Review Article

Biofortification in Vegetable Crops – A Review

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Abstract

About 800 million people suffer from hunger, but even more suffer from micronutrient malnutrition, also called "hidden hunger", particularly in the developing countries. Iodine, vitamin A, iron, and zinc malnutrition are major concerns. The malnutrition of minerals (Fe, Zn) and vitamin A are major food-related primary health problem among populations of the developing world including India where there is a heavy dependence on cereal-based diets and limited access to fruits and vegetables. One such approach to combat the issue of micronutrient malnutrition is through biofortification, a process of breeding nutrients into food crops which provides a comparatively cost-effective, sustainable, and long-term means of delivering more micronutrients to rural populations in developing countries. Currently, agronomic, conventional, and transgenic biofortification are three common approaches. Agronomic micronutrient biofortification can provide temporary increases through fertilizers.

In conventional plant breeding, parent lines with high vitamin or mineral levels can be crossed over several generations to produce plants that have the desired nutrients. Transgenic approaches are advantageous when the nutrient does not naturally exist in a crop (example, provitamin A in sweet potato and cassava. Recently, there have been several reports on the development of transgenic crops to enhance levels provitamin A content in crops like tomato, potato, cassava, sweet potato, beans and other vegetable crops.

Keywords: Biofortification, malnutrition, transgenic approaches, nutrition

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Introduction

Increasing population, inadequate food and nutrition, hunger, malnourishment of vitamins and micronutrients etc. are the biggest challenges to address most of the nations across the world. Vitamin A deficiency (VAD) predominant in developing countries among children and women which leads to >600, 000 deaths each year globally among children <5 year of age. Among the micronutrient malnourishment of population about 60% of iron, 30% of zinc, 30% of iodine and 15% of selenium are predominant. Inadequate availability of these important vitamins and micronutrients resulted in many health and physical disorders in human beings. Traditional agricultural practices can partly enhance the nutritional content in plant foods but biofortification is a practice of nutrient fortification into food crops using agronomic, conventional and transgenic breeding methods to provide a sustainable and long term strategy to address negative impacts of vitamin & nutrient deficiencies. Biofortification works have been practiced in most of the horticultural crops like banana, cassava, beans, potato, orange sweet potato (OSP), cowpea, pumpkin etc. several conventional and transgenic varieties have been released, while additional varieties are in the pipeline. The results of efficacy and effectiveness studies, as well as recent successes in delivery, provide evidence that biofortification is a promising strategy for combating hidden hunger. Biofortification is the process of adding nutritional value to the crop. It refers to nutrient enrichment of crops to address them negative economic and health consequences of vitamin and mineral deficiencies in humans [49].

Bio-fortification refers to increasing genetically the bioavailable mineral content of food crops [9]. Developing biofortified crops also improves their efficiency of growth in soils with depleted or unavailable mineral composition [7]. Breeding plants with increased phytonutrients is most easily achieved with crops with short juvenile periods to reach fruiting stage such as vegetables, berries and melons, but is a much longer term strategy for tree-fruit and nuts, which usually require a juvenile period of many years before fruit-set is possible. Alternative strategies include the identification of plant variants with enhanced phytonutrient levels within germplasm collections or within existing commercial cultivars. This can identify lines that may be already acceptable to consumers, or alternatively identify a potential donor parent with the appropriate phytonutrient background for transfer in to a more acceptable plant-type for consumption [28].

Importance of Biofortification

Biofortification provides a comparatively cost-effective, sustainable, and long-term means of delivering more micronutrients in relatively remote rural areas and it also deliver naturally-fortified foods to population groups with limited access to commercially-marketed fortified foods. Biofortified staple foods cannot deliver as high level of minerals and vitamins per day as supplements or industrially fortified foods, but they can help by increasing the daily adequacy of micronutrient intakes among individuals throughout the lifecycle [8]. Biofortification is not expected to treat micronutrient deficiencies or eliminate them in all population groups. No single intervention will solve the problem of micronutrient malnutrition, but biofortification complements existing interventions to sustainably provide micronutrients to the most vulnerable people in a comparatively inexpensive and cost-effective way. For instance, according to World Health Organization (WHO) estimation, biofortification could help cure two billion people suffering from iron deficiency-induced anemia [38].

