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Bio-fortification in Horticultural Crops

B.V.G. Prasad[1], Smaranika Mohanta[2], S. Rahaman[3] and Prerna Bareily[4]

[1,2,3,4]Division of Horticulture, Institute of Agriculture, Visva-Bharati University, Sriniketan - 731236 (West Bengal)
E-mail: [1]prasadbvg@hotmail.com

Abstract—*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. This article provides an overall idea on bio fortification methods, SWOT analysis of each method, present and future bio fortification works in horticulture crops.*

1. INTRODUCTION

1.1. What is Biofortification?

Biofortification is the process of adding nutritional value to the crop.It refers to nutrient enrichment of crops to address the negative economic and health consequences of vitamin and mineral deficiencies in humans.

1.2. Nee for Biofortification

Vitamin A deficiency (VAD) is an important health concern in developing countries among children and women of childbearing age[1] and is estimated to account for >600,000 deaths each year globally among children <5 years of age.According to Government of India statistics provided to the World Health Organization (WHO) 62% [1] of all preschool-age children are VAD. Iron (Fe), zinc (Zn), and selenium (Se) deficiencies are serious public health issues and important soil constraints to crop production, particularly in the developing countries.

2. METHODS OF BIOFORTIFICATION

*Agronomic bio fortification *Conventional plant breeding * Genetic engineering

2.1. AgronomicBiofortification

Application of fertilizers to increase the micronutrients in edible parts. The degree of success in agronomic biofortification is proportional to the mobility of mineral element in the soil as well as in the plant[2].Most suitable micronutrients for agronomic bio fortification Zinc**,** (foliar applications of $Znso_4$),Iodine(Soil application of iodide or iodate), Selenium(as selenate).The application of inorganic Se fertilizers resulted in over 10-fold increase in Se concentrations .The use of inorganic I and Zn also had an impact on plant enrichment at a country scale in China and Thailand.Fe ($FeSO_4$) shows a low mobility in soil due to conversion of Fe^{+3} which is unavailable to plant roots. Bottleneck of Phytoavailability, to overcome this, Synthetic metal chelators (e.g. EDTA- Fe- and Zn-chelates which were effective in increasing mineral concentration in edible vegetable and fruit tissues) [3].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 [4].AM-fungi increases the uptake and efficiency of micronutrients like Zn, Cu, Fe etc.

2.2. Conventional plant breeding

Traditional breeding mainly focused on yield attributes and resistance breeding from last four decades and lack of priority on nutritional aspects lead's 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 [5].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.

Fe and Zn levels and up to a 6.6-fold variation has been reported in beans and peas [6].Apparently, this genotypic variation is generally more reduced in tubers [2] and in fruits (e.g. Fe, Zn, Ca and Mg concentrations in strawberry differed less than 2-fold;[7].Steps in biofortification by conventional plant breeding are: **Discovery (**1**.**Identify target populations,2. Set nutrient target levels, 3. Screen germplasm and gene), **Development** (4. Breed biofortified crops, 5. Test performance of new crop varieties, Measure nutrient retention in crops/food, Evaluate nutrient absorption and impact) **Dissemination (**8. Develop strategies to disseminate seeds, 9. Promote marketing & consumption of biofortified food**), Outcomes (**10. Improve nutritional status of target populations**)**

2.3. Genetic engineering

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; e.g. banana) then genetic engineering offers a valid alternative for increasing the concentration and bioavailability of micro nutrients in the edible crop tissues.One of the main concerns is theso-called 'gene flow' environmental problem, i.e. the concern of transfer of foreign genes to non-target species [8].Targets for transgenes include, redistributing micronutrients between tissues, increasing the efficiency of biochemical pathways in edible tissues, or even the reconstruction of selected pathways. Some strategies involved in the removal of 'antinutrients'.For Instance, one of the first biofortified crops was golden rice, which was engineered to produce beta-carotene or provitamin A in the edible portion of the grain [10].Since then, there have been similar successes with other crops, giving us a variety of carotenoid-enriched foods [9]as well as crops enriched with other micronutrients such as vitamin E [10]andfolate[11]. In the same way this approach is also being applied to other crops, including maize, orange cauliflower, tomato, yellow potatoes and golden canola [5]. Feeding trials by[12] demonstrated that calcium absorption was significantly increased in both mice and humans by biofortified carrots. The genetic engineering has moved into a new phase that aims at (i.e. 'multigene transfer') [8]Ex: 'Multivitamin corn' Which is engineered to produce higher levels of provitamin A, vitamin B_9, and vitamin C (b -carotene, folate, and ascorbate).Promissing lines has been identified which contained 169-fold more b -carotene, 6.1-fold more ascorbate, and double the amount of folate as foundin endosperm[13-15].Micronutrient powders, popularly known under their original name of Sprinkles, are a form of 'home fortification' that also provide several nutrients at once. In sachets for a single serving, they are sprinkled on top of normal foods. Beginning with just, encapsulated iron, they have now developed numerous varieties, with as many as 15 vitamins and minerals, appropriate for the nutritional problems of specific areas.

