## **REVIEW ARTICLE**

## Biofortified Crops for Nutritional Security with Special Reference to the North-Eastern India

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## Abstract

Malnutrition resulting from the consumption of an unbalanced diet has emerged as one of the major health concerns worldwide. Due to its negative impact on growth and development in humans, it adversely affects the livelihood and prosperity of a society too. Among various alternatives, crop biofortification has emerged as the most sustainable and cost-effective approach to alleviate malnutrition. The high-yielding crop cultivars generally lack adequate levels of nutritional quality required for optimum metabolism in humans. Intensive research efforts by the Indian Council of Agricultural Research (ICAR) led National Agricultural Research Education and Extension System (NAREES) involving State Agricultural Universities (SAUs) and Central Universities (CUs), have led to the development of >150 biofortified crop cultivars across cereals, pulses and oilseeds. While nutritional qualities viz., protein, iron, zinc, calcium, lysine, tryptophan, provitamin-A, oleic acid and linoleic acid have been enhanced, the concentration of several anti-nutritional factors viz., phytate, erucic acid, glucosinolates and trypsin inhibitor have also been significantly reduced through breeding approaches. North-Eastern Region (NER) harbors a rich reservoir of genes spread over various crops like cereals, pseudocereals, pulses, oilseeds, vegetables, fruits, and medicinal and aromatic plants (MAPs). Here, we present a review of the work done by the NAREES on the biofortification of various crops and enlist the biofortified crops developed and released for commercialization by the National system of the country. The presence of a reservoir of genes in NER can help in further biofortifying the wide array of crops which can assist in improving the nutritional security of the people in India in general and the dwellers of the NER of India in particular. In addition, attempts made to popularize these crops by licensing their seed production through private seed companies and farmers' groups like farmers' producer organizations have also been dealt with.

Keywords: Anti-nutritional factors, Biofortification, Hybrid, MAS, Nutrition, Variety

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## Introduction

Consumption of food inadequate in nutritional balance leads to malnutrition in humans (Bouis et al., 2019; Gupta et al., 2019; Yadava et al., 2023). It not only affects growth and development in humans but also has severe socio-economic implications (Vasal et al., 1993; Gupta et al., 2015; Hossain et al., 2024a). Of the 3.1 billion global population that cannot afford healthy food, 735 million people are hungry too (FAO, IFAD, UNICEF, WFP and WHO 2023). Globally, 148.1 million (22.3%) children (<5 years of age) are stunted, while 45 million children (6.87%) face wasting and 37 million (5.6%) children are affected by obesity. In India, 35.5% of the children (<5 years) are stunted, 19.3% wasted and 32.1% are under-weight (NFHS-5 2019-21). Among adults, 57.0% of women (15-49 years) are anemic, while the same was 25.0% among men. Considering the paramount importance of alleviating malnutrition, world leaders at the United Nations framed the 'Sustainable Development Goals' (SDGs) for meeting the current needs (UNDESA 2023). Of the 17 goals, 12 are associated with nutrition. Of these goals, 'Goal-2 (zero hunger)' primarily addresses alleviating malnutrition. In fact,

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alleviating malnutrition is the most cost-effective step as every \$1 invested in a proven nutrition programme offers benefits worth \$16 (Global Food Policy Report 2016). Thus, a balanced and nutritious diet for people assumes great significance in mitigating malnutrition (Bouis and Welch, 2010; Mishra *et al.*, 2025).

