Review Article

Biofortification: Sources of Food Security

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Abstract | Micronutrients are an integral part of human health, normal growth as well as for optimal development of plants. More importantly, zinc and iron deficiencies have been associated with malnutrition which can be alleviated by dietary supplements in the form of pills, capsule, tablet, powder and liquid. Dietary supplements can also contain substances that have not been confirmed as being essential to life. The use of microorganisms to enable agriculture plants in efficient and productive zinc and iron absorption and translocation is a possible approach, which needs to be united into agronomic and genetic breeding strategies. Biofortified food crops including cereals, legumes and vegetables provide sufficient micronutrients to targeted populations. This method holds a lot of potential in terms of improving people's nutritional health. The objective of this study is to provide a general overview of reasons, treatments of micronutrient malnutrition across the world, as well as to explore existing knowledge and advances of biofortification for improving important food crops.

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1. Introduction

The human population has doubled by 3.5 billion to 7.5 billion, in the last 50 years; possibly it will reach 10 billion in the next decade. For agriculture probably 20% of surface is acceptable. Therefore about 6% of world's surface is used to fulfil human food requirements. Rapid increase in world's population and restricted land area for agriculture require high concentration of food production. Micronutrients and macronutrients both are necessary elements of our diet (Dubock, 2017). Nowadays the population of the world is suffering due to the deficiency of various micronutrients including 60% Fe, 30% zinc, 15% selenium deficiency, other than this, calcium and copper deficiency is very common (Kaur *et al.,* 2020). Major crops of Pakistan such as wheat and rice contain insufficient amount of major nutrients namely zinc, iron, boron (B), copper, manganese (Rehman *et al.,* 2018a).

Defective early growth and development in children due to malnutrition is major cause of economic loss of about US $7.6 billion annually in Pakistan. In Pakistan people especially pregnant women, infants, children, breast feeding mothers are facing catastrophe of malnutrition (Ali, 2020). Zinc deficiency in the human body may lead to some health issues like growth, a non-functioning immune system, the chance of infection, DNA injury, and various cancers (Zaman *et al*., 2018). Calcium performs a significant role in development of teeth and bones (Pravina *et al*., 2013). Deficiency of calcium is an important health problem that prevails worldwide. Inadequate intake of calcium may lead to diseases such as osteoporosis and rickets (Sharma *et al*., 2017). Selenium is the most important nutrient for animals and humans as it forms seleno-proteins such as thioredoxen reductases and glutathione peroxidase (Kaur *et al*., 2014). In developing countries women and children are more affected by micronutrient or vitamin deficiencies (Uchendu *et al*., 2012).

Human diet encompasses the energy, vital micronutrients needed, to fulfil dietary requirements of the human body (Halimi *et al*., 2020). Wheat accounts for more than half of the daily calorie consumption in poor countries (Chattha *et al*., 2017).

2. Approaches to relieve deficiency of Zinc and Iron

2.1. Change of diet

Change of diet is the first approach to reduce the iron deficiency as it can be easily performed. To fulfil iron requirements dietary change includes balanced recipes (Man *et al*., 2021). Daily zinc consumption by wheat-derived processed meals is an effective method of reducing zinc deficiency (Wang *et al*., 2020). Animal products including butter, liver, milk and eggs are the major sources of vitamin-A. Human and animals obtain vitamin-A from carotenoid chemicals that occur in colored fruits, plant leaves and vegetables (Dubock, 2017).

2.2. Supplementation

Humans get calcium from dietary intake and calcium supplementation. Calcium supplementation has been taken by millions of people including men, women and children in order to improve their skeletal health (Li *et al*., 2018). Zinc supplementation plays a significant role to reduce stunting and improves the child growth. Trials examining effects of zinc supplementations on child growth had beneficial effects on child growth (Liu *et al*., 2018). Oral iron supplementation is regarded as the best treatment in order to reduce iron deficiency in women (Stoffel *et al*., 2020).

