



Review Article



Review: Biofortification of Cereals with Zinc through Agronomic practices

Shahzad Ali^{*1}, Tariq Mahmood², Sami Ullah², Zarmina Aslam³, Sohaib Nasir⁴, Rimsha Zain⁵ and Samia Zain⁵

^{*1}Department of Soil and Environmental Sciences, University of Sargodha, Sargodha 40100, Pakistan

²Department of Forestry, University of Sargodha, Sargodha 40100, Pakistan

³PMAS-Arid Rawalpindi, Rawalpindi 6800, Pakistan

⁴Department of Plant Pathology, University of Sargodha, Sargodha 40100, Pakistan

⁵Women University Multan, Multan 59300, Pakistan

Corresponding author e-mail: amdadchoudhary33@gmail.com

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ABSTRACT

In the developing world, Zinc deficiency is a bigger most socio-economic concern for the human and constraints for crop production. The deficiency of micronutrients in humans is generally overlapped with the deficiency in the soil. Mostly zinc deficiency is prevalent due to the consumption of cereals because their grains are genetically low in zinc concentration. The zinc deficiency is mostly occurred in soil due to cropping intensification. Intensification of zinc deficiency in humans mainly occurred due to the regular consumption of cereals as a staple food. Zn plays a key role to regulate the different functions in plants, animals, and humans. In humans, especially zinc is required for the immunity boost up. Therefore, a rapid and complementary approach is required for the biofortification of cereal crops to control zinc deficiency in the developing world. In this scenario, the breeding strategy is cost-effective, competent, and requires a lot of time to biofortified the cereals; but, on the other hand, agronomic biofortification is one of the most competitive approaches and rapid strategies. In the agronomic strategies of biofortification, (I) the priming of cereal seeds increases the germination ability and their strength/resistance against biotic & abiotic stressors (II) Soil and foliar application of zinc fertilizer ($ZnSO_4$) greatly increase the zinc concentration in grain cereals. Generally, soil with foliar application increases the zinc concentration in grains many folds. Chelation-based strategies play a key role in zinc uptake and remobilization. So, agronomic strategies with chelation help to increase zinc content many folds in cereal grains.

Keywords: Zinc deficiency, Socio-economic, Intensification, Agronomic strategies, Breeding Approach, Chelation.

INTRODUCTION

Zinc is one of the most important bio-element for the growth and reproduction of plants, animals, and humans. In the development of human life, cycle zinc plays a critical role, especially in pregnant women and children. Zn is a necessary element for human growth and development (Zinc 2005). Reports indicate that annually about 500,000 children are died due to zinc and iron deficiency and they are under the age of 5 years (Black et al. 2008). Zinc performs different functions in the plant body like enzymes activation, free radical detoxification, helps in the retaining & tolerance to plant stressors, and is especially needed for healthy root structure (Cakmak 2008; Peck & McDonald 2010). In developing countries, low dietary intake of zinc can cause impaired growth of offspring. Zinc and iron deficiency is most prevalent due to cereal consumption because their grains are genetically low in concentration

of zinc. Most cereal-based foods are regularly consumed in these countries that are why they have more incidence of zinc deficiency (Cakmak 2008).

In developing countries, about 80% of the population are living at the risk of zinc deficiency, and also estimated that more than 20% of the populations are facing zinc deficiency globally (WHO, 2002; Bouis et al., 2012). Human dependency on zinc-deficient cereals, especially in developing counties may be serious for those how to deal with this condition. The availability and required amount gap for the good status of health is 40-50ppm (Cakmak 2010). Cereals are grown on micronutrient deficient soils, then results in a many times reduction in cereal grains. These micronutrient deficiencies particularly zinc occur due to the cropping intensification that causes the depletion of soil micronutrients. Process of fast degradation results in the

deficiency of micronutrients in soil and plants (Manojlovic et al., 2019).