## Approaches to Alleviate Malnutrition

Four major approaches viz., (i) food fortification (ii) medical supplementation (iii) dietary diversification and (iv) crop-biofortification are generally used for alleviating malnutrition (Hossain et al., 2021, 2022; Yadava et al., 2022). 'Food fortification' is a process of physically adding vital nutrients to the food to enrich it. In India, as for example (i) iron, folic acid and vitamin  $B_{12}$  fortified wheat and rice flour, (ii) iron and iodine fortified salts, (iii) vitamin-A and vitamin-D fortified oil and milk, have been permitted by Food Safety and Standards Authority of India (FSSAI), an agency of Govt. of India (Gol). 'Medical supplementation' is a process of providing vital nutrients through pills/ tablets. Gol-sponsored programmes, viz., (i) Weekly Iron, Folic Acid Supplementation (WIFS) programme for school adolescent boys and girls (10-19 years) and out-of-school girls (10-19 years) in urban and rural areas, and (ii) Vitamin-A Supplementation (VAS) programme for children under five, are some of the successful supplementation programmes in India. 'Dietary diversification' is a process of including diverse cereals, pulses, oilseeds, vegetables and fruits in the diet to enhance the nutritional status. 'POSHAN Abhiyaan' (Nutrition Campaign) by Gol is promoting 'Poshan Thali' (nutritious platter) comprising of cereals, pulses, oilseeds, vegetables, fruits, milk and other components like sugar, jaggery and oils for balanced nutrition to increase low birth weight and reduce the level of stunting, undernutrition and anemia among young children, women and adolescent girls. However, these programs are not sustainable in the long run. Lack of purchasing power, poor infrastructure, crop seasonality, higher expense, and lower bioavailability are some of the reasons that affect the successful implementation of these progarmmes (van Lieshout and de Pee 2005).

'Crop biofortification' is defined as a process of enhancing the nutritional qualities of edible parts of the plants through genetic approaches such as plant breeding, gene-editing and transgenics (Neeraja *et al.*, 2022). Development of (i) iron, zinc and protein-rich wheat grains, (ii) protein- and zinc-rich rice grains, and (iii) vitamin-E, vitamin-A, lysine and tryptophan-rich maize grains developed through breeding approaches are some of the examples of 'Crop Biofortification'. Besides, provitamin-A-rich 'Golden rice' generated through transgenic approach is another example of a crop biofortification approach. 'Crop biofortification' is regarded as the most sustainable approach, through which nutrients are provided in natural form (Bouis and Welch 2010). Further, people can easily afford the 'biofortified food' as it does not involve any additional cost during cultivation. Besides, biofortified crop varieties are as high-yielding as traditional varieties, thus no yield penalty is suffered by the farmers. More importantly, it does not require elaborate infrastructure facilities as in 'food fortification'. In addition, it also does not need an elaborate distribution system as required in 'medical supplementation' programmes (Yadava *et al.,* 2022).

Globally, the plant breeding-based biofortification program led by HarvestPlus has assisted in developing >293 biofortified varieties, some of which are (i) iron-rich pearl millet and bean, (ii) zinc-rich rice, wheat and maize, and (iii) provitaminA-rich maize, cassava and sweet potato (HarvestPlus Annual Report 2022). More than 1,00,000 tonnes of seeds of biofortified crops were produced and distributed in 2022 by HarvestPlus throughout the globe. An estimated 17.3 million farming households grew biofortified varieties during 2022 thereby enriching the diets of 86.5 million people (www.harvestplus.org). The positive impact of biofortified crop varieties is well documented worldwide. The beneficial effect of iron-rich rice (Haas et al., 2005), zinc-rich wheat (Sazawal et al., 2018), iron-rich pearl millet (Finkelstein et al., 2015), lysine- and tryptophan-rich maize (Gunaratna et al., 2010), provitamin-A rich maize (Sheftel et al., 2017), and provitamin-A rich orange-fleshed sweet potato (Low et al., 2007) have shown great promise in alleviating malnutrition in humans.

