**CHARACTERIZATION OF LENTIL GENOTYPES FOR ZINC BIOFORTIFICATION AND SOARING GRAIN OUTPUT**

**ABSTRACT**

Along with food security, micronutrient malnutrition, especially zinc (Zn) deficiency, is primarily a problem in South Asia. Pulses, being an inexpensive source of protein, play a major role in giving calories to the people in poor nations. In addition to increasing profits, zinc biofortification of pulses, particularly lentils can help alleviate human zinc deficiency. The genotypes of lentils were categorized in a field experiment according to their greater grain production and Zn content. A two-factor randomized design was used to cultivate the 12 lentil genotypes at Zn concentrations of 0, 12.5, and 25 ppm with three replicates.The results showed that compared to a lower application rate and control, a 20 ppm Zn treatment greatly boosted plant growth, grain output, and grain Zn content. The large grain production of the BARI Masoor-8 genotype was exceptional. The genotypes of lentils DPL-62, BARI Masoor-3, BARI Masoor-8, and BARI Masoor-5 were identified as high grain yield cultivars and ranked as Zn receptive genotypes and was determined, these genotypes improved crop yield and grain Zn content by responding differently to Zn fertilization. However, further exploration is needed to furnish broad recommendations for the finestpedigree and application know-how while maintaining the necessary quantity of bioavailable zinc.

**Keywords:** Lentil genotypes, Zinc Concentration, Biofortification, Categorization

**INTRODUCTION**

Considering the present rate of increase, especially in South Asia(USDA 2015), it is anticipated that the earth populacemay approach 10 zillion people by 2050(FAO 2017). An estimated 60-80% increase in present food output is required to feed the population in 2050 (FAO 2009). Furthermore, the region faces a significant risk of malnutrition due to a large reliance on certain grains, such as wheatand rice,32which are squat in micronutrients, especially zinc. One of the most common micronutrient deficiencies in developing nations, particularly in South Asia, is zinc deficiency (Seth et al. 2018;Haideretal.2018; Ullah et al. 2019). However, pursuant to approximates from World Health Organization (WHO), a third of the earth populace lacks enough zinc (Swain et al. 2016).Therefore, there is a clear necessity to boost food output and choose alternatives to cereals as a main diet. Because they are an inexpensive source of protein and calories, pulses can be an excellent choice for feeding the populace as well as addressing the problem of malnutrition. The consumption of pulses in India has decreased from 0.96 kg to 0.86 kg, as populations are moving from vegetable sources of protein (such as pulsesandbeans) to more costly fount like animal proteins as developing nations become sufficiently wealthy (National Sample Survey Office, 2001, 2014). Additionally, lentils are a great source of micronutrients, particularly iron (Fe) and zinc (Ganesan and Xu 2017, Rahman et al. 2013, Rochfort and Panozzo 2007, Dueñas et al. 2002).

The greatest strategy to avoid micronutrient deficiency is to eat foods that are richer in absorbable micronutrients (Bouis and Saltzman 2017, Miller and Welch 2013, Hotz and Brown 2004). Previously, it was suggested that the best ways to address human zinc deficiency were dietary fortification, food diversification, and supplementation. However, these approaches are seldom viable, therefore they are sidelined to be a better choice in impoverished nations. Because there isn’t enough domestic research, biofortification-the process of adding minerals to grainsisn't yet commonly used in developing nations, despite being universally acknowledged as a cost-effective strategy. The current Zn shortage issue is addressed temporarily by using Zn fertilizers (such as agronomic biofortification) as opposed to breeding programs (Hussain et al. 2011 and 2013, Cakmak 2008a and b).

A number of dietary parameters, primarily the amount of zinc in the human diet, have a revelatory influence on bioavailability of zinc(White and Broadley 2005, Lönnerdal 2000). Moreover, nutritional promoters and anti-nutrients have an impact on the bioavailability of zinc in the human gut(Cakmak and Kutman 2018; Hussain et al. 2018, FAO/WHO2004, Brown et al. 2001). A significant amount of phytate, an anti-nutrient and the principal embodiment of P storage in grains, is rested in cereals and pulses. Phytate can bind zinc by creating insoluble complexes. Consequently, the bioavailability of zinc in dietary products has often been cataloged using the [Phytate]:[Zn] ratio(Brown et al. 2001). Grain Zn and phytate levels vary significantly among crop genotypes(Hussain et al. 2011, 2013). Cultivars and zinc treatment have an impact on crop production and nutritional position(Manaf et al. 2019, Hidoto et al. 2017). Nonetheless, it is preferable to screen the current lentil germplasm for low phytate concentrations and increased grain zinc values. In order to produce the highest grain yield on Zn-deficient soil, several lentil cropgenotypes of with stubby phytate-to-Zn molar ratios and greater bioaccessible Zn must be selected out. However, competency of zinc does not guarantee a soaring concentration of zinc in grain (Torunetal, 2000). Therefore, genotypes of lentil produced on soils which are Zn-deficient, it is necessary to assess Zn treatment in order to optimize grain production and enhance grain Zn contents. The current study concentrated on classifying germplasm of lentil for optimal yield and boosting zinc bioavailability in grain by foliar spray application. In addition to aiding in crop promotion, this study might assist researchers and policymakers in increasing the intake of lentils in order to promote overall health.

**MATERIALS AND METHODS**

**Seed collection**

For the purpose of determining the Zn biofortification, seed samples of twelve genotypes of grown lentils were gathered. All the genotypes are collected from two sourcesPulse and Oilseed research station (PORS) Berhampore and All India Coordinated Research Project (AICRP) on MULLaRP (Mung bean, Urd bean, Lentil, Lathyrus, Rajmash and Pea).

