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## Wheat Biofortification: Agricultural investment to refrain malnutrition especially in developing world

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### Abstract

Malnutrition is a reticent emergency and persistent threat to the future of children especially in developing countries. 27 percent of all children globally are stunted which is the ultimate cause of malnutrition. Global Nutrition Report 2018, insights 22.7% of world's children under the age of five are stunted. 9.7% of female (aged 20-29) and 5.7% of adolescent girls (aged 15-19) are underweight. These figures signified the unacceptably high burden of malnutrition on world. Biofortified staple food seems the best solution to fight and overcome malnutrition problem worldwide. Breeding for nutrient dense wheat is a sustainable and cost-effective approach for availability of micronutrients and further, adoption of biofortified wheat varieties by farmers and release of new biofortified varieties in public domain will be helpful in fighting against malnutrition globally which also damages the world economy.

**Keywords:** Biofortification, malnutrition, micronutrient deficiency, conventional and biotechnological approach, wheat

### Introduction

With the millions of births every year, the burden of micronutrient deficiency is also increasing especially in developing countries. One in three people in the world suffer from micronutrient deficiency which leads to negative health consequences <sup>[1]</sup>. Malnutrition or hidden hunger causes extremely negative effects on the world economy <sup>[2]</sup>. The term malnutrition covers 2 broad groups of conditions. One is 'undernutrition'-which includes stunting (low height for age), wasting (low weight for height), underweight (low weight for age) and micronutrient deficiencies or insufficiencies (a lack of important vitamins and minerals). The other is overweight, obesity and diet-related non-communicable diseases (such as heart disease, stroke, diabetes and cancer). Women have a higher prevalence of obesity than men, at 15.1% compared with 11.1% <sup>[3]</sup>.

Worldwide the progress in addressing underweight and anemia among females has been extremely slow while overweight and obesity among adults is getting worse. Underweight has been declined slightly since 2000 though not significantly-9.7% of female (aged 20-29) and 5.7% of adolescent girls (aged 15-19) are still underweight and anemia has risen slightly to 32.8% <sup>[3]</sup>. It is the leading risk factor in poor health outcomes in Sub-Saharan Africa and fourth highest in Asia <sup>[4]</sup>. Fe and Zn are major micronutrients deficient in the world <sup>[5]</sup> and the main reason is dependency on cereal based diet which itself has low nutrient concentration. People need iron for vital functions. Infants are at high risk of iron deficiency and iron deficiency anemia <sup>[6]</sup>. There is urgent need of multiple complementary strategies to address micronutrient deficiencies at global level <sup>[7]</sup>.

Cereal-based foods have been a staple dietary source for the world's population for centuries. Cereal grains contain the macronutrients (protein, fat, and carbohydrate) required by humans for proper growth and maintenance, contributing approximately 70% and 50% of the total calories and protein, respectively <sup>[8]</sup>. They also provide 20% of magnesium and zinc, 30-40% of carbohydrate and iron, 20-30% of riboflavin and niacin, and over 40% of thiamine in the diet <sup>[9]</sup>. Wheat is a major staple food crop for 40% of the world's population <sup>[10, 11]</sup>. Wheat, like many other staple cereals, contains sub-optimal levels of the essential micronutrients particularly iron and zinc <sup>[12]</sup>. Because of its widespread geographic distribution, acceptance, stability and versatility, wheat flour is a suitable vehicle for delivering micronutrients to human being <sup>[13]</sup>.

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Generally, in today's commercial wheat cultivars, the grain Zn and Fe concentrations are 20-35 mg kg<sup>-1</sup> which is not sufficient for human nutrition where wheat constitutes the main source of essential minerals. It has been suggested that in order to have a measurable biological effect on human health, concentrations of Fe and Zn grain should be increased by at least 10 to 25 mg kg<sup>-1</sup> in the present cultivars [12, 14]. Biofortification targets for Fe and Zn in the wheat grain are approximately 60 and 40 mg kg<sup>-1</sup>, respectively [15].