Table 1 Sources of nutrients from vegetables [19]		
Nutrients	Vegetables	
Carbohydrate	Sweet potato, potato, cassava	
Protein	Pea, lima bean, French bean, cowpea	
Vitamin A	Carrot, spinach, pumpkin	
Vitamin B ₁	Tomato, chilli, garlic, leek, pea	
Vitamin C	Chilli, sweet pepper, cabbage, drumstick	
Calcium	Hyacinth bean, amaranthus, palak	
Iron	Amaranthus, palak, spinach, lettuce, bitter gourd	
Phosphorous	Pea, lima bean, taro, drumstick leaves	
Vitamin B ₅	Palak, amaranthus, bitter gourd, pointed gourd	
Iodine	Tomato, sweet pepper, carrot, garlic, okra	
Sodium	Celery, green onion, Chinese cabbage, radish	

Methods of Biofortification

Biofortification can be achieved through three strategies

- Agronomic Biofortification
- Conventional plant breeding
- Genetic engineering

Agronomic Biofortification

Application of fertilizers to increase the micronutrients in edible parts [49]. Most suitable micronutrients for agronomic biofortification are Zinc (foliar applications of ZnSO₄), Iodine (Soil application of iodide or iodate), Selenium (as selenate).Foliar application is the quick and easy method of nutrient application to fortification of micro nutrients (Fe, Zn, Cu etc.) in plants. Several studies have found that the mycorrhizal associations increase Fe, Se, Zn and Cu concentrations in crop plants. AM-fungi increase the uptake and efficiency of micronutrients like Zn, Cu, and Fe etc. Sulphur oxidising bacteria increases the sulphur content in onion.

Biofortification of crops with Iron

Tomato plants can tolerate high levels of iodine, stored both in the vegetative tissues and fruits at concentrations that are more than sufficient for the human diet and conclude that tomato is an excellent crop for iodine-biofortification programs. The fruit concentration of iodine detected in 5 mM iodide–treated plants was more than enough to cover a daily human intake of 150 μ g [37]. Increasing iron levels of *Amaranthus* plants by using *S.platensis* as microbial inoculant when compared with control and he also reported that *Spirulina platensis* has been used as biofortifying agent to enhance the iron status in *Amaranthus gangeticus* plant [31].

Biofortification of crops with Zinc

The relationship between tuber Zn concentration and foliar Zn application followed a saturation curve, reaching a maximum at approx. 30 mg Zn kg⁻¹ DM at a foliar Zn application rate of 1.08 g plant⁻¹. Despite a 40-fold increase in shoot Zn concentration compared to the unfertilised controls following foliar Zn fertilisation with 2.16 g Zn plant⁻¹ [**58**]. The use of fertilizer "Riverm" during cultivation of sweet pepper, eggplant and tomatoes helps to be enriched by zinc. Biofortified vegetables contain 6.60-8.59 % of Zn more than control [60].

Biofortification of crops with Selenium

Se-enriched *S. pinnata* is valuable as a soil amendment for enriching broccoli and carrots with healthful forms of organic-Se. Onions and carrots were bio-fortified by foliar application of a solution of 77Se(IV) that was enriched to 99.7% as 77Se [4]. In brassica vegetables selenium application did not affect the yield or oil content [32]. High accumulation of Se in the seeds and meal of $(1.92-1.96 \ \mu g \ Se \ g^{-1})$ was detected [51].