Table 1: SWOT analysis of bio fortification methods[5]

Agronomic Bio fortification
STRENGTHS: Comparatively simple method than other methods and suitable for immediate results.
WEAKNESS: Success limited to minerals and dependent on several factors, Needs regular application of nutrients, expensive and difficult to distribution.
OPPORTUNITIES:Often used as a compliment to other strategies
THREATS:Negative environmental impact, reverse exhaustion (Eg:Se)
Conventional plantBreeding
STRENGTHS: Successful for minerals and vitamins, one-off cost, easier distribution,Longterm strategy
WEAKNESS:Long development time, Success limited to minerals available in the soil.
OPPORTUNITIES:Wide public acceptance, simple legal frame work
THREATS: Requires genetic variation
Genetic engineering
STRENGTHS: Successful for minerals and vitamins, one-off cost, easier distribution, long term strategy,speed up process of conventional plant breeding.
WEAKNESS:Long development time,Success limited to minerals available in the soil,interactions among transgenes(may limit the process)
OPPORTUNITIES:Fast 'omics'developments
THREATS:Low public acceptance,high sociopolitical problems,Environmental impact(gene flow)

3. TARGETED HORTICULTURAL CROPS PRESENT STATS AND FUTURE PROJECTS

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.

Orange sweet potato (OSP)

To increase targeted level of 30 ppm of provitamin A in sweet potato, International Potato Center (CIP) in south Africa and Uganda(Harvest plus) + National agriculture Research and Extension System (NARES) started project in 2002-2007 and the first variety released in 2002.This variety have ability to grater provitamin A retention more than 80% after boiling or steaming and at least 75% after solar or sun drying but also high yielding and drought tolerant[16]. HarvestPlus and its partners distributed OSP to more than 24,000 households in Uganda and Mozambique. Biofortified varieties are now being introduced in many parts of Africa and South America, as well as China. In 2009, CIP launched its Sweet Potato for Profit and Health Initiative (SPHI), which seeks to deliver OSP to 10 million households in Africa by 2020.

Bio Cassava+

Project on Bio Cassava Plus initiative started in 2009 by Donald Danforth Plant Science Center to target Nigeria, Kenya with 6 major objectives namely to increase the minerals zinc and iron, vitamins A and E, protein contents and decrease cyanogen content, delay postharvest deterioration, and develop virus-resistant varieties. The scientists of Nigeria have developed three new yellow colour varieties of cassava by hybridization and selective breeding methods. The se varieties can produce higher amount of beta-carotene which helps to fight against vitamin A malnourishment in the region [17] and release of the varieties will be in 2017.

Potato

CIP (International centre for potato) started project on development of Fe rich potatos by conventional biofortification method in 2009 and the varieties will be release in 2017.

Cow pea

Pioneer research on biofortifcation 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) has been developed by conventional plant breeding and released in 2008 and 2010.Pant Lobia-3 (67 ppm Fe and 38 ppm Zn), Pant Lobia-4(51ppm Fe and 36 ppm Zn) released in 2013 and 2014 respectively. Brazil also released three varieties of high-iron cowpeas, developed by Embrapa, in 2008 and 2009 and bio availability.

Nutri banana

Breeding banana/plantain (*Musa*) is complex, as commercial varieties are sterile triploids (3X). Among the fertile groups, a high degree of cross incompatibility can exist. Further, the *Musa* crop cycle is long.Genetic engineering method of biofortification is suitable for banana because most of the edible bananas are vegetative propagated and transgene outflow are minimum and therefore genetically modified bananas can be grown alongside non-GM bananas in the same field. Also since the GM bananas are sterile, the existing diversity of bananas in India will not be affected and there won't be any heritable mixing of GM and nonGM cultivars in nature.Unfortified bananas have 0.4 mg/100 gm Fe of banana while the fortified banana would supplement this to 2.6 mg/ 100grams [18].The bio fortification of banana by increasing their beta carotene (up to 20ppm), alphatocopherol and iron content.Biofortification works on banana will be beneficial where bananas are the major staple food source and good consumer acceptance.The biofortification works on banana had been initiated at Queensland University of Technology (QUT), Australia to develop provitamin A (β-carotene),alfatocopherol and iron rich varieties besides they succeed in improving the disease resistant varieties against Banana Bunchy Top Virus (BBTV) and FusariumWilt.These varieties are under field and selection for enhanced level of micronutrients that may match pro vitamin A (PVA) and iron requirements is desirable for India. Works initiated to transfer of specific traits in two Indian banana varieties cv. Grand Nain and Rasthali.Donald Danforth Plant Science Center working on nutribanana to develop 20 ppm provitamin A bananas by Conventional breeding for Nigeria, Kenya. Bio availability trails were started 2013 and release of variety will be in 2017.