Zinc deficiency is widespread across developing countries and threatens the population with different problems and diseases. Among the developing nations, Zinc deficiency is at the 5th number, and leading risk for causing diseases (Maret & Sanstead, 2006). Globally, one-third of the population is threatened by zinc deficiency problems (Hotz & Brown, 2004), in term developing countries 50% of pregnant females and children are facing zinc deficiency. Particularly, micronutrients deficiencies occur where they are relying only on a cereals-based diet as a staple food. Micronutrient deficiency is more adverse due to cereals-based diets like wheat, maize, rice (Pfeiffer & McClafferty, 2007 a, b).

In the human body, Zn performs a large number of important functions which are very necessary for human health, and are also known to be the “metal of life” (Kaur, Gupta, Saraf, & Saraf, 2014). Zn is also behaving as an anti-depressant in human life (Amani, Saeidi, Nazari, & Nematpour, 2010). Some diseases like Alzheimer’s and Parkinson’s occurs in humans having lower Zn levels in the blood (brewer et al., 2010). The inhibition of diseases such as hepatitis and liver cirrhosis can be done with the use of Zn (Nakayama et al., 2002). The functioning of the endocrine system via modulation of thymus glands activity is regulated by the Zn micronutrient (Hasse & Rink, 2009).

The deficiencies of the micronutrients like Zn, Mg, and Cu especially during pregnancy may cause infertility, pregnancy wastage, pregnancy-induced hypertension, placental abruption; stillbirth, and low birth weight (Pathak & Kapil, 2004). An important function of the Zn transporter was detected in the cultured breast cells and mammary glands (Kelleher, Seo, & Lopaz, 2009), which realize the importance of Zn in lactation. Especially in children the Zn deficiency cause many health problems, it may also become chronic in case of serve deficiency of Zn like stunted growth, weight loss, increased susceptibility to infection, and mortality (Salgueiro et al., 2002).

Syndrome of Zn deficiency syndrome may result in erosive skin changes especially alopecia if occurs in breastfed infants (Heinen, Matern, Pringsheim, Leititis, & Brandis, 1995).

The normal concentration of Zn also regulates the immune functions in the human body (Rink & Gabriel, 2001). The deficiency of Zn in the human body also provides the susceptibility to a wide variety of pathogens to cause infections and diseases (Shankar & Prasad, 1998). Oxidative stress may cause a variety of neurological diseases in the rapid change of Zn metabolism (Cuajungco & Lees, 1997). A suitable concentration of Zn plays a very crucial role in the division of cells, growth of cells, differentiation, and death of cells (apoptosis; Maclean, 2005).

Individuals with Zn deficiency are predisposed genetically predisposed to deregulation from

inflammatory responses, this deficiency may cause the reduction of the probability of successful aging process in human beings (Vasto et al., 2006). Delayed injury or wound healing is also associated with Zn deficiency. Pneumonia recovery is also associated with micronutrients, it’s also needed for the regulation of chronic hemodialysis, homeostasis, diarrhea recovery, antimicrobial activity, eyesight, and genome (Reviewed in Kaur et al., 2014).

Focus on the importance of Zn and Fe in human life and child health, currently focused on the production of biofortified cereals to overcome malnourishment and also to focus on the associated hardships with the production of Zn-enriched cereal crop. Cereals are selected as a targeted crop because of their edibility and leading to malnutrition of Zn in maize crop, almost 100% of the Pakistani soils are Zn deficient.

Zinc Bioavailability

The development of Zn enrichment diets is also important in bioavailability in dietary foods. Zn and Fe bioavailability are useful in the case of fraction intake of maize absorbed into the circulated blood system and this micronutrient is used by the body for different physiological functions. By assessing the Zn bioavailability, the three main factors in healthy individuals, which are the status of individuals, total Zn concentration in diet, and availability of soluble Zn in food (Lonnerdal, 2000). Dependency of Zn absorption is in form of available Zn in food and also in the presence of the inhibitors and enhancers of Zn absorption available in food.

