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INVITED ARTICLE
Prospects of Bio-fortified Varieties in Cereal Crops

Malnutrition stances a serious socio-economic implication throughout the world, more specifically in the underdeveloped and developing countries. The inadequate consumption of balanced diet deficit of several essential micronutrients has led to deprived health, reduced immunity. Micronutrient malnutrition also referred as "hidden hunger", which afflicts more than two billion people worldwide, especially women and pre-school age children (White and Broadley, 2009) in the developing countries, who are largely dependent on staple food crops. The major staple food crops lack important micronutrients, if present is in traces; and it is very difficult for poor people to afford the different kinds of fruit and vegetables due to financial condition which force them to be malnourished. For better nutrition of human beings, Zn or Fe content of wheat grains should be improved to around 40–60 mg/kg, from the existing available amount of 10–30 mg/kg (Cakmak *et al*., 2000).

Worldwide, the most common forms of micronutrient malnutrition (MNM) are dietary deficiencies of iron, vitamin A and iodine. These deficiencies affect at least one-third of the world's population, the majority of who live in developing countries. In addition to MNM, about 820 million people in developing countries are undernourished, i.e. ingest fewer calories per day than they require; and has been estimated that MNM accounts for about 7.3 per cent of the global burden of disease, with iron, zinc and vitamin A deficiency ranking among the 15 leading causes of the global disease burden (FAO 2006). On the other hand, one-third of the world population is at risk due to low dietary intake of Zn including 2 billion people in Asia; and is a major cause

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of stunting among children. More than 85% of total body zinc is found in skeletal muscles and bones. Zinc deficiency is responsible for the development of a large number of illness and diseases including stunting of growth, compromised immune system function, cancer, susceptibility to infectious diseases and poor birth outcomes in pregnant women, hair and memory loss, skin problems, weakening of body muscles, infertility in men and pneumonia in children.

The importance and need of the micronutrients in diet and health have forced plant scientists to develop the genotypes, which may have the optimum requirement of the micronutrient for healthy people and can fulfil the daily need as fed alone. The biofortification emerges as technology to develop such genotypes which refer to genetically increasing the bioavailable mineral content of food crops (Brinch-Pederson *et al*., 2007). Biofortification is the process by which the nutrient density of food crops is increased through conventional plant breeding, and/or improved agronomic practices and/or modern biotechnology without sacrificing any characteristic that is preferred by consumers or most importantly to farmers (Nestle *et al*., 2006). Developing biofortified crops also improves their growth and production in soils with depleted or unavailable minerals (Borg *et al*., 2009). Conventional and molecular breeding and genetic engineering techniques are the approaches that may be used to biofortify the crops with iron, zinc, and vitamin A (Tiwari *et al*., 2010).

Significance of Biofortification in Cereal Crops

About more than half of the humanity depends on cereals for their daily calorie intake out of which rice, wheat and maize provide about 23%, 17% and 10%, respectively, of the calories acquired globally. Developing countries depend more on cereal grains for their nutritional needs than the developed world. Close to 60% of calories in developing countries are derived directly from cereals, with values exceeding 80% in the poorest countries. By comparison, approximately 30% of calories in the developed world are derived directly from cereals (Awika, 2011). Cereals accounts a major portion of the daily diet for most of the world, so biofortified cereal can serve as a means to fight against the malnourishment throughout the world. The cereal can serve the purpose due to its daily intake and utilised quantity of it daily. Few of the advantages of cereal as a candidate for biofortification are listed as follows:

1. It capitalizes on the regular daily intake of a consistent and large amount of food staples by all family members.
2. One-time investment to develop seeds that fortify themselves, recurrent costs are low, and the germplasm can be shared internationally and will be cost-effective.
3. Biofortified crop system is highly sustainable and provides a feasible means of reaching undernourished populations in remote rural areas, delivering naturally fortified foods to people with limited access to commercially marketed fortified foods.
4. Breeding for higher trace mineral density in seeds will not incur a yield penalty. With mineral-dense seeds, more seedlings survive, and initial growth is more rapid. Ultimately, yields are higher, particularly in trace mineral "deficient" soils of arid and calcareous regions (Monasterio *et al*., 2007).

Harvest Plus programme leads the global effort to make the familiar staple foods more nutritious and available to those suffering from hidden hunger. Founded in 2003, it is now a part of the CGIAR Research Program on Agriculture for Nutrition and Health. Harvest Plus uses biofortification to breed higher amounts of vitamins and minerals in staple foods, including bean, cassava, orange sweet potato, rice, maize, pearl millet, and wheat. The Harvest Plus biofortification program mainly focuses on three micronutrients viz; iron, zinc, and vitamin A-that are widely recognized by the World Health Organization (WHO) as limiting.