Beans

Iron(Fe) content in common bean is about 50 parts per million (ppm) and target in biofortification of bean by conventional breeding is 94 ppm, biofortified beans provide about 60% of the Estimated Average Requirement (EAR).Average bean yields in Rwanda. Non-biofortified beans produce approximately 0.8 tons/hectare (bush and climbers combined)but biofortified bush beans yield around 1.5 t/ha and biofortified climber beans 2–3 t/ha. Among the different varieties released in Rwanda in 2012 and 2014 MAC-42 from CIT contains 91ppm iron and ability to resistance against anthracnose and bean common mosaic virus and ability to produce 3.5t/ha.

Fig. 1: Altering transport gene in carrot results in more bioavailable calcium

Fig. 2: Introduction of single bacterial gene increases 'folate' levels in tomato to levels comparable to spinach

Fig. 3: Engineered potato contains3 times more calcium

Fig. 4: “Super Broccoli” created by classical genetics contains 3.4x sulforaphane, potential anti-cancer agent

Fig. 5: Nutri banana(yellow colour) (Pro vitamin-A rich)

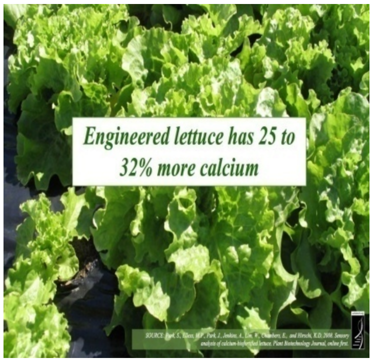

Fig. 6: Engineered lettuce has 25 to 32% more calcium

Fig. 7: Biocassva plus(yellow) Beta carotene rich

Fig. 8: Orange fleshed sweet potato

Fig. 9: Iron(Fe) rich beans

Table 1: Biofortified targetcropsandcountries-releaseschedule [19]

crop	Nutrient	Targeted country	Leading institutions	First release year
Banana /Plantain	Provitamin A Carotenoids	Nigeria, Ivory Coast, Cameroon, Burundi, DR Congo	IITA, Bioversity	Unknown
	Provitamin A Carotenoids,Iron	Uganda	Queensland University of Technology, NARO	2019
Bean	Iron (Zinc)	Rwanda, DR Congo	CIAT, RAB, INERA	2012
		Brazil	Embrapa	2008
Cassava	Provitamin A Carotenoids	DR Congo Nigeria Brazil	CIAT	
	Provitamin A Carotenoids,Iron	Nigeria, Kenya	Donald Danforth Plant Science Cente	2017
Cowpea	Iron, Zinc	India, Brazil	G.B. Pant University Embrapa	2008 2008
Irish potato	Iron	Rwanda, Ethiopia	CIP	Unknown
Pumpkin	Provitamin A Carotenoids	Brazil	Embrapa	2015
Sweet potato	Provitamin A Carotenoids	Uganda	CIP, NaCCRI	2007
		Mozambique	CIP	2002
		BraziL	Embrapa	2009
		China	Institute of Sweet Potato, CAAS	2010

IITA: International Institute of Tropical Agriculture,NARO: National Agricultural Research Organisation; CIAT: International Center for Tropical Agriculture, RAB: Rwanda Agriculture Board; INERA: Institut National pour l'Etude et la RechercheAgronomiques, CIP: International Potato Center, CAAS: Chinese Academy of Agricultural Sciences,NaCCRI: National Agricultural Crops Resources Research Institute.

4. CONCLUSION

Biofortified crops, either by conventional breeding methods or by modern biotechnological tools, are not a solution for malnurishment. The ultimate aim in global nutrition remains a sufficient and diverse diet for the world's population. However, biofortified crops can complement existing micronutrients interventions; can have a significant impact on the lives and health of millions of people, especially those most in need.

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