# Genetic Diversity of Crops for Nutritional Traits in North-Eastern India

North-Eastern Region (NER) of India comprising eight states viz., Arunachal Pradesh, Assam, Meghalaya, Manipur, Tripura, Mizoram, Nagaland and Sikkim, is known for its diverse nature of soil, climate, and topography (Asati and Yadav et al., 2004). This region possesses rich reservoir of many crops including cereals, pulses, fruits, vegetables, flowers particularly orchids, spices and medicinal and aromatic plants (MAPs) (Deka et al., 2012; Prakash et al., 2024). Among cereals, rice, barley, coix, finger millet and foxtail millet are native to this region (Upadhaya and Sundriyal 1998). Wide genetic diversity in pulses such as pigeon pea, rice bean, mung bean, winged bean, Dolichos bean and sword bean has also been reported (Borthakur 1992). NER also possesses a large diversity of maize (Singh and Rana 1995; Prasanna and Sharma 2005; Sharma and Pradhan 2023). Farmers in the NER still grow traditional landraces of many of these crops for family and community needs (Devi et al., 2023). 'Chakhao' rice landraces of Manipur possess deep-pigmented kernels rich in anthocyanins (Bhuvaneswari et al., 2020). 'Malbhog' landrace of rice is rich in protein and oil. While rice landraces viz., 'Bhaja', 'Komal Chaul', 'Komal Ronga', 'Komal Sakowa', 'Sakowa Bora' and 'Sakowa Dhan' are zero cooking soft rice. 'ARC10075', a rice accession collected by Dr. B.C. Patra in 2013

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from West Garo Hills, Meghalaya, is rich in protein and has been used as a donor for the development of protein-rich rice varieties *viz*. CR Dhan-310 and CR Dhan-311. Similarly, 'Mimban' landraces of maize are rich in amylopectin (Rathod *et al.*, 2019). Baruah *et al.*, (2023) also reported some of the maize landraces being rich in pigments such as anthocyanins and phlobaphenes. Thus, landraces rich in nutritional qualities such as vitamins, antioxidants and minerals, can serve as suitable donors for the development of high-yielding biofortified crop cultivars.

## Crop Biofortification Programs in India

The National Agricultural Research Education and Extension System (NAREES) led by the Indian Council of Agricultural Research (ICAR) in collaboration with State Agricultural Universities (SAUs) and Central Universities (CUs) have contributed immensely to making India self-sufficient in food production (Singh et al., 2022). The food grain production of 50.82 million tons (MMT) in 1950-51 has been increased to 332.22 MMT during 2023-24 (www.indiastat. com). The research efforts on the enhancement of grain yield were initiated during the 'Green Revolution' in the mid-1960s under the leadership of Prof. MS Swaminathan (Kesavan and Swaminathan 2006). So far NAREES has developed 6,563 high-yielding cultivars of various field crops. In the process of yield enhancement, nutritional quality was not given due importance in the breeding program, and as a result, the majority of these varieties do not possess the desired level of nutritional attributes (Debnath et al., 2023). Realizing the importance of nutritional quality, the research efforts of NAREES have now led to the development and release of a series of biofortified crop varieties through All India Coordinated Research Projects (AICRPs) for different crops through breeding approaches (Yadava *et al.*, 2022). Besides, a special project on the Consortium Research Platform (CRP) on 'Crop Biofortification' was launched in 2014-15, and it is continuing with the development of several biofortified crop cultivars.

## Biofortified Crop Cultivars Released for Commercial Cultivation

Research effort at ICAR-led NAREES has led to the release of 152 field crop cultivars across rice, wheat, maize, pearl millet, finger millet, little millet, proso millet, foxtail millet, brown-top millet, lentil, field pea, mung bean, urd bean, chickpea, faba bean, groundnut, linseed, mustard, soybean and grain amaranth (Figure 1). These cultivars have been improved for essential nutrients *viz.*, iron, zinc, calcium, protein, lysine, tryptophan, provitamin-A, oleic acid and linoleic acid (Table 1). The concentration of several anti-nutritional factors *viz.*, phytate, erucic acid, glucosinolates and trypsin inhibitor has been significantly reduced in some of the cultivars (Table 1). Off-flavour of soybean grains has also been reduced.