**Field Experiment**

The study was carried out at the Bidhan Chandra Krishi Viswavidyalaya research area in West Bengal, India (22°93' N and 88°53' E). To create a composite sample from the field, several soil samples were chosen at random. Three composite samples with a surface layer of up to 30 cm were gathered, sieved using 2.0 mm sieve, and their physico-chemical characteristics were examined. The availability of zinc is most frequently linked to soil conditions. The alkaline nature and pH of the soil have a significant impact on the obtainability of soil zinc and its captivation by plants (Alloway 2009).

Furthermore, the solubility of zinc and, eventually, the zinc levels in lentil genotypes are influenced by the soil habitat, moisture, and precipitation. Atomic absorption spectrophotometer (AAS) was utilized to quantify the zinc that plants could access in soil extract (Lindsay and Norvell 1978). The texture of the soil samples is composed of 14% sand, 65% silt, and 21% clay. The pH and EC of soil extract was 6.64 and 1.58 dS m-1 correspondingly. While the concentrations of accessible phosphorus and potassium were 27.38 and 168.46 kg ha-1, respectively, the organic matter and nitrogen in total of soil were found to be 0.49% and 291.15 kg ha-1.

Twelve different genotypes of lentil seeds were cultivated in separate 15 × 30 cm plots. Each genotype was cultivated in a two-factorial randomized complete block design with three replicates at three different Zn doses: 0, 12.5, and 25 ppm. At 30- and 60-days following seeding, the zinc treatments were administered twice. Zinc was showered on the leaves as ZnSO4. Recommended amounts of N and P (30:57 kg/ha) were met by using urea and DAP (di-ammonium phosphate) fertilizers, respectively. The crop was grown with residual soil moisture, and for improved growth, a single, life-saving irrigation was given during the flowering stage. After thirty and sixty days of seeding, weeds were manually removed. When the plants reached maturity, they were picked, and the lentil seeds were manually separated. After the grains were separated, straw samples were gathered, and metrics related to yieldwere computed.

**Zinc and Phytate Analysis**

Lentil genotype plant and seed samples were pulverized in a grinding mill after being desiccated in ovenat 65 °C for 48 hours (Liu et al. 2006). With minor adjustments recommended by Singh et al. (2005), these samples (grainand straw) then assimilated (each of 0.2 g sample) using HNO3 and HClO4 created in 9:4 ratio in accordance with the procedure outlined by Zarcinasetal in 1987. AAS was used to measure the digestate's zinc content. Samples (50 mg) were extracted using 50 mL of a 3% TCA solution and centrifuged at 5000 rpm for 30 minutes at room temperature in order to determine the amount of phytate.Phytate was determined by the indirect approach, which involved developing a pink color using 2,2′-bi-pyridine and un-reacted Fe (III) (Haug and Lantzsch 1983) and measuring their absorption at 519 nm using a spectrophotometer.

**Categorization of Lentil Genotypes**

Exercising indicator scores of 3, 2 and 1 for high, medium and lowscoring genotypes, respectively, the lentil genotypes were categorized. By measuring the mean of population (μ) and standard deviationof intended parameters, genotypic categorization is created depending on how well applied zinc performs in comparison to control (Bilal et al. 2018, Aziz et al. 2011). Genotypes of lentils were classified as low if the genotype mean is less than μ-SD, medium if the mean is betwixt μ + SD and μ-SD, and high if mean is larger to μ + SD.

**Statistical Analysis**

Analysis of variance was executed onyield and associated parameters, phytate, and Zn concentration data using the Windows-based SPSS 12.0 application. At the 5% level of significance, treatment means with significant differences were separated using the Standard Error of Mean (SEm±).

**RESULTS AND DISCUSSION**

**Yield and Related Parameters**

Table 1 provides information on the yield and associated characteristics of lentil genotypes at various zinc treatment rates. When matched to control, the application of zinc greatly escalated grain size, which improved the test weight. For test weight, genotypes varied considerably across all treatments. When compared to other lentil genotypes, the genotype DPL-62 with 12.5 and 25 ppm Zn administration showed the highest test weight (27.7 g). Similarly, when grown without Zn treatment, BARI Masoor-3 had the lowest test weight (19.86 g) (Table 2). The figure of pods per plant upsurged with applying zinc. Comparing Zn application at 25 ppm to other treatments and control, pod numbers increased.Similarly, it was discovered that when Zn was applied at 25 ppm, the lentil variety BARI Masoor-8 produced more pods in a single plant (118), whereas the control treatment's Precoz produced the fewest pods in a single plant (50). Zinc treatment has significantly increased plant output by increasing the number of grains in a single pod, regardless of the genotype of lentils. In comparison to other genotypes, the lentil variety Subrata had the highest mean number of grains in a single pod (1.72), while Ranjan, DPL-62, and Precoz had lower grain counts (Table 2). Compared to treatment which did not receive any zinc application, a significant impact on grain yield was observed in the applied treatments (Table 2).When 25 ppm Zn was administered, the highest grain production compared to the control was noted. When zinc was applied, the lentil variety Precoz produced less grain (1.59 g plant-1) than the lentil variety BARI Masoor-8, which produced a higher grain yield (4.53 g per plant). Table 2 shows information on the genotypes of lentils' straw yield at various Zn applications. It shown that, as connected to the control, a significant rise in straw output was seen with the subsequent Zn treatment. With Zn applied at 25 ppm, the lentil genotype Ranjan produced the least amount of straw (5.23 g plant-1), whereas the lentil variety BARI Masoor-8 produced the most (6.50 g plant-1).

When zinc is applied to calcareous and alkaline soil that has a low plant-available zinc content, genotypes react differentially (Maqsood *et al.* 2015, Abid *et al*. 2013, Alloway 2008). Plant development and production characteristics were enhanced by zinc treatment (Manaf *et al