3. Biofortification

Biofortification is a natural process for enhancing micronutrient availability in food crops as a strategy against hunger and malnutrition (Andersson *et al*., 2017). It is an agricultural approach which enhances the absorption and accumulation of essential micronutrients that involves increasing micronutrient availability in staple foods by using techniques such as conventional breeding, agronomic and transgenics (Agrawal *et al*., 2020; Wu *et al*., 2015). Biofortification is often times considered the additional beneficial approach for serving populations with worldwide zinc shortage, compared with regulating zinc supplements to individuals in backcountry (Velu *et al*., 2014).

3.1. Techniques of Biofortification

Biofortification of essential nutrients into staple crops may be obtained through major biofortification approaches namely transgenics, conventional breeding, and agronomic, including use of biotechnology, fertilization, and breeding of crops. Many crops including rice, pea, tomato, wheat, banana, potato and maize are biofortified by using agronomic, transgenic and conventional breeding approaches (Garg *et al*., 2018).

3.2. Biofortification through organic manure

To enhance plant growth and improve biofortification of iron in cereals, chemical fertilizers and organic improvements are used (Alburquerque *et al*., 2015). Poultry manure, has high micronutrient composition and nitrogen contents. High nitrogen enhance iron (Fe) absorption in wheat crops by improving Fe activity also induces plenty of iron transporter proteins like yellow stripe 1(YS1) in root cell the membrane (Ramzani *et al*., 2016). According to evidence nitrogen (N) fertilizers are an important factor in increasing the iron concentration in wheat grain. So, plant nitrogen condition must be paid extra attention (Aciksoz *et al*., 2011).

A major disadvantage of fertilizer approach for biofortification is its price which may not have an economic return (Joy *et al*., 2016). Apart from this their high dose may lead to several environmental issues including ozone layer depletion, global warming and pollution (Parsaad and Shivey, 2020). Nitrogen fertilizers are not acidic but their enormous use makes the soil acidic. An example of that is the soil in the Great Plains becoming acidic due to constant uses of fertilizers which results in low productivity of crops (Schroder *et al*., 2011).

3.3. Biofortification through an agronomic technique

Biofortification through agronomic approach demands the solid practice of nutriments to momentarily enhance the nutrient level of staple foods and the intake of these foods makes the human health better (Cakmak *et al*., 2017). Micronutrient biofortification through an agronomic approach is a different scheme to enhance the zinc and iron value in wheat, and rice crops (He *et al*., 2013; Ackisoz *et al*., 2011).

The nutrient value of barley has also been improved by using different forms of organic and inorganic biofertilizers. The iron and zinc value was improved by using combination of biofertilizers with inorganic fertilizers (Maleki *et al*., 2011). Agronomic biofortification of wheat regarding zinc is usually supposed to be quite cost-effective as it involves frequent annual applications (Velu *et al*., 2014). Therefore, this technique for wheat with zinc deficiency has not been widely accepted (Joy *et al*., 2015).

3.4. Biofortifcation through transgenic approach

Biofortifcation through transgenic technique can be a sustainable and alternate way for producing biofortified foods. Genes are inserted into the genome of a crop to produce micronutrients in the transgenic method. Transgenic technique also useful in that sense when a functional gene has been discovered it can be used for selecting different plants. Various useful genes are carotene desaturase, phytoene synthase (PSY), ferritin and nicotinamide synthase (Garg *et al*., 2018).

Banana is the fourth major food crop and has been mainly selected for beta-carotene, which have been attained by producing transgenic banana by the expression of PSY gene (PSY2a) of Asupina Banana (Waltz, 2014). Sorghum is the significant food crop. It has been selected to increase provitamin A by expressing Homo188-A gene (Lipkie *et al*., 2013). Maize is a significant food crop in advanced countries. Their mineral, vitamins and protein quality has been improved through genetic engineering (Decourcelle *et al*., 2015). By expressing the phytase gene in Barley phytase action has been enhanced in order to enhance the concentration of Fe and Zn (Holme *et al*., 2012). Iron availability in rice also has been increased by decreasing anti nutrient like phytic acid (Hurrell and Egli, 2010). A bacterial PSY and carotene desaturase gene has been expressed to increase the micronutrient provitamin-A in the wheat (Wang *et al*., 2014).