Biofortification strategy had been ranked fifth among other most cost-effective solutions to address global challenges such as reducing hidden hunger [16]. In this approach, the density of minerals and vitamins in food staples eaten widely by the poor may be increased either through conventional plant breeding or through the use of transgenic techniques [17]. It takes just one upfront investment; once the trait is bred in, it will be retained in successive generations. In India, the burden of disease that arises due to iron deficiency could be reduced from 19% to 58% by crop biofortification [18]. Global biofortification can have a significant role in reducing the burden of micronutrient malnutrition in the developing world in a highly cost-effective manner [19]. Hence, plant breeding/transgenic technologies are promising and long-term strategies to overcome human malnutrition by releasing new cultivars with enhanced levels of Fe and Zn to the target

regions. Wheat biofortification is a better alternative for genetically enhancing the nutritional content of wheat crop and increasing the bioavailability of the micronutrients in humans [20].

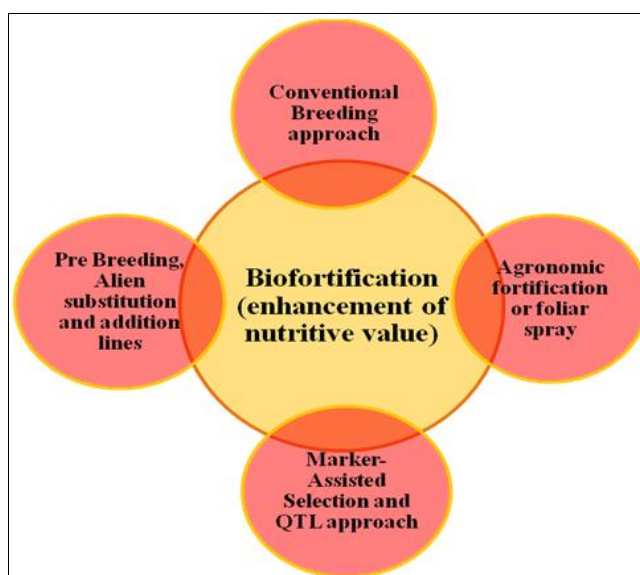
### Ways for Biofortification:

#### A. Conventional method

The breeding steps include-

1. Identification of a useful genetic variation and the promising parents for hybridization,
2. Long-term crossing and back-crossing activities for incorporation of gene in the background of the recipient parent,
3. Stability and expression of the target traits (e.g., high grain Zn and Fe concentrations) across the different environments,
4. Adaptation of the newly developed biofortified genotypes over a range of crop and soil management practices applied in the target regions or countries.

Breeding for biofortified crops offers a sustainable solution to malnutrition problems by exploring natural genetic variation. Breeders selectively breed nutritious cultivars of major staples, rich in Zn and Fe concentrations and with substances that promote the bioavailability of micronutrient (Figure 1).



**Fig 1:** Bio fortification tools for improving protein, iron (Fe) and zinc (Zn) content in the cultivars.

Authors at CCS HAU, Hisar, Haryana are working on wheat bio fortification using conventional approach using the genotypes containing Fe and Zn > 40 ppm and > 45 ppm respectively (Table 1) as donor parents and high yielding cultivars as recipient parents in crossing programme. RILs have been generated for QTL mapping for high Fe and Zn content.

**Table 1:** Best performing CCSHAU genotypes for Fe and Zn

Sr. No.	Fe > 40 ppm	Zn > 45 ppm
1	WH1136	WH1136
2	WH1164	WH1164
3	WH1179	WH1189
4	WH1183	WH1193
5	WH1193	WH1195
6	WH1063	WH1063

#### B. Exogenous application of micronutrients (Fortification)

This includes application of FeSO<sub>4</sub> and ZnSO<sub>4</sub> by foliar spray to the crop which may lead to increased availability of these elements in the grain which ultimately improves the nutritional value of cereal. Bread wheat bio fortification for high Fe and Zn by exogenous application of micronutrients or trace mineral element like Fe and Zn at 4 mg kg<sup>-1</sup> has significant effect on plant available nutrients and nutrient concentration in wheat straw and grain [21]. Kumar *et al* 2016 [22] concluded that when an increase in grain Zn, Fe or Mn concentration in grain is required, in addition to soil fertilization, foliar application would be beneficial. Soil Zn deficiency in major wheat growing areas leads to inherently low grain Zn concentration and is considered as a lead factor in low Zn uptake in human. A foliar Zn application as compared to soil Zn applications results in remarkable increases in grain Zn concentration in wheat. The application

of Zn and Fe encompassed fertilizers (i.e. agronomic bio fortification) is a short-term solution and represents only a parallel path to breeding [23].