Some of the important biofortified crop varieties developed across crops are zinc-rich varieties of rice such as 'DRR Dhan-49' (25.2 ppm), 'Zinco Rice MS' (27.4 ppm) and 'CR Dhan-315' (24.9 ppm). Besides, 'CR Dhan-310' with 10.3% protein (in milled rice), and 'CR Dhan-411' with 10.1% protein were also released. In addition, 'CR Dhan 311' possesses both



Figure 1: Crop-wise biofortified cultivars developed through breeding

high-grain protein (10.1%) and zinc (20 ppm). In wheat; the concentration of iron, zinc and protein has been improved. Of these, 'HD-3171' (47.1 ppm zinc), 'DBW-187' (43.1 ppm iron) and 'HI-8801' (12.4% protein) are the biofortified wheat varieties released for cultivation. Besides, Pusa Tejas (41.1 ppm iron, 42.8 ppm zinc, 12.0% protein) is a multinutrient-rich wheat cultivar released for commercial cultivation in India (Yadava *et al.*, 2022). In maize, the quality of protein has been enhanced by increasing two essential amino acids, lysine and tryptophan. Though the first quality protein maize (QPM) cultivar (composite), Shakti-1 was released way back in 1997 (Hossain *et al.*, 2019), the first marker-assisted selection (MAS)-derived QPM hybrid, 'Vivek QPM-9' (lysine:

 Table 1: Important crops and traits targeted for crop biofortification in India

Crop	Nutrients	Baseline levels	Levels achieved
Rice	Protein (%)	7.0-8.0	>10.0
	Zinc (ppm)	12.0–16.0	>20.0
Wheat	Protein (%)	8–10	>12.0
	Iron (ppm)	28.0-32.0	>38.0
	Zinc (ppm)	30.0-32.0	>37.0
Maize	Provitamin-A (ppm)	0.5–1.5	>5.0
	Lysine (%)	1.5–2.0	>2.5
	Tryptophan (%)	0.3-0.4	>0.6
	Vitamin-E (ppm)	6–8	>14
Pearl Millet	Protein (%)	8.0-9.0	>15.0
	Iron (ppm)	45.0-50.0	>70.0
	Zinc (ppm)	30.0-35.0	>39.0
Finger Millet	Iron (ppm)	25.0	>38.0
	Zinc (ppm)	16.0	>24.0
	Calcium (mg/100 g)	200.0	>400.0
Small Millet	lron (ppm)	25	>55
	Zinc (ppm)	20	>33
Lentil	Iron (ppm)	45.0-50.0	>62.0
	Zinc (ppm)	35.0-40.0	>50.0
Groundnut	Oleic acid (%)	45.0-52.0	>70.0
Linseed	Linoleic acid (%)	20–25	>58
Soybean	Oleic acid (%)	22–25	>40.0
Antinutritional factors			
Maize	Phytate (mg/g)	3–8	<2.5
Mustard	Erucic acid (%)	>40.0	<2.0
	Glucosinolates (ppm)	>120.0	<30.0
Soybean	Kunitz trypsin inhibitor	30–45 mg/g of seed meal	Negligible
	Lipoxygenase	High beany flavor	Low beany flavor

4.19%, tryptophan: 0.83%) was developed by introgressing opaque2 gene and was released during 2008 (Gupta et al., 2013). Besides, several opaque2-based QPM hybrids viz., 'Pusa HM4 Improved' (lysine: 3.62%, tryptophan: 0.91%), 'Pusa HM8 Improved' (lysine: 4.18%, tryptophan: 1.01%) and 'Pusa HM9 Improved' (lysine: 2.97%, tryptophan: 0.68%) have been released using MAS (Hossain et al., 2018). Furthermore, maize has also been biofortified with provitamin-A. 'Pusa Vivek Hybrid-27 Improved' with high provitamin-A (5.49 ppm) developed through MAS for crtRB1 gene has been released for cultivation. Besides, 'Pusa Vivek QPM-9 Improved' with higher provitamin-A (8.15 ppm), lysine (2.67%) and tryptophan (0.74%) were developed through marker-assisted introgression of opaque2 and crtRB1 genes (Muthusamy et al., 2014). 'Pusa Vivek QPM-9 Improved' earns the distinction of being the world's first multi-nutrient-rich maize hybrid with higher provitamin-A and protein quality. Maize has also been improved for vitamin-E, and 'Pusa Biofortified Maize Hybrid-5' with enhanced α-tocopherol/ vitamin-E (21.60 ppm), provitamin-A (6.22 ppm), lysine (4.93%) and tryptophan (1.01%) has been recently released. This hybrid has been developed by marker-assisted stacking of vte4, crtRB1, lcyE and opaque2 genes (Das et al., 2021).

