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REVIEW ARTICLE
AGRONOMIC BIOFORTIFICATION OF FOOD CROPS WITH MICRONUTRIENTS

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Abstract

Micronutrient malnutrition in human population can be very well strategized by using Agronomic biofortification, which remains a key tool for healthy populations and reduced mortality. The approach related to different micronutrient is reviewed. The efficacy studies of various elements like iron (Fe), iodine (I), colbalt (Co), Zinc (Zn) and Selenium (Se) and its concentration in different parts are also viewed. All the five micronutrients were equally important and contribute for better human health nutrition in food crops. Eventhough some nutrients have their own restrictions in the plant, but due to various factors (like, temperature, climate, time of applications, crop adaptability, etc), few micronutrients have played a significant role in the food nutrition making easier access in the plant edible parts by its applications.

Effective studies were carried out widely with Fe, I, Co, Zn and Se, which is found more valuable and highly resourced, against food crops in developing countries. Few among them are closely related to plant – animal interactions (Co and Se), rather than human alone. For Zn, combination of soil and foliar (two applications) were most effective than individual application. It is predominantly efficient when applications were tried in deficient soils, with yield increase and Zn content in grain during harvest. Also tolerance to Zn deficient soils is achieved through genetic studies. It is important to note that foliar application of Zn at the time of mid booting stage or early milking stage is important and effective. Both soil and foliar is found to be effective if Se is applied. When compared sodium selenate is more effective than selenite for soil application.

Keywords: biofortification, iron (Fe), zinc (Zn), iodine (I), cobalt (C), selenium and micronutrients

Introduction

Due to dietary deficient nutrients, high mortality of 50% deaths occurs across the globe. This is due to high dependence of cereal crops by the population in developing countries. Lack of awareness about the nutritional crops and in food system has lead to a huge decline in promoting such need based crops for the specified regions. This has also taken a different route in the food systems, which fail to deliver optimum nutrition to the populations. Agriculture has a key role to play in both food and nutritional security. As of now, agriculture was viewed and considered solely to improve the productivity rather than it should be focused on sustainable nutrition food system. The important factor is the bioavailability of micronutrients in food crops (Welch and Graham, 2004), which is a major task for the breeders and agronomist to work for the healthy environment.

Biofortification in staple crops has been initiated in three different ways; by Transgenic, Plant Breeding and Agronomic measures to achieve the micronutrient levels in edible parts of the crop. This has been designed as a special strategy to target the dietary deficiencies among the rural poor. Many such similar methods have also been tried to increase dietary diversity, process in fortification and supplementation for livestock, etc (Lyons *et al.*, 2003; Haug *et al.*, 2007).

It is of paramount importance to attract and retain the producers and consumers by adopting the biofortification strategy without compromise on agronomic applications. In general farmer will not be interested in high Fe content in wheat, if he has to compromise on yield (Bouis and Welch, 2010; Cakmak *et al.*, 2010). There is a major concern about the consumers, who shows little interest in identifying the levels of minerals and nutrients, which affects the branding aspects (Pfeiffer and McClafferty, 2007). Agronomic biofortication may be more effective for Zn, Se, and I (Lyons *et al.*, 2008).

In case of genetic engineering, biofortification with micronutrient is proved by the high carotenoid golden rice (Potrykus, 2003). Similarly under conventional breeding approach, crops like sweet potato, banana and cassava were tried for pro-vitamin A carotenoids (Chavez *et al.*, 2000, 2005; Bouis and Welch, 2010; Genc *et al.*, 2010).

The process involving of transgenic and breeding may take its own time, which remains a long term process, whereas agronomic biofortification is a short term solution to the malnutrition problem. Because readily available micronutrients like Fe, I, Co, Zn, Se will represent as a complimentary to genetic biofortification. Also the possible efficiency and their role of individual micronutrients will be discussed.