3.4. Biofortifcation through Conventional Breeding

Conventional breeding is most believed approach of biofortification to develop required agronomic traits and micronutrients. In this technique parent crops are crossed with receiver crops over various generations (Lafiandra *et al*., 2014). It is regarded as a significant and cheap method to improve nutrients in staple foods worldwide (Velu *et al*., 2014). While genomic selection, quantitative trait loci (QTL) mapping, and marker-assisted selection (MAS), are genomics techniques that has been used to biofortifying wheat crops globally (Saini *et al*., 2020). However, conventional breeding has been used for so many years, but it is restricted to only sexually congenial plants so rely on natural variation of desired micronutrient. For instance, variation of Fe and Zn within wheat crops and its wild species have been used to produce new cultivars with improved iron and zinc concentration. But cassava plants contain low concentration of protein therefore breeding cannot be used for biofortification of cassava (Hirschi, 2020).

Biofortified crops produced by conventional breeding are provitamin A orange sweet potato, orange maize, Fe pearl millet, Zn rice, Zn wheat; Fe bean and have been released in above 30 countries for production (Saltzman *et al*., 2017). Because of the high concentration of phenolic, colored wheat (Purple, black, blue) have been utilized in many plant breeding strategies and released various varieties in some regions (Sharma *et al*., 2018). So far there are four zinc biofortified varieties which have been revealed such as Zincol 2016, Zinc Shakti, HPBW-01 and WB020 which are enriched with zinc content approximately 25%, 40%, 20% and 20%. These varieties are presently being cultivated in Pakistan and India. As zinc biofortifcation of wheat has been successful but no iron biofortified variety has been developed through breeding to date (Saini *et al*., 2020).

3.5. Microbe-based Biofortification

In biofortification of micronutrients and macronutrients of staple crops, growth promoting microorganisms, also play significant role by several processes including N fixation, transformation, phosphorus immobilization, and siderophore production (Khan *et al*., 2019). Plants nutrient bioavailability is affected by both rhizospheric and endophytic microorganisms but endophytic microbes supposed to be much more important for increasing iron and zinc absorption and translocation, because they can indirectly affect the metal transporters regulation (Weyens *et al*., 2013).

Endophytes from bacteria and fungi have been associated with the biofortification of wheat and rice crops with micronutrients including iron and zinc (Abaid-Ullah *et al*., 2015). Species such as *Bacillus subtilis* and *Arthrobacter* have been effective in improvement of zinc concentration to about 75% in zinc deficit soils. In wheat crops, iron content has been improved by using *Enterococcus hirae* and *Arthrobacter sulfonivorans* species (Singh *et al*., 2018). This approach is also believed to enhance plants resistance to salinity, drought, metal and pesticide toxicity, also providing growth regulators, nutrients, and improving ethylene induced stress by, 1-aminocyclopropane-1-carboxylate (ACC) deaminase synthesis (Singh and Singh 2017).

4. Methods to enhance up regulation of iron and zinc uptake

4.1. Up-regulation of Zinc and Iron transporters

Nutrients absorption and translocation are two distinct mechanisms; although nutrients uptake efficiency is good in some crop genotype, nutrients translocation from shoot to seed and root to shoot is low (Singh *et al.,* 2018). Therefore nutrient translocation and upregulation is a critical process, that must be regulated in order to enhance the nutrient concentration of the plant's consumeable parts (Singh and Prasanna, 2020). Since zinc cannot pass across cell membrane, it must be carried into the cytoplasm by several zinc transporters. In the last few years plants have been found to have a number of metal transporters. These transporters are natural resistance-associated macrophage protein (NRAMP) family, iron-regulated transporter (IRT)-like protein (ZIP), P1B-ATPase family, zinc-regulated transporter (ZRT) and cation diffusion facilitator (CDF) family (Li *et al.,* 2013). The zinc ions are carried out from cell membranes to cytoplasm through ZIP and IRT transporters (Krishna *et al.,* 2017).

