.1038/ncomms6110>.
- Wang, Yanfang, Yanzi Zhang, Chongjing Dai, Jun Ma, Yewei Zhou, Sirpaul Jaikishun, Jiahui Yan, Zhenbiao Yang, Tongda Xu, and Shikui Song. 2021. 'The Establishment of Two Efficient Transformation Systems in Quinoa (Preprint)'. *Plant Cell, Tissue and Organ Culture*, April. <https://doi.org/10.21203/rs.3.rs-364280/v1>.
- Welsch, R. 2010. 'Provitamin a Accumulation in Cassava (*Manihot Esculenta*) Roots Driven by a Single Nucleotide Polymorphism in a Phytoene Synthase Gene'. *Plant Cell* 22: 3348–56.
- Win, A.Z. 2016. 'Micronutrient Deficiencies in Early Childhood Can Lower a Country's GDP: The Myanmar Example'. *Nutrition* 32: 138–40.
- Wu, T.Y., W. Gruissem, and N.K. Bhullar. 2019. 'Targeting Intracellular Transport Combined with Efficient Uptake and Storage Significantly Increases Grain Iron and Zinc Levels in Rice'. *Plant Biotechnology Journal* 17: 9–20.
- Wu, Xingbo, and Matthew W. Blair. 2017. 'Diversity in Grain Amaranths and Relatives Distinguished by Genotyping by Sequencing (GBS)'. *Frontiers in Plant Science* 8. <https://www.frontiersin.org/articles/10.3389/fpls.2017.01960>.