Agronomic biofortification

Iron

Iron is required by humans for its proper functioning of muscle and brain tissues. It also acts a carrier of oxygen from lungs to various tissues in the form of red blood haemoglobin. The anaemia is the major problem for iron deficient people. At an average the requirement of iron for non-pregnant and non-lactating women its is 1460 µg/g and for a age group of 406 yr old it is 500 µg/g per day. Iron nutrient in grain is achieved through biofortification in crops like wheat, Pearl millet and lentil which is as; 28.0-32.0 ppm (max >38.0) and 45.0-50.0 ppm (max >70 ppm) and 45-50 ppm (max >62 ppm) respectively (Yadav *et al.*, 2018).

Similar findings were also reported that ancient wheat have higher concentrations of micronutrients than modern wheat with, Fe concentration ranges from 40 to 100 mg/kg

(Cakmak *et al*., 2004; White and Broadley, 2005, 2009; Cakmak *et al*., 2010).

On a precautionary note, Fe is drastically removed during milling process in wheat flour manufacturing, leaving more than 50% down against the target of 40 mg/kg Fe. Under agronomic measures, in general Fe is proved to be difficult in biofortification due to its rapid conversion of ferrous to ferric. The ideal Fe, provided in chelate form is more available and effective against Fe deficiency, which is quite expensive and found to be marginally better in increasing Fe concentration in grain. Foliar application of ferrous sulphate $(Fe_2So_4)_3$ has shown a better effect than soil application in increasing Fe concentration in cereals (grain), which also enhances the yield of crops. Fe when used under process fortification has been successful for Fe fortification which includes rice, fish, soy sauce, wheat flour & maize flour, milk and infant formulas. The easiest way to reach rural poor is by using a large scale fortification of flour or salt.

Iodine

Iodine reduction causes thyroid gland enlargement and stomach cancer. The Source of iodine for human diet should be achieved with a strategy through biofortification of crops with cost-effective, well accepted and in bioavailable form. Iodine required for different age groups are; young children (1 to 8 years) – 90 µg d^{-1}, older children (9 to 13 years) – 120 µg d^{-1}, adults > 14 years $^{-1}$ 150 µg d^{-1}. Pregnant women are prone to iodine deficiencies. They require 220 and 270 µg d^{-1} for pregnancy and breastfeeding. One third to one half of the population are affected by iodine (other micronutrients too) deficiencies. In order to counteract iodine deficiency (apart from salt iodination), the cost effective and most promising agronomic biofortification can be effectively used. Some corrective measures like fortification of livestock feed and biofortification of plants plays a major role.

Agronomic biofortification has numerous advantages like; Iodine supplied to plants is taken from soils. A biochemical action takes place during the conversion, when process of incorporation into tissues happens. Iodination in irrigation water is an indigenous technology which is easier, cost-effective and has a direct benefit to the plants. Fertigation is much useful for leafy vegetables, because of its faster solubility. Application of potassium iodate @ 40 µg dm−3 at 2-day intervals for 4 weeks have shown a significant increase in iodine content in vegetables (Ujowundu *et al*., 2010). Agronomic Biofortification shows a positive sign in leafy vegetables than root vegetables due to iodine movement in plant parts.

In contrast with consumption and human benefits, tomato crop plays a key role in iodine biofortification, because of its presence in the fruit up to 10 mg kg^{-1} of fresh weight. The concentration of iodine in tomato is more than sufficient to the daily human consumption of 150 µg (Cakmak *et al.,* 2017). Many promising results have been obtained with iodine biofortification due to its enhanced levels of bioactive compounds. It was viewed that concentrations of glucose, total sugars, and fructose were increased in biofortified carrot (Li *et al*., 2016). Inorder to determine the beneficial and toxin iodine concentrations, higher amount of iodine salts to be utilized for biofortification. Inorder to increase the iodine concentration in plant tissues, iodine salts of <50 mg kg^{-1} can be applied (Gonzali *et al*., 2017). The plant growths are not affected in soil when iodine is applied upto 10 mg kg^{-1} (Caffagni *et al*., 2012).

Many experiments have successfully proved that for effective biofortification, spraying in leaves with iodine salts was found to be best. Similar foliar spraying contribution was effective in lettuce cultivation than soil application with positive impact on crop quality (Lawson, 2016). In order to produce more nutritious crops, seaweed manure application was done as an iodine biofortification.