Conventional plant breeding

Traditional breeding mainly focused on yield attributes and resistance breeding from last four decades and lack of priority on nutritional aspects leads to decreased amount of nutrient status in the existed varieties. Examples of minerals that their mean concentration in the dry matter has declined in several plant-based foods are Fe, Zn, Cu and Mg. Recent progress in conventional plant breeding has given emphasis on fortification of important vitamins, antioxidants and micronutrients. The potential to increase the micronutrient density of staple foods by conventional breeding requires adequate genetic variation in concentrations of β -carotene, other functional carotenoids, iron, zinc, and other minerals exists among cultivars, making selection of nutritionally appropriate breeding materials possible [49].

Crop	Biofortified element/mineral/vitamin	References
Tomato	Chlorogenic acid, stilbene, flavonoids, anthocyanin,	[50];[42][23]
	Folate, phytoen, lycopene β -carotene, provitamin A	[12];[21]
	Zinc, Iodine	[60];[37]
Potato	Amin acid, protein, anthocyanin, starch, carbohydrate (fructan)	[59];[11];[34];[53]
Onion, Broccoli	Selenium	[1]; [2]
Lettuce, Beans	Iron	[24]; [6]
Carrot	Calcium	[47]
Radish	Selenium	[18]
Brassica spp.	Selenium, carotene	[51];[62]
Cassava	Protein, carotene and mineral contents	[43]
	Zn, Se, Cu, I	[35]
Sweet potato	Protein	[16]
	Carotene	[29]
Broccoli	Selenium	[1]
Cucumber	Potassium	[41]
Spinach	Iodine	[63]
Pumpkin	Carotenoids	[10]

Table 2 Examples of biofortification in vegetable crops

Targeted Vegetables Crops for Bio-Fortification through plant breeding *Cassava*

Cassava, an important crop in many developing countries, contains iron and zinc only in low concentrations. Thus, the focus of bio-fortification initiatives is exclusively on increasing beta-carotene concentration [40]. Analyzing 632 accessions from the CIAT germplasm collection of 5500 accessions and detected germplasm with beta- carotene concentrations above $20 \ \mu g/g$, suggesting a high genetic variability that would make it possible to successfully bio-fortify cassava and meet the daily retinol requirements of adults [25]. It was possible to increase true protein in

cassava roots measured by amino acid contents by inter-specific hybridization and the interspecific hybrid has 10-fold lysine and 3-fold methionine than common cassava cultivar [43].

Sweet Potato

The major aim of the bio-fortification programs is the replacement of white fleshed low pro-vitamin A sweet potato varieties with orange fleshed high pro-vitamin A plants. Harvest Plus has set the target level for sweet potatoes at 32 μ g/g but varieties with concentrations up to 100 μ g/g already exist [45]. Workers provided the children with either orange fleshed potato with a beta carotene concentration of about 100 μ g/g in the cooked root or white fleshed potato without any beta- carotene over a period of 11 weeks. Vitamin A liver stores were increased in the treatment group compared to the control group [55]. Furthermore it has been shown that retention of beta carotene from orange fleshed sweet potatoes when boiled is very high with about 80% of the initial concentration [56].

Beans

In nutritional terms, beans are often called the "poor man's meat" for their inexpensive price as a protein source and their rich content of minerals (especially iron and zinc) and vitamins. The advantages and needs of mineral biofortification in common bean, starting with the steps of breeding for the trait such as germplasm screening, inheritance, physiological, or bioavailability studies and finishing with product development in the form of new biofortified varieties [6].

Cow pea

Pioneer research on biofortification of cow pea has initiated G.B. Pant University of Agriculture and Technology, Pantnagar, India. Two early maturing high iron and zinc fortified varieties namely Pant Lobia-1(82ppm Fe and 40ppm Zn), Pant Lobia-2(100ppm Fe and 37 ppm Zn) have been developed by conventional plant breeding and released in 2008 and 2010.

Recent years, biofortification in vegetables is mainly focused through transgenic approaches. Hence this review article is clearly discussed with gene manipulation.