Several factors are known to be the influence of Zn absorption in the human body. The well-known Zn inhibitor Phytic acid (PA) (Hexa and Penta-Phosphate) binds Zn in the gastrointestinal tract (Hambidge, Miller, & Krebs, 2011). In maize or other cereals, phytate is a major form of storing phosphorus compound which may up to 80% of the almost total seed phosphorus (King & Cousins, 2006).

Phytic acid contents are ranging from 0.62-1.17 g/100 g detected in the different samples based on the molecules having 28.20% phosphorus (Hidvegi & Lasztity, 2002). However, the phytate content in seeds varies across the different regions of the world depending on the heredity of germplasms and the application of phosphatic fertilization. The ratio of phytate to Zn (Phytate: Zinc) in foods is used in the prediction of the phytate inhibitory effects. According to World Health Organization (WHO), molar ratio up to 15:1 and according to the International Zinc Nutrition Consultant Group (IZiNCG), the molar ratio of 18:1 is associated with inhibition of zinc absorption and indeterminate the concentration of Zn in the human body (Allen, Benoist, Dary, & Hurrell, 2006; Brown et al., 2006).

Enhancement of Zn-Bioavailability

Various factors like food type, physiological status of the body, level of anti-nutritional and promoting factors play a critical role in making Zn bioavailability to humans (Gupta et al., 2015).

Nearly 80% of phosphorus in maize grain is present as phytic acid and the primary function of phytate in the seed is to store phosphorus as an energy source and antioxidants essentially required for seed germination. But the negative charge of the phytic acids chelates the positively charged minerals like Fe and Zn and makes them unavailable in the animal gut.

The bioavailability of Zn can be increased in two ways the (1) by hydrolysis of the Phytic Acid (PA) and (2) by enhancing the activation of the phytase enzyme. The activity of endo and exogenous phytase enzymes results in the hydrolysis of Phytic Acid (Hurrell, 2004). Zn bioavailability is also affected by the interaction between iron and zinc. It is reported that Zn uptake affects badly after application high dose of inorganic iron (Solomons & Jacob, 1981). The protein uptakes have a positive impact on the Zn uptake because protein uptake increases the uptake of Zn. Animal protein sources like meat; eggs, cheese, and chicken improve zinc bioavailability. Zn absorption and solubility also increase with the binding of soluble chelators (Lonnerdal, 2000).

Variable Age	WHO Weight (Kg)	Requirement (mg/Kg)	Variable Age	FNB Weight (Kg)	Requirement (mg/Kg)
6-12 months	9	0.84	6-12 months	9	0.84
1-3 Years	12	0.83	1-3 Years	13	0.53
3-6 Years	17	0.97	4-8 Years	22	0.83
6-10 Years	25	1.12	8-13 Years	40	1.53
10-12 Years	35	1.40	14-18 Years (Male)	64	2.52
12-15 Years	48	1.82	14-18 Years (Female)	57	1.98
15-18 Years (Male)	64	1.97	Pregnancy	-	2.68
15-18 Years (Female)	55	1.54	Lactation	-	2.98
Pregnancy	-	2.27			
Lactation	-	2.89			

In adults, the Zn absorption requirement is 3 milligram/day (institute of medicine, 2001). Miller's model shows that 300g maize kernels can provide 55-59% of daily human requirements. A study reported that biofortified maize significantly lower molar ratio which is 34:1 due to higher Zn contents than normal maize having a 38:1 molar ratio (Chomba et al., 2015).

There are also different strategies to improve the dietary Zn bioavailability which will be discussed in the subsequent sections. The improvement in the Zn bioavailability or absorption first needed to know recommended dietary intake dose of Zn.