4.2. Reduction of anti-nutritional factors in grains

Compounds that inhibit the nutritive value and food intake of plants or plants products used as human foods are known as anti-nutritional factors (Thakur *et al.,* 2019). Phytate is a key anti-nutrient that chelates calcium and other nutrients including iron, zinc and copper, affecting their absorption. Other anti-nutrients, including oxalates, and polyphenols are thought to decrease the absorption of minerals in foods (Kaushik *et al.,* 2018). Various ways has been devised to lower the amount of phytic acid, in food grains and enhance the nutritional value of crops that have become deficient as a result of these anti-nutritional factors. Genetic improvement is one of them, as are numerous pre-treatment processes like as germination, fermentation, soaking, and phytase enzyme treatment of grains (Gupta *et al.,* 2015).

5. Conclusions

Deficiency of micronutrients results in many diseases worldwide. Therefore, in developing countries food fortification is an essential and important approach to fight with malnutrition. To address the hidden hunger some techniques such as fortification, biofortification and supplementation plays a significant role. The absence of key nutrients and minerals are main causes of malnutrition. Vitamin-A, Zn and Fe are three key micronutrients identified by the WHO as being most deficient in people in poverty. Among various methods biofortification is a simple, crops based and cost effective method which play significant role in solving problem of nutrient deficiency. It is based on the techniques including genetic manipulation, breeding and application of fertilizers. Also with plant breeding and agronomic biofortification, considerable attempts to involve microorganisms as partner in such techniques are required. However a scientific method alone is insufficient to address the problem of micronutrient deficiency, although requires a strategic plan. There is need to raise public awareness about the benefits of food diversity and to find useful solution for improving people's dietary needs.

6. Author's Contribution

Usaal Tahir designed the study. Maham Mazhar and Ammara Moon did the article write up. Maryam Zameer was responsible for the overall formation and finalization of the article.

8. Conflict of Interest

Authors have declared no conflict of interest.

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Annexure 1

Table 1: Tabulation of crop, micronutrient and growing country

Sr.	Crop	Micronutrient	Country	Reference
1.	Wheat	Zinc and iron	India and China	(Yadava *et al.*, 2017)
2.	Rice	Iron and zinc	USA, India, China, Bangladesh	(Singh and Prasanna, 2020)
3.	Pearl millet	Zinc and Iron	India	(Rai *et al.*, 2014)
4.	Sweet potato	Provitamin A	China	(Saltzman *et al.*, 2013)
5.	Cassava	Provitamin A	Nigeria	(Njok *et al.*, 2015)
6.	Potato	Iron and Zinc	Rwanda	(Singh and Prasanna, 2020)
7.	Sorghum	Iron and Zinc	India	(Ashok *et al.*, 2013b)
8.	Beans	Iron	Rwanda	(Haas *et al.*, 2016)
9.	Banana	Provitamin A	Uganda	(Schnurr *et al.*, 2020)
10.	Maize	Provitamin A	Zimbabwe, Malwi, Rwanda, Nigeria, Ghana, Tanzania, Zambia and Mali	(Pixely *et al.*, 2013)
11.	Lentil	Iron and Zinc	Bangladesh, Ethiopia, Nepal and Syria	(Kumar *et al.*, 2016)

Table 2: Anti-nutritional factors present in some plant crops (Thakur *et al.*, 2019)

Sr.	Crops	Anti-nutritional factors
1.	Wheat	Tannis, phytic acid, saponins, polyphenols
2.	Barley	β- glucans
3.	Peas	Oligosaccharides, tannis, lectins
4.	Lupin	Alkaloids
5.	Soybean meal	Phytic acid, lectins, trypsin inhibtors, oligosachrides
6.	Sunflower meal	Tannins