Genetic Engineering

Genetic engineering (GE) is often described as a technology that is critical for future food, feed and energy needs. Biotech crop hectares increased by more than 100-fold from 1.7 million hectares in 1996 to 179.7 million hectares in 2015. Since the first large-scale introduction of Flavr-Saver tomato in 1996, a record 175.2 million hectares of biotech crops were grown globally in 2013, at an annual growth rate of 3 per cent [30]. This unprecedented high growth rate starting from 1.7 million hectares in 1996 to 175.2 million hectares in 2013 makes biotech crops the fastest adopted crop technology in recent history, increasing approximately 100 folds between 1996 and 2013 [27]. Transgenic crops, commonly referred to as genetically modified (GM) crops enable plant breeders to bring favorable genes, often previously inaccessible, into elite cultivars, improving their value considerably and offer unique opportunities for controlling insects, viruses and other pathogens, as well as nutritional quality and health benefits.

Lack of sufficient variation among the genotypes for the desired character/trait within the species, or when the crop itself is not suitable for conventional plant breeding (due to lack of sexuality;) then genetic engineering offers a valid alternative for increasing the concentration and bioavailability of micro nutrients in the edible crop tissues[49].

Genetic engineering enables vegetable breeders to incorporate desired transgenes into elite cultivars, thereby improving their value considerably. It further offers unique opportunities for improving nutritional quality and bringing other health benefits. Many vegetable crops have been genetically modified to improve traits such as higher nutritional status or better flavour, and to reduce bitterness, slow ripening, higher nutritional status, seedless fruit, increased sweetness and to reduce anti-nutritional factors.

Transgenic Approaches for Biofortification

Tomato

Antioxidants

Fruits and vegetables contain a wide range of antioxidants including anthocyanins and carotenoids such as lycopene and β -carotene and vitamins C and E. In transgenic fruit which accumulate trans resveratrol, there is an increase in the

levels of ascorbate and glutathione, the soluble antioxidants of primary metabolism, as well as in the total antioxidant activity [23].

Carotenoids -rich tomato

Lycopene is a potent antioxidant with the potential to prevent epithelial cancers and improve human health. Therefore, there is considerable interest in elevating the levels of carotenoids in tomato fruit by genetic manipulation and thereby improving the nutritional quality of the crop. The *Psy-1* enzyme catalyzes the first committed step of the carotenoid biosynthesis pathway by producing phytoene from GGPP (geranylgeranyl diphosphate). In order to increase the carotenoid content of fruit, the *Psy-1* gene was constitutively expressed in tomato. [5].

Anthocyanin-rich tomato

To enrich the anthocyanin content of the fruits of a commercially cultivated tomato cultivar, Arka Vikas by fruitspecific expression of two transcription factors *Ros*1 and *Del* by Agrobacterium-mediated transformation. The average anthocyanin content of the transgenic fruit was 0.1 mg g⁻¹ fresh weight, which were 70-100 folds higher than that of the control fruits. [36].

Flavonols -rich tomato

Transformation of tomato with the *Petunia chi-a* gene encoding chalcone isomerase. Resulting transgenic tomato lines produced an increase of up to 78 fold in fruit peel flavonols, mainly due to an accumulation of rutin [42]. By increasing 78-fold of total fruit flavonols was achieved through ectopic expression of a single biosynthetic enzyme, chalcone isomerase [57].

Folate -rich tomato

Engineering a moderate increase in pteridine production can significantly enhance the folate content in food plants and that boosting the PABA supply can produce further gains [21]. When transgenic PABA- and pteridine-overproduction traits were combined by crossing, vine-ripened tomato fruit accumulated up to 25-fold more folate than control[22]. Expressing a yeast S-adenosylmethionine decarboxylase gene (*ySAMdc; Spe2*) fused with a ripening-inducible E8 promoter to specifically increase levels of the polyamines spermidine and spermine in tomato fruit during ripening. This led to an increase in lycopene, prolonged vine life, and enhanced fruit juice quality [39].