Recommended dietary intake of Zinc

Daily dietary intake of humans for the different age groups is recommended by the World Health

Organization (WHO) and Food and Nutrition Board (FNB) of Institute of Medicine (Food and Nutrition Board/ Institute of Medicine, 2002; WHO, 2002). Different parameters or different statistical methods are used for the estimation of the daily dietary intake of humans for the different ages of humans. Recommended dietary intake of Zn could be improved by applying the different strategies or methods which are discussed in the following sections.

Methods of Zinc Biofortification

Grains like maize are hereditarily not capable to aggregate the broadly huge measure of Zn substance. Numerous sorts of grain like maize and wheat generally become on the Zn insufficient soils in undeveloped nations like Pakistan, India, and Iran. The significant reasons for micronutrient inadequacies in soils are the green revolution advanced the high return assortments varieties, enormous water irrigation systems rehearses, and macronutrient (N, K, P) fertilizer use. There is a negative relationship among's irrigations and phosphorus was seen with Zn take-up which prompts a decrease of Zn content in Cereals. Zn insufficiency is the significant reason for Zn malnutrition for Cereal consumers. Zn biofortification focus of Cereals can be accomplished through agronomic techniques. These strategies are discussed in subsequent sections to overcome the hidden hunger for Zn.

Agronomic Practices

These practices include the treatment before sowing like seed priming, soil application, and foliar application Zn. Zn fertilization practices have various outcomes for the different modes of application and cereal crops. The Zinc contents are variables in different agricultural soils and their range from 10-300 mg per Kg. Overall mean zinc content in soils is around 5-55mg per Kg. Different assays are used to determine the threshold level of Soil Zn deficiency.

Zn concentration in soils' lower critical limits for most of the crops ranges between 0.5 to 2.0 mg Zn/Kg for DTPA and 0.5 to 3.0 mg Zn/Kg for Mehlich-1. Zinc deficient soils are also correlated with these conditions: neutral to alkaline soils, low organic matters soil of sandy texture, high level of phosphate, highly weathered parent material produce the acidic soil with inheritably low Zn status, calcareous soils, mucks, and peat soils with inheritably low concentration of Zinc, permanently wet or waterlogged fields; irrigation water, high carbonate and magnesium in the Soil. Across the world, Zn deficiency in soil is also linked with human Zn deficiency.

Priming of Cereal seeds

Seed priming is a technique in which different cereals seeds are treated with different solutions or for attempting to gain a high level of biofortification. Commonly two types of practices are used which are hydro-priming and osmo-priming. Mainly seed priming performs to improve seed germination and resistance against abiotic stress at early stages but some are performed for the maturity stages of cereals especially

maize. Seed priming which had done with ZnSO₄, and increased the yield up to 27%. Different reports show that the increase in biological yield, grain yield, and Stover can be done by using ZnSO₄ as a priming agent in different maize varieties.

Seed priming which had done with Zn solutions can improve the Zn contents of kernels maize at the stages of maturity along with other traits of maize. The combined application of seed priming and foliar application of Zn can cause an improvement in kernels' Zn contents by 43.61% in hybrid maize varieties. It was also found the Zn application of seed priming and foliar application was greatly improved the kernel's Zn contents. Therefore, many studies show that seed priming combined with different methods can be useful rather than allowing only seed priming.

Soil and Foliar application of Zn-Fertilizer

Soil and foliar applications were reported to have differential effects on the growth of cereals. But on the other hand, the timing of application of Zn fertilizer has significant effects on the concentration of maize. Soil application in Zn fertilizers showed up to a 40% increase of Zn concentration in (maize) cereals. The foliar application also increases Iron concentration and reduces cadmium toxicity in cereals. Zn concentration in maize cereals could be improved through soil application of Zn fertilizer or in combination with seed priming.

Crop plants uptake a small portion (1-5%) of the soil-applied Zn fertilizer. The small portion of the uptake depends on the proportion and types of fertilizers applied. The inefficiency in uptake is attributed to the quick fixation of Zn into unavailable forms and ZnSO₄ with limited mobility of Zn. However, an increase in the rate of Zn fertilizer application may increase the Zn uptake by the provision of more available Zn, but this is not economical to approach the fertilizer rate.