There has been only a limited success for biofortification through conventional breeding which involves human selection out of extensive natural and induced variation. As the need is for heritable but not environment influenced trait enhancement; and as conventional breeding techniques have been either inadequate or even impossible to adopt in the case of many crops, the tools of Genetic Engineering (GE) are (a) elegant as they seamlessly combine several bioscience techniques, (b) predictable as we can determine the kind of change being introduced, (c) precise as the gene of interest can be inserted at a specific position on a specific chromosome and (d) trackable as we can map back and forth the genes and the changes they cause (Kameswara Rao and Seetharam 2014).

Benefits of Bio-fortified Varieties

The deployment of biofortified cultivars holds great promise for the health and wellbeing of the human population. Several studies have demonstrated the positive effects of these biofortified crops on humans. A study conducted on 246 children of 12-16 year of age in Maharashtra, by feeding them with 'bhakri' (round flat unleavened bread) made from iron-rich and conventional pearl millet grains, demonstrated that feeding iron-rich pearl millet was an efficient approach to improve iron status in school-age children. The development and promotion of biofortified varieties thus would help address malnutrition. Since nutritional traits such as minerals and vitamins do not affect the grain yield, these need to be brought into the mainstream where varieties across crops developed in future should possess at least set level of one/few selected nutritional traits. The high yielding biofortified crops assume great significance for nutritional security.

Criteria for Biofortification of Cereals

Apart from high quantity and need, the cereal crop should have the following criteria for the biofortification:

a. High yielding: Crop productivity must be maintained /enhanced to guarantee farmer acceptance.
b. Effective: Micronutrient enrichment levels must have a significant impact on human health.
c. Stability: Enriched levels must be relatively stable.
d. Efficacious: Bioavailability in enriched lines must be tested in humans to ensure that they improve the micronutrient status of people preparing and consuming them.
e. Consumer acceptance: Taste and cooking quality must be tested.

Strategies for Cereal Biofortification

Different strategies have been employed for biofortification successfully. These strategies can be used alone or in combination with others. To effectively target biofortification of cereals, the main strategies are:

(i) Enhanced uptake from the soil
(ii) Increased transport of micronutrients to grains
(iii) Increased sequestration of minerals to endosperm rather than husk and aleurone
(iv) Reduction in antinutritional factors in grains and
(v) Increase in promoters of mineral bioavailability in grains

Steps in Biofortification

Success of the biofortification through conventional plant breeding depends on the available variation for the micronutrient as the transgenics faces the biosafety issues throughout the world for human consumption and animal feed. The steps of biofortification include:

a. Identification of genetic variability within the range that can influence human nutrition.
b. Introgressing this variation into a high yielding, stress-tolerant genotypes possessing acceptable end-use quality attributes.
c. Testing the stability of micronutrient accumulation across the target environment.
d. Large scale deployment of the seed of improved cultivars to farmers.

Advantages of Biofortification

Biofortification possess several advantages in addition to the crop improvement, major advantages are (i) Reaching the malnourished in rural areas and biofortification strategy seeks to put the micronutrient dense trait in the most profitable, highest-yielding varieties targeted to farmers and to place these traits in as many released varieties as is feasible. These crops makes their way into retail outlets, reaching consumers in both rural and urban areas. (ii) Cost-effectiveness and low cost after the one-time investment are made to develop seeds that fortify themselves, recurrent costs are low, and germplasm may be shared internationally. It is this multiplier aspect of plant breeding across time and distance that makes it so cost-effective. (iii) Behavioural change in mineral micronutrients make up a tiny fraction of the physical mass of seed, 5–10 parts per million in milled rice. Whether, such small amounts will alter the appearance, taste, texture or cooking quality of foods is needed to investigate. If increased densities in iron and zinc are not noticeable by consumers, the dissemination strategy for trace minerals could rely on the existing producer and consumer behaviour. (iv) Sustainability of biofortification crop system is high; and nutritionally improved varieties will continue to grow and consumed year after year, even if government attention and international funding for micronutrient issue fade. (v) Relies on the plant's biosynthetic (Vitamin) or physiological (mineral) capacity with no effect of policy change or weak funding.