Table 3 Engineering for quality improvement in tomato [5, 13]		
SI. No.	Fruit trait	Inserted target
1.	Carotenoid content	Dxs, CrtB, CrtI, CrtY, PSY-1, CRY-2, CYC-B, LCY-B, LCY-B, CHY-B, DET-
		1, COP1LIKE, CUL4, FIBRILLIN, Spermidine synthase, PG
2.	Flavonoid content	CHI, CHS, CHI, F3H, FLS, MYB12, STS, CHR, FNS-II, Del, Ros1
3.	Ascorbic acid content	GaLDH, GME, GCHA and/or ADCS
4.	Nutritional value	Crt1, Samc

Potato

The single gene overexpression of genes encoding chalcone synthase (CHS), chalcone isomerase (CHI) and dihydroflavonol reductase (DFR) resulted in a significant increase of measured phenolic acids and anthocyanins in potato [34].

Starch-rich potato

Starch is the primary storage component of carbohydrate in potato tubers accounting up to 70 per cent of tuber dry matter. Bacterium *Escherichia coli* gene *glg C16* encoding bacterial ADPGP Pase when transferred into potato, the transgenic plant showed high starch content in the tubers [53].

Protein-rich potato

Increased nutritive value may be achieved in potato by expressing a non-allergenic seed albumin gene from

Amaranthus hypochondriacus by protein-rich potato expressing the seed protein gene AmA1 (Amaranth Albumin 1). At the biochemical level, expression of AmA1 in both categories of transgenics leads to a high increase in all essential amino acids, particularly lysine, tyrosine, and the sulfur amino acids with corresponding increase in total protein content [11].

Project on Bio Cassava Plus to increase the minerals zinc and iron, vitamins A and E, protein contents and decrease cyanogen content, delay postharvest deterioration and development of virus-resistant varieties through hybridization and selective breeding methods [11].

Amino acids-rich potato

High essential amino acid encoding *heaae* gene was transferred to potato clones K-2 and K-7 and it showed increase in essential amino acids. This synthetic gene fragment (HEAAE-DNA), 292 base pairs in length, codes for a protein composed of about 80 per cent essential amino acids [59].

Sl.No.	Quality trait	Gene	source
1.	Amino acid-rich storage protein	AmA1	Amaranthus hypochondriacus
		tar1 (tarin)	Colocasia esculenta
		Boxla, BoxIIa, BoxlaIIa 2	Bertholletia excels (Brazil nut)
2.	High amylose starch	SBE I antisense	Potato
3.	Carbohydrate engineering	SUSI (sucrose synthase)	Potato
4.	High tuber galactose	stUGE451 stUGE51	Potato
5.	High tuber fructose	xylA (glucose isomerase)	Thermus thermophilus

Table 4 Recent up	pdates of trans	sgenic research	n in potato	for qualit	y improvement [46]
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β -carotene-rich potato

The single gene overexpression or simultaneous expression of genes encoding chalcone synthase (CHS), chalcone isomerase (CHI), and dihydroflavonol reductase (DFR) resulted in a significant increase of measured phenolic acids and anthocyanin [34]. The *crtB* gene was also transformed into S. phureja (cv. Mayan Gold), resulting in an increase in total carotenoid content to 78 μ g carotenoid g⁻¹ DW in the most affected transgenic line [15].

In order to enhance the carotenoid content of potato tubers, transgenic potato plants have been produced expressing an *Erwinia uredovora crtB* gene encoding phytoene synthase, specifically in the tuber of *Solanum tuberosum* L. In developing tubers of transgenic *crtB* Desiree lines, carotenoid levels reached 35 μ g carotenoid g⁻¹ DW and the balance of carotenoids changed radically compared with controls. The *crtB* gene was also transformed into *S. phureja* (cv. Mayan Gold), again resulting in an increase in total carotenoid content to 78 μ g carotenoid g-1 DW in the most affected transgenic line. In these tubers, the major carotenoids were violaxanthin, lutein, antheraxanthin, and β -carotene [15].