The significant increase in grain iodine concentrations was found in wheat, brown rice and maize, when foliar application was done. This increase was noticed in endosperm part of wheat grain and in polished rice.

Cobalt

Cobalt is an essential element for the synthesis of vitamin B12, which is required for human and animal nutrition (Young, 1983 and Smith, 1991). The requirement of cobalt in nutrition is essential for animals rather than plants. It is important for ruminants that without Co, rumen bacteria are unable to synthesise the cobalt-containing vitamin B12. Cobalt is safe for human consumption and up to 8 mg can be consumed on a daily basis without health hazard (Young, 1983).

Cobalt is a necessary element to legumes. For legumes, Cobalt at lower doses enhances nodule formation, nitrogen fixation, growth, seeds and oil yield of oilseed and leguminous seeds. All ruminants (including sheep, cattle and goats) require cobalt in their diet for the synthesis of vitamin B12. Vitamin B12 is essential for energy metabolism and the production of red blood cells. Cobalt deficiency in soils can cause vitamin B12 deficiency in livestock.

Mankind also adds cobalt to the soil, primarily through three mechanisms. The major mechanism is use of cobalt salts, e.g. cobalt sulphate, as a feed additive to keep cattle and crops healthy in areas where there is insufficient natural bioavailable cobalt. Smaller amounts of cobalt also enter the soil from the airborne transport of particulate emissions and application of sewage sludge onto fields.

The growth parameters of soybean plants (Length of shoot, Length of root, number of nodules/plant, number of leaves/plant number of branches/plant, fresh weight and dry weight of shoots and roots) at a higher levels were obtained when 12 mg kg^{-1} cobalt was applied. Increase in cobalt concentration above 12 mg kg^{-1}, significantly reduces all growth parameters. Application of 12 mg kg^{-1} cobalt had a significant primitive effect on the macronutrients (N, P and K) and micronutrients (Mn, Zn and Cu), with those obtained by Boureto *et al.* (2001). Thus ruminants consumed biofortified Co plants, has a direct impact on humans vitamin B_{12}.

Zinc

Zinc is a mineral element, involved in major role in maintaining cellular integrity and immune system. Zinc deficiency occurs mainly of low dietary intake and consumption of cereal foods. Low zinc is observed in cereal based foods than animal based foods. Majority of the rural people are based on cereal based foods because of their poverty and cultural adaptability. The average zinc required for non-pregnant and non-lactating women is 2960 µg/g and for children of 4-6 yr old it is 1390 μg/g per day. There found to be a significant difference between the actual Zn concentration and the target of human (40-50 mg kg^{-1}).

Zinc nutrient in grain achieved through biofortification in crops like rice, wheat, pearl millet and lentil, which is as; 12.0-16.0 ppm (max >20.0 ppm), 30.0-32.0 ppm (max >40.0 ppm), 30.0-35.0 ppm (max >40.0 ppm) and 35-40 ppm (max >50.0 ppm) respectively (Yadava *et al.,* 2018).

On a cautionary note, during wheat grain milling process, it removes the Zn rich parts and leaves the endosperm behind. Due to which, wheat flour contains only 5-10 mg Zn per kg. This is where the required concentration for human is not met.

Two different sources of Zn in the grain is noticed, they are; Zn that is absorbed continuously by roots from soil and translocated into grain and that is deposited in vegetative tissues and translocated into reproductive stage. These two sources vary depending on soil conditions, available soil Zn, Water conditions, Period of applications, etc., which decides the Zn accumulation in grain. Only under certain conditions, it can effectively be monitored and utilized for better crop productivity and quality of Zn available. Two such practices are in agriculture, mainly soil and foliar application of Zn. Timing of foliar application is a critical factor, which determines the accumulation of Zn in grains. It can be performed by using Zn So_4 or Chelated forms of Zn (eg., Zn-EDTA).

In general foliar application of Zn is effective at a late growth stage. Therefound to be increase in whole grain Zn through soil and foliar applications. The results were noted based on the predominantly consumption of the food products in certain countries (Yilmaz *et al.*, 1997; Cakmak, 2008). The foliar application of Zn was effective against reducing phytate concentration in grain (Erdal *et al.*, 2002; Cakmak *et al.*, 2010). The bioavailability of Zn in diets is promising based on phytate/Zn molar ratio.