Limitations of Biofortification

Though biofortification has several advantages, it has few limitations too. (i) Low acceptability: There may occasionally be difficulties in getting biofortified foods to be accepted if they have different characteristics to their unfortified counterparts. For example, vitamin A enhanced foods are often dark yellow or orange– this, for example, is problematic for many in Africa, where white maize is eaten by humans and yellow maize is negatively associated with animal feed or food aid or where white-fleshed sweet potato is preferred to its moister, orange-fleshed counterpart (ii) Varying impact throughout the life cycle: Biofortified staple foods can contribute to body stores of micronutrients such as iron, zinc, and vitamin A (the three target nutrients) throughout the lifecycle, including those of children, adolescents, adult women, men, and the elderly. The potential benefits from biofortification are, however, not equivalent across all of these groups and depend on the amount of staple food consumed the prevalence of existing micronutrient deficiencies, and the micronutrient requirement as affected by daily losses of micronutrient from the body, and special needs for processes such as growth, pregnancy, and lactation. (iii) Risk: There is a theoretical risk that a gene inserted by a genetic engineering (GE) process (such as the gene that codes for beta-carotene, the precursor of Vitamin A) could pass to related crop or wild plants with unknown effects. There is no evidence to support this risk but for this and other reasons, GE crops require mandatory field testing to assess environmental risks. These are likely to be costly and regulations in many countries may mean that a GE approach to

Table 1. Bio-fortified varieties released in various cereal crops

Crop	Bio-fortified varieties
Rice	CR Dhan 310, DRR Dhan 45, DRR Dhan 49, CR Dhan 315
Wheat	WB 02, HPBW 01, Pusa Tejas (HI 8759), Pusa Ujala (HI 1605), MACS 4028, HI 8777, HI 1633, HD 3298, DBW 303, DDW 48
Maize	Pusa Vivek QPM9 improved, Pusa HM4 improved, Pusa HM9 improved, Quality Protein Maize Hybrid 1, Quality protein maize hybrid 2, Quality protein maize Hybrid 3, Shakti-1 (composite) and hybrids, namely Shaktiman-1, Shaktiman-2, HQPM-1, Shaktiman-3, Shaktiman-4, HQPM-5, HQPM-7, Vivek QPM-9, HQPM-4, Pratap QPM Hybrid-1 and Shaktiman-5
Pearl Millet	HHB 299, AHB 1200
Finger Millet	CFMV1, CFMV2

Table 2. Crops currently undergoing bio-fortification process with targeted nutrient range (ìg/g) (Meena et al., 2018)

Sr. No.	Crop	Target Nutrient	Nutrient Range	Nutrient Target Level
1	Rice	Zinc	13-18	Polished rice
		Iron	24-Jun	
2	Wheat	Zinc	25-65	Whole wheat
		Iron	25-56	
3	Maize	Zinc	13-58	Whole Maize
		Iron	Oct-63	
		β- Carotene	5-8.6	
4	Pearl Millet	Zinc	47	Whole pearl millet
		Iron	47	

biofortification is only justified in using a conventional breeding technology is impossible. In general, GE approaches face resistance in many countries. Marketing in the developing countries is not easy and consumer acceptance is essential for a biofortification strategy to reduce malnutrition.

Future Projections

Biofortification stand as economically and sustainably efficient mode to fight against malnutrition. The strategy of most biofortification programs is to put the traits for biofortification into the most profitable and highest yielding varieties available, which would then to some extent address the issue of insufficient food availability. For biofortified cereals to make a broad impact on the nutritional status of such children in rural Africa, ideally the predominant cereals consumed should have enhanced levels of multiple critical nutrients, as with Africa Biofortified Sorghum and the improved 'ProVitaMinRice' Golden Rice. Intensive efforts by public sector institutions and policy for intense promotional campaigns can effectively ensure a significant increase in the adoption and acceptance of biofortified crop varieties. Strengthening the seed chain to produce and supply good quality seeds is one of the important steps for the popularizing biofortified varieties. The maintenance of genetic purity is very essential for keeping the quality trait intact; hence, special seed production areas need to be identified. Providing subsidized seeds and other inputs would further contribute to the rapid dissemination of nutritionally improved cultivars among the farmers. Assured premium remunerative price through minimum support price for biofortified grains in the market will encourage the farmers to grow more biofortified crops. Investment on extension activities would make the farmers, industry and consumers aware of the availability and benefits of biofortified crops.

Areas for further research include robust new trials that test the efficacy of biofortified crops for a wider range of age and gender groups, including infants, and over a longer period (for example, before conception through infancy). Other research will test the efficacy of consuming several different biofortified crops, each providing different vitamins and/or minerals to the food basket. Nutritionists agree that biofortified crops can improve nutritional status in micronutrient-deficient populations, but additional research is needed, using other, more sensitive biochemical indicators, as well as functional indicators, to more fully understand the health impact of consuming biofortified foods. The challenge is to get consumer acceptance for biofortified crops, thereby increasing the intake of the target nutrients. With the advent of good seed systems, the development of markets and products, and demand creation, this can become a reality.

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