Enhancing carotenoid accumulation by expression of Or in transgenic potato

Cauliflower *Or* gene represents a novel gene mutation. It causes many low pigmented tissues of the plant, most noticeably the edible curd and the shoot meristem to accumulate high levels of β -carotene and turns these tissues orange. The *Or* gene has been isolated by a map-based cloning strategy. This gene appears to represent a regulatory gene in controlling carotenoid accumulation. It functions in increasing the sink capacity rather than altering the expression of genes involved in carotenoids biosynthesis [62].

To examine whether Or can be used as a new genetic tool to enhance carotenoid content in a major staple crop, Or was transformed into potato plants under the control of a granule-bound starch synthase gene to obtain tuberspecific expression. Remarkably, expression of Or in the transgenic potato tubers results in the production of orangeyellow tubers. HPLC analysis confirmed that this color change is indeed associated with enhanced levels of carotenoids, including the accumulation of β -carotene that is present at negligible amounts in the controls. The total carotenoid contents in the tubers expressing the Or transgene were six fold higher than those in the controls. This successful transformation result demonstrates that Or functions across plant species and can be used as a novel molecular tool to enrich carotenoid contents for improving the nutritional value of crops [33].

Low sugar potato

Genetically modified potato plants have also been produced in the Czech Republic. These are potatoes with an inserted gene for phosphofructokinase from bacterium *Lactobacillus bulgaricus*. Moreover, potatoes containing higher amounts of simple sugars turn brown during frying and are consequently less attractive for consumers. Transgenic potato plants not only have lower sugar content, but moreover, chips prepared from such potatoes are lighter in colour than those prepared from non-modified ones [44].

Cauliflower

Successful cloning of a cauliflower *Or* gene reveals that manipulation of chromoplast formation to provide an effective metabolic sink for carotenoid sequestration and deposition exerts a profound effect on carotenoid accumulation. The demonstration of use of the *Or* gene to increase carotenoid content in transgenic potato illustrates an alternative new approach to complement effects relying on expression of carotenogenic genes for enhancing carotenoid levels in food crops [62].

Cabbage

Red cabbage providing big amounts of anthocyanins and presenting high antioxidant properties which may decrease the risk of cardiovascular diseases, brain disorders and cancer [14].

Carrot

Genetically engineered carrot containing increased Ca levels may boost Ca uptake, thereby reducing the incidence of Ca deficiencies such as osteoporosis. Transgenically modified carrots expressed increased levels of the plant Ca transporter SCAX1 [47].

Pumpkin

The total carotenoid and β -carotene isomers contents increased according to the cooking methods applied and high contents of total carotenoids in pumpkin Carvalho *et al.* (2012) [10].

Cassava

Cyanogen-free cassava [52]

Cassava, however, contains potentially toxic levels of the cyanogenic glucoside, linamarin. The cyanogens levels in leaves (200-1, 300 mg CN equivalents/kg dry weight) and roots (10-500 mg CN equivalents/kg dry weight) of many cassava cultivars are higher than the maximum recommended cyanide levels (10 mg CN equivalents/kg dry weight) in foods established by the FAO. The cyanogen content of cassava foods can be reduced to safe levels by maceration, soaking, rinsing and baking; however, short-cut processing techniques can yield toxic food products.

To generate cyanogenic cassava plants targeted the genes (CYP79D1 and CYP79D2) encoding the cytochrome P450s that catalyze the first dedicated-step in linamarin synthesis for reduced levels of expression. As a result both leaf and root levels of linamarin are reduced up to 94% and 99%, respectively, in CYP79D1/CYP79D2 antisense plants.