Rates of 25 to 50 kg $ZnSO_4$ per ha are generally used in fertilization of soils with Zn (Cakmak, 2008). Depending on the severity of the Zinc deficiency problem, a cost effective model in fertilization need to be worked out, to understand the better practices adopted in the region. At the initial stage, farmers need to be motivated about the strategy of Agronomic Biofortification use in staple crops, quality seeds and awareness about the Zinc nutrition to human health. This will bring a potential growth in fertilization (biofortification) of Zn as foliar spray or foliar + soil application, irrespective of stages it reaches for maintaining yield and helps in accumulation of available Zn in grain.

Selenium

Selenium is an essential element linked mainly to human and animal nutrition (Lyons *et al.*, 2003). Many vulnerable diseases such as cardiovascular and cancers were found due to low Se intakes. Selenium is highly suitable for agronomic biofortification process in food crops due to its quick adaptability in plant systems. Se is compatible in the wide range of pH from 5.5. – 9.0, as selenate.

When applied it gets accumulated in edible parts and conversion of seleno methionine in organic forms takes place. This is also evenly distributed in cereal grains and found abundant in milled white flour and polished rice. Effectiveness of Se on soil or foliar applications depends mainly on soil characteristics, method of basal application and time of foliar spray. Different rate of applications were noticed in combined form of basal and foliar selenate to crops (Ylaranta, 1983). Considering the overall performance, it was economical and promising when foliar spraying is done (Ylaranta, 1984). There is a direct co-relation between increase of Se in plants versus climate, organic matter, Soil pH, soil aeration, soil temperature and moisture. For cancer prevention, Se acts as a supplementation target (Combs, 2001).

Under agronomic biofortification, Se concentrations in cereal grains are drastically increased when applied in selenate form (Broadley *et al.*, 2010; Hart *et al.*, 2011). When compared to selenite, Se-enriched manure is found to be more effective in biofortifying crops, for ex., Tea (Hu *et al.*, 2002). Even with varied pH, Fe, S and organic content, sodium selenate to the soil at seeding stage was effective than post-anthesis foliar applications under drought conditions of South Australia.

Sodium selenate as foliar application of 10 g ha^{-1} was effective against maize, groundnut and soyabean (Allan *et al.*, 2014). It shows that 10 g selenate per ha have increased wheat Se concentration at varied levels of 30-100-200-500 μg Se kg^{-1} grain. Also the optimum level is recorded as 300 μg Se kg^{-1} (Lyons *et al.*, 2004). Individual Se increase will happen, if wheat meal bread is consumed over a week with fortified wheat. Agronomic biofortification (Fertilization) with sodium selenate is economical and can be achieved practically at field level in wheat, where the concentrations of Se remains high provided health benefits to the consumers from major chronic diseases.

Conclusion

The deficiency of micronutrient is prevalent among the populations. It's possible to rightly identify the micronutrients like Fe, I, Co, Zn and Se for agronomic biofortification strategies. These elements can be well used within its limits for required concentrations and such effective measures can target the consequences of malnutrition. Some of the effective measure can be taken for better biofortified promotions:

1. Enriched micronutrient germplasm to be developed either through transgenic or plant breeding for efficacy towards agronomic biofortification
2. Studies on minimizing toxicity to be undertaken
3. Standardizing nutrient availability under post processing stages
4. Various ways to be adopted for promotion to farmers, with premium price for harvest produce been recommended

A multilayered approach should be positioned, right from the research institutes (identifying suitable crops/varieties for the region) to promotion at farmers field (Suitable crop or variety, its cost effectiveness, additional benefits, etc) and to the processing unit (harvested grains, packed and sent), all should be endurable (without any deviation) for better success. Even after the proposed plan, there need to be continuous trial and error experiment, in terms of yield, nutrient presence in grain, etc., which remains a major socio-economic issues in nutritional pattern of consumer is concerned. Such multistage strategy will depend on technology efficacy and dietary effects.

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