In pearl millet, 'AHB-1200Fe' (73.0 ppm iron), 'HHB-299' (73.0 ppm iron, 41.0 ppm zinc), 'AHB-1269Fe' (91.0 ppm iron, 43.0 ppm zinc) have been released. 'HHB-67 Improved2' possessing high protein (15.5%), iron (54.8 ppm) and zinc (39.6 ppm) have also been released for commercial cultivation in India. In addition, biofortified finger millet variety, 'VR-929' (131.8 ppm iron), 'CFMV-1' (428 mg/100g calcium, 58.0 ppm iron, 44.0 ppm zinc) and 'CFMV-2' (454 mg/100g calcium, 39.0 ppm iron, 25.0 ppm zinc) were released. In little millet, 'CLMV-1' with higher iron (59.0 ppm) and zinc (35.0 ppm) was developed and released for commercial cultivation (Yadava et al., 2022). Besides, 'HB-1' (55 ppm iron, 35 ppm zinc) in proso millet, 'Mahanandi' (440 ppm of calcium) in foxtail millet, and 'Hagari Browntop-2' (70 ppm iron, 42 ppm zinc) in brown-top millet, have been released.

Lentil has been improved for both iron and zinc (Yadava *et al.*, 2022). Biofortified lentils, 'Pusa Ageti Masoor' (65.0 ppm iron) and 'IPL 220' (73.0 ppm iron, 51.0 ppm zinc) have been released. In field pea, 'Arpan' with 91.5 ppm of iron and 50.5 ppm of zinc has been released for cultivation. Besides, 'Shreejan' (33.0 ppm zinc, 215.8 ppm iron) in mungbean, 'TJU-339' (104.6 ppm iron) in urdbean, 'IPC 2005-62' (27.25% protein) in desi chickpea, 'HFB-3' (28.05% protein) in faba bean, and 'RMA 62' (11.83% protein, 5.17% lysine) in grain amaranth have also been released for commercial cultivation.

Groundnut varieties, 'Girnar-4' (78.5 % oleic acid) and 'Girnar-5' (78.4% oleic acid) were developed by introgressing *ahFAD2a* and *ahFAD2b* genes through MAS. In soybean, oleic acid content has also been improved in 'NRC-147' (42%).

In linseed, 'TL-99' possessing 58.8% linoleic acid has been developed.

## Reducing the Antinutritional Factors

Antinutritional factors have been significantly reduced through breeding. Phytate, a natural chelator of positively charged ions such as iron and zinc, has been reduced in maize kernel. 'PMH-1-LP' - a single cross maize hybrid developed through marker-assisted introgression of the lpa2 gene with reduced phytate (1.89 mg/g) has been released for commercial cultivation in India. In the case of mustard, two anti-nutritional factors erucic acid and glucosinolates have been reduced below the threshold level (Yadava et al., 2022). Single zero mustard varieties, 'Pusa Mustard-30' (1.20% erucic acid) and 'Pusa Mustard-32' (1.32% erucic acid) have been developed and released. Double zero varieties, 'PDZM-31' (0.76% erucic acid, 29.41 ppm glucosinolates) and 'PDZM-33' (0.58% erucic acid, 15.17 ppm glucosinolates) were also developed. In soybean, Kunitz trypsin inhibitor (KTI) free varieties were developed through marker-assisted introgression of the null allele of the KTi3 gene, while lipoxygenase activity was reduced using the null allele of the Lox2 gene. 'NRC- 127' and 'MACSNRC-1667' are KTI-free varieties. 'NRC-132' is devoid of Lox2 activity. 'NRC-142' is a double biofortified soybean variety with null alleles of both KTi3 and Lox2.