Enhancing Protein content in cassava

In order to increase the nutritional quality of cassava storage roots, which contain up to 85% starch of their dry weight, but are deficient in protein, a synthetic *ASP1* gene encoding a storage protein rich in essential amino acids (80%) was introduced into embryogenic suspensions of cassava via *Agrobacterium*-mediated gene transfer [61]

Biosafety concerns in the development and commercialization of transgenics

- 1. Biosafety refers to policies and procedures adopted to ensure environmental safety during the course of the development and commercialization of the transgenic organisms.
- 2. Escape of engineered gene by the gene flow or gene disposal.
- 3. Non-target or ecological effects.

- 4. Invasiveness or weediness of transgenics. It means tendency of plant to spread beyond the field where first planted.
- 5. Creation of super-weeds and super-viruses.
- 6. Toxicity and allerginicity to human beings and animals.
- 7. Expression of undesirable phenotypic traits.
- 8. Genetic erosion of biological diversity.

Potential risks or concerns from use of GM Foods [54]

- 1. Alteration in nutritional quality of foods
- 2. Antibiotic resistance
- 3. Potential toxicity and allergenicity from GM foods
- 4. Unintentional gene transfer to wild plants
- 5. Possible creation of new toxins
- 6. Limited access to seeds through patenting of GM food plants
- 7. Threat to crop genetic diversity
- 8. Religious/cultural ethical concerns
- 9. Concerns for lack of labeling
- 10. Concerns of organic and traditional farmers
- 11. Fear of the unknown.

Substances	Nutrient
Certain organic acids (ascorbic acid, malate),	Iron and Zinc
β -carotene, cysteine containing peptides (metallothioneine)	
Haemoglobin	Iron
Long chain fatty acids (palmitate)	Zinc
Fats and lipids	Vitamin A
Selenium	Iodine
Iron and zinc	Vitamin A

Bioavailability

According to [17] bioavailability is the proportion of the total nutrient in the food utilized for the normal body function. Others consider bioavailability to reflect the efficiency with which consumed nutrients are absorbed from the alimentary tract and are thus available for storage or use [20].

Conclusion

Biofortification provides a feasible means of reaching malnourished populations in relatively remote rural areas, delivering naturally fortified foods to people with limited access to commercially-marketed fortified foods, which are more readily available in urban areas. Biofortification and commercial fortification, therefore, are highly complementary. Ultimately, good nutrition depends on adequate intakes of a range of nutrients and other compounds, in combinations and levels that are not yet completely understood. Thus, the best and final solution to eliminating under nutrition as a public health problem in developing countries is to provide increased consumption of a range of non-staple foods. However, this will require several decades to be realised, informed government policies, and a relatively large investment in agricultural research and other public and on-farm infrastructure.

To sum up in the words of M.S. Swaminathan, "GM foods have the potential to solve many of the world's hunger and malnutrition problems, and to help protect and preserve the environment by increasing yield, quality and reducing reliance upon chemical pesticides. Yet there are many challenges ahead for governments, especially in the areas of safety testing, regulation, industrial policy and food labeling."

Future Thrust

Biofortification of crops is a challenging endeavor. Many plant breeding programs focus on improvement of productivity, resistance to biotic and abiotic stresses, and food palatability. The improvement of nutritional quality has

been added as an additional breeding objective in recent years. In order to achieve these objectives, collaboration between plant breeders and nutrition scientists is essential. Moreover, it is not possible to implement some of the biofortification programs due to the lack of sufficient genetic variation for the micronutrients in the germplasm. In such situations, the application of genetic engineering approaches is needed and collaboration between plant breeders and molecular biologists is essential. The biggest hurdle in the commercial use of GM crops is the regulatory approval process which is very expensive and time consuming. Biofortification is a promising agriculturally based strategy for improving the nutritional status of malnourished populations throughout the world. Therefore, major resources should be allocated to biofortification programs.

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