- Xu, Xun, Shengkai Pan, Shifeng Cheng, Bo Zhang, Desheng Mu, Peixiang Ni, Gengyun Zhang, et al. 2011. 'Genome Sequence and Analysis of the Tuber Crop Potato'. *Nature* 475 (7355): 189–95. <https://doi.org/10.1038/nature10158>.
- Yadav, Chandra Bhan, Rakesh K. Srivastava, Prakash I. Gangashetty, Rama Yadav, Luis A. J. Mur, and Rattan S. Yadav. 2021. 'Metabolite Diversity and Metabolic Genome-Wide Marker Association Studies (Mgwas) for Health Benefiting Nutritional Traits in Pearl Millet Grains'. *Cells* 10 (11): 3076. <https://doi.org/10.3390/cells10113076>.
- Yadav, K., P. Patel, A.K. Srivastava, and T.R. Ganapathi. 2017. 'Overexpression of native ferritin gene *MusaFer1* enhances iron content and oxidative stress tolerance in transgenic banana plants'. *PLoS One* 12.
- Yadav, O. P., and K. N. Rai. 2013. 'Genetic Improvement of Pearl Millet in India'. *Agricultural Research* 2 (4): 275–92. <https://doi.org/10.1007/s40003-013-0089-z>.
- Yadav, Yuvraj, G. R. Lavanya, Sushil Pandey, Manjusha Verma, Chet Ram, and Lalit Arya. 2016. 'Neutral and Functional Marker Based Genetic Diversity in Kodo Millet (*Paspalum Scrobiculatum* L.)'. *Acta Physiologiae Plantarum* 38 (3): 75. <https://doi.org/10.1007/s11738-016-2090-1>.
- Yasui, Yasuo, Hideki Hirakawa, Mariko Ueno, Katsuhiko Matsui, Tomoyuki Katsube-Tanaka, Soo Jung Yang, Jotaro Aii, Shingo Sato, and Masashi Mori. 2016. 'Assembly of the Draft Genome of Buckwheat and Its Applications in Identifying Agronomically Useful Genes'. *DNA Research: An International Journal for Rapid Publication of Reports on Genes and Genomes* 23 (3): 215–24. <https://doi.org/10.1093/dnares/dsw012>.
- Yazdani, M. 2019. 'Ectopic expression of ORANGE promotes carotenoid accumulation and fruit development in tomato'. *Plant Biotechnology Journal* 17: 33–49.
- Ye, X., and P. Beyer. 1979. 'Engineering the Provitamin A (β -Carotene) Biosynthetic Pathway into (Carotenoid-Free) Rice Endosperm'. *Science* 287: 303–5.
- Yemataw, Zerihun, Sadik Muzemil, Daniel Ambachew, Leena Tripathi, Kassahun Tesfaye, Alemayehu Chala, Audrey Farbos, et al. 2018. 'Genome Sequence Data from 17 Accessions of *Ensete Ventricosum*, a Staple Food Crop for Millions in Ethiopia'. *Data in Brief* 18 (June): 285–93. <https://doi.org/10.1016/j.dib.2018.03.026>.
- Yuan, C. Y., C. Zhang, P. Wang, S. Hu, H. P. Chang, W. J. Xiao, X. T. Lu, S. B. Jiang, J. Z. Ye, and X. H. Guo. 2014. 'Genetic Diversity Analysis of Okra (*Abelmoschus Esculentus* L.) by Inter-Simple Sequence Repeat (ISSR) Markers'. *Genetics and Molecular Research: GMR* 13 (2): 3165–75. <https://doi.org/10.4238/2014.April.25.1>.
- Yuan, H. 2015. 'A single amino acid substitution in an ORANGE protein promotes carotenoid overaccumulation in *Arabidopsis*'. *Plant Physiology* 169: 421–31.
- Yun, Z. 2019. 'Integrated Transcriptomic, Proteomic, and Metabolomics Analysis Reveals Peel Ripening of Harvested Banana under Natural Condition'. *Biomolecules* 9.
- Zainuddin, Ima M., Ahmad Fathoni, Enny Sudarmonowati, John R. Beeching, Wilhelm Gruissem, and Hervé Vanderschuren. 2018. 'Cassava Post-Harvest Physiological Deterioration: From Triggers to Symptoms'. *Postharvest Biology and Technology* 142 (August): 115–23. <https://doi.org/10.1016/j.postharvbio.2017.09.004>.
- Zanklan, A. Séraphin, Heiko C. Becker, Marten Sørensen, Elke Pawelzik, and Wolfgang J. Grüneberg. 2018. 'Genetic Diversity in Cultivated Yam Bean (*Pachyrhizus* Spp.) Evaluated through Multivariate Analysis of Morphological and Agronomic Traits'. *Genetic Resources and Crop Evolution* 65 (3): 811–43. <https://doi.org/10.1007/s10722-017-0582-5>.
- Zehr, Usha Barwale, Madhavan Narendran Nair, and Satish Govindrao Deole. 2014. Method for plant regeneration of okra. United States US8697445B2, filed 13 September 2011, and issued 15 April 2014. <https://patents.google.com/patent/US8697445B2/en>.
- Zhang, Lijun, Xiuxiu Li, Bin Ma, Qiang Gao, Huilong Du, Yuanhuai Han, Yan Li, et al. 2017. 'The Tartary Buckwheat Genome Provides Insights into Rutin Biosynthesis and Abiotic Stress Tolerance'. *Molecular Plant* 10 (9): 1224–37. <https://doi.org/10.1016/j.molp.2017.08.013>.
- Zhang, P., G. Legris, P. Coulin, and J. Puonti-Kaerlas. 2000. 'Production of Stably Transformed Cassava Plants via Particle Bombardment'. *Plant Cell Reports* 19 (10): 939–45. <https://doi.org/10.1007/s002990000224>.

Zhao, Zuo-yu, Tishu Cai, Laura Tagliani, Mike Miller, Ning Wang, Hong Pang, Marjorie Rudert, et al. 2000. 'Agrobacterium-Mediated Sorghum Transformation'. *Plant Molecular Biology* 44 (6): 789–98. <https://doi.org/10.1023/A:1026507517182>.

Journal Pre-proof

Competing interests statement

The authors declare no financial or commercial conflict of interest.

Journal Pre-proof