Zn applied in the form of Fertilizer (ZnSO₄) may get fixation about 75% and rendered unavailable form for plant uptake. More than pH 7 may enhance the fixation of Zn due high availability of carbonates. However, the availability of Zn in plant-available form can be increased due to the production of gluconic acid.

Gluconic acids are readily biodegradable acids and can produce the major anion involved in the solubilization of fixed Zn in soil, making Zn readily available for plant uptake. Arbuscular mycorrhizal fungi (AMF) improve the host plant nutrition by colonizing the roots and enhancing the uptake of immobile metal micronutrients. Mycorrhizal colonies acidify the rhizosphere and urge the plants to uptake the Zn through the solubilization of nutrients.

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the plants to uptake the Zn through the solubilization of nutrients.

Chelation-based strategies and their role in Zinc remobilization and uptake

The uptake and remobilization is a major bottleneck to increasing Zn concentration in cereals. Many strategies are adopted mainly chelation-based strategies are adopted for plant mineral uptake. Chelation-based strategies in cereals are adapted to mobility in soil and increase in Zn & Fe uptake. In reality, chelation-based methodologies are characterized as such procedures in which Phyto-siderophores are delivered in the soil which prepare the Fe and Zn in soils and resultantly increment their take-up. Phytosiderophores are having high infinity to form the complex with Fe (III), and these types of complexes are taken up by plants that have YS1/ YSL (Yellow Stripe Like) are the member of this gene family. PS also can chelate Zn and which are easily taken up by the plants from the soil.

PS (65Zn-label) increases the uptake of the Zn (II) under soil deficiency conditions. Phytosiderophores are phytochelators (PCs) and metallothioneins (MTs). These special types of metal chelators can bind with different heavy metals like Zn and Fe and therefore keep the free metal concentrations at a low level. Special types of metal chelators like phytochelators (PCs) and Metallothioneins (MTs) are having the ability to bond with different metals like Zn and Fe and make it freely unavailable for uptake.

Zn stored in vacuoles is needed under Zinc-deficient conditions to perform different functions in the cytosol. Transporters are involved in the remobilization of Zn vacuoles to cytosol under Zn-deficiency conditions are very little known.

CONCLUSION

Zn deficiency mostly occurs in the rural areas of developing countries and this is very crucial for pregnant women and children's development. Zn deficiency can correct by using supplements or highly nutritional nourishment. The people of rural areas with low-income status are actual victims of Zn malnourishment. Cereals are used as a staple food in Latin America, African countries, and some countries of Asia. Dietary diversification is not reachable for the actual victims. Although, different solutions are available biofortification of cereals is the most appropriate approach to overcome the Zn deficiencies. Mineral contents of the plants depend on the different environmental factors (Soil Profile), so agronomic approaches are useful for the biofortification of the cereals.

Different agronomic approaches are used, the seed priming used at early stages to maximize the seed germination ability and resistance against biotic and abiotic stresses but the soil and foliar application or combined application of different Zn solutions at different stages are more beneficial to improve the Zn concentration in cereal grains. The combined application

of soil and foliar application is more beneficial for agronomic practices.

Foliar application with either band placement or broadcast is used to maximize the phyto available Zn to optimize the cereal yields with increases in biofortification level of Zn in grains. All of these agronomic combinations and approaches are to enhance the biofortification level. Foliar with soil application of fertilizer at plow depth are very helpful for Zn distribution in gains and increases the Zn bioavailability and protein concentration in cereal grains. These agronomic practices will be helpful for dietary diversification as well as improve the nutritional quality and yield of cereals. At that present scenario of increasing population, there is a bigger challenge to maintain the nutritional quality of dietary diversification along with an increase in production.

However, an increase in desired Zn concentration in cereals by agronomic practices recommended the breeding and molecular approaches must be practiced in the field and used for future research in biofortification programs.

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