### Screening of germplasm, wild relatives and Prebreeding

Identification of useful genetic variability for enhancement of iron and zinc in wheat using conventional approach, major emphasis has been given on screening of elite germplasm of wheat including wild relatives, progenitor and non-progenitor of cultivated wheat species [22]. Germplasm screening of wheat and its wild relatives has revealed substantial genetic variation for grain Fe and Zn concentrations (Table 2). The source of genetic variation is essential for the next breeding generations. Identification of potential donors for the trait and understanding of how micronutrients are accumulated in the plant/crop is essential [24]. If variation is present in the strategic genepool, prebreeding is required. If variation is present in the tactical genes pool, the materials can be used directly to develop competitive varieties [11]. Individuals with

the highest Zn and Fe concentrations are progenitors of wheat such as einkorn wheat and wild emmer wheat, and landrace. Unfortunately, little variation exists in improved adapted wheat varieties for Zn and Fe content. Researchers, therefore, focused on a more in-depth evaluation of wheat landraces, then, the secondary gene pool, i.e. tetraploid and diploid progenitors of hexaploid wheat, was evaluated for micronutrient concentration. *Aegilops tauschii*, *Triticum monococcum*, *Triticum boeoticum* and *Triticum dicoccoides* were among the most promising sources of Fe and Zn grain concentration. Among the hexaploids wheats, spelt wheat found to have high grain Zn and Fe and is easily crossable with *Triticum aestivum* [25]. Pre-breeding activities designed to identify desirable characteristics and/or genes from unadapted materials that cannot be used directly in breeding populations and to transfer these traits to an intermediate set of materials that breeders can use further in producing new varieties for farmers.

**Table 2:** Mean and range of Zn concentration in bread wheat, durum wheat, and their wild relatives in different studies.

Germplasm	Number of genotypes	Zn concentration (mg/kg)		Source (Year)
		Mean	Range	
<i>Triticum boeoticum</i>	12	89	45–177	2000 [26]
Hard red winter wheat	1605	33.3	14.3–74.4	2002 [27]
<i>Triticum dicoccoides</i>	518	61	30–98	2004 [28]
Spring and winter wheat	66	28	20–32	2007 [29]
Bread wheat	150	21.4	13.5–34.5	2009 [30]
Einkorn wheat	5	22.4	20.1–27.8	
Emmer wheat	5	22.8	15.8–30.3	
<i>Triticum spelta</i>	5	22.9	16.8–28.0	
Winter wheat	137	31.6	11.7–64.0	2009 [31]
Durum wheat (old)	10	36.4	33.7–41.4	2009 [32]
Durum wheat (modern)	57	33.9	28.5–46.3	
Durum wheat (advanced)	17	32.7	29.1–40.9	
Wild emmer wheat	19	77.0	39–115	2010 [33]
Winter and spring wheat	321	39.45	37.7–41.2	2010 [34]
Durum wheat	50	38	31.9–44.1	2017 [35]

### Alien addition and substitution lines

Biofortification through a combination of conventional and molecular breeding methods is the most feasible, economic, and sustainable approach. For disease resistance, quality, enhancing micronutrient content like high grain Fe and Zn, protein, wider adaptation, high yield etc. wide crosses can be performed. In alien addition lines, addition of chromosomes of a wild species (foreign species) to the normal compliments of a cultivated species occur while in alien substitution, there occurs replacement of one pair of chromosomes of a cultivated species with those of a wild donor species. Tiwari *et al* 2010, confirmed the substitution of chromosomes 2S and 7U for their homoeologues of the A genome, suggesting that some of the genes controlling high grain micronutrient content in the *Ae. Kotschy* accessions are on these chromosomes [35].