So far, >37,000 quintals of breeder seeds of various biofortified crop varieties have been produced and distributed. A large number (>90,000) of front-line demonstrations (FLDs) of these biofortified crop cultivars have been undertaken. Currently, >16 mha area is under cultivation of biofortified crops in India. Biofortified cultivars have been licensed to >300 private seed companies through >1,300 memoranda of understanding (MoU) for seed production and marketing leading to faster spread.

## Challenges and Way Forward

Despite the great promise, the full potential biofortified crop cultivars are yet to be exploited (Gupta et al., 2015; NAAS 2022; Yadava et al., 2023). Apprehension of the lowyielding potential of biofortified varieties has been the most important factor for slow popularization among farmers. It is now a well-established fact that nutritional traits targeted so far do not pose any penalty on yield. However, some of the nutritional traits (lysine and tryptophan) are governed by recessive genes and contamination by foreign pollen grains from neighboring fields dilutes the quality of the produce in cross-pollinated crops. Adoption of the entire village for cultivation of the same type of biofortified cultivars would solve such a problem. In addition, there is also an urgent need to develop climate-resilient biofortified crop cultivars by stacking various genes and QTLs of biotic- and abiotic stress tolerance with loci governing nutritional qualities. Novel approaches such as gene editing and genomic selection should be an integral part of the crop improvement program (Hossain *et al.*, 2023, 2024b).

Awareness generation on the health benefits of biofortified crops is a key factor for the rapid adoption of biofortified cultivars by farmers (Yadava *et al.*, 2018). Besides, in cases where the enrichment of nutrients such as provitamin-A leads to an altered appearance of the produce (yellow vs. white maize), consumers generally hesitate to accept the newly developed biofortified produce as food (Gupta *et al.*, 2015). Strong promotional extension activities such as field demonstrations, and conveying a message through television talk, radio broadcasts, live-drama and various social media platforms would make the farmers, industry and consumers aware of the existence and benefits of biofortified crops. Further, robust linkages with agrifood-processing and poultry industry would help in the dissemination of biofortified crops.

Strong policy support would help in the rapid dissemination of biofortified varieties and hybrids (Yadava et al., 2018; Hossain et al., 2024b). Strengthening the seed chain to produce and supply good quality seeds is an important step for the popularization of biofortified cultivars. Providing subsidized seeds and other inputs would further contribute to the rapid dissemination of nutritionally improved cultivars among the farmers. Segregation of grains of biofortified crops in the market and assurance of remunerative price through minimum support price and/or premium price for biofortified grains in the market would also encourage the farmers to grow more biofortified crops. Inclusion of these biofortified cereals in different government-sponsored programs such as 'The National Food Security Mission', 'Rashtriya Krishi Vikas Yojna' as well as nutrition intervention programs such as 'Integrated Child Development Services' scheme, 'Mid-day Meal' and 'Nutrition Education and Training through Community Food and Nutrition Extension Units' would help in providing the much-needed balanced food to poor people. Through these schemes, children, pregnant women, and elderly people would greatly benefit, and would also help in the dissemination of biofortified crop cultivars. 'National Nutrition Strategy - 2017' by NITI Aayog, Gol envisages the alleviation of malnutrition in the country through food-based solutions. Dedication of biofortified crop varieties to the nation by the Hon'ble Prime Minister on October 16, 2020, September 28, 2021, and August 11, 2024, is a testimony of the commitment of Gol to alleviate malnutrition through a food-based system.

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