### QTL identification and marker breeding:

Molecular markers have been used to identify the genetic regions involved in grain Fe and Zn content in several species of crop plants. Gpc-B1 (GRAIN PROTEIN CONTENT - B1) is wheat QTLs linked with increased grain protein, zinc and iron content [36]. In a study conducted by Xu *et al.*, 2012 for mapping of QTLs for grain zinc, iron and protein concentration of wheat across two environments, nine additive and four epistatic QTLs were identified for Fe and

Zn. They mentioned that *Xgwm154* is the marker closely linked to the QTLs and may be useful in wheat biofortification breeding by MAS [37]. QTLs for Fe and Zn concentration were identified in two wheat recombinant inbred line (RILs) populations in wheat [30]. Thirty eight polymorphic SSR makers known to be linked with micronutrient content in bread wheat (as stated in literature) showed positive results in various crosses attempted [38]. Hence marker-assisted breeding is useful in improving the efficiency of selection in advance generation leading to improve characters with low heritability. Recently, significant genomic regions have been identified through QTL mapping and Genome Wide Association Mapping approach for better understanding of genetic architecture of grain accumulation in cereals [93].

### Inheritance, Genotype x Environment interaction for grain iron and zinc concentrations in wheat along with association with the grain yield

The inheritance of nutritional traits appears to be mostly quantitative, influenced by the environment, but more specific to source genotypes. Reports are there showing significant and positive association among Fe and Zn content in wheat and significant genotype × location interactions for Zn and Fe in wild and improved wheat cultivars [15, 33, 40, 41]. Although negative correlations between the concentrations of Fe and Zn

in grain and grain yield were reported in many studies [32, 33, 42]. Quantitative inheritance along with low heritability of Fe and Zn (grain) and the large genotype x environment interactions associated with them have negatively affected the biofortification programme.

### Bioavailability

The bioavailability of micronutrients in unfortified wheat cultivars in developing regions is typically low, which raises questions about the efficacy of these crops to improve population micronutrient status [43]. So, it is recommended to first evaluate the micronutrient bioavailability of biofortified cultivars in order to derive lessons that may help direct plant breeding and to infer the potential efficacy of food-based nutrition interventions. The bioavailability of iron and zinc can be increased by reducing the concentration of inhibitors which hinder the human absorption of dietary iron and zinc or increasing the concentration of enhancers which favor iron and zinc absorption. Dietary fiber, Phytic acid (PA), tannins and calcium are the most potent inhibitors while organic acids are known to promote iron absorption [44, 45]. By reducing the phytate content, bioavailability of iron and zinc content could be enhanced significantly.

### Ground level Problems incurred in the biofortified food:

In the past 50 years, the fundamental objective of wheat breeding programs has been to raise productivity by developing high yielding cultivars. This has been brought about largely by choosing the lines with disease resistance, dwarf & semi-dwarf height, enhanced biomass and grain yield to biomass ratio, among other agronomic features. Moreover, yield enhancement is essential to feed the world's explosively widening population. Although, the nutritional values of staple crops, especially micronutrient concentrations and protein quality, is equally important but often neglected. There may be problems in getting biofortified foods to be preferred if they have different attributes to their unfortified counterparts. Lack of price incentive is also the ill cause behind the better outcome. Creating awareness among resource-poor farmers in developing countries will enhance adoption of biofortified cultivars to combat the malnutrition.

### Conclusion

Biofortification is a feasible and cost-effective means of delivering micronutrients to those that may have limited access to diverse diets and other micronutrient interventions [12]. Studies have shown that lots of genetic variation is present for micronutrient contents in wild relatives and unexploited germplasm which needs to be exploited substantially. Further, creating awareness among local population and resource-poor farmers will help in adoption of effective agronomic applications for increased soil and crop micronutrient contents as with the industrialization of wheat processing, the demand for specific and consistent qualities has been increased in wheat. The differentiation of wheat products may create extra employment along value chains, this may, in turn improve the incentives for farmers to adopt new varieties with enhanced grain quality traits, ultimately building higher world economy by preventing losses incurred due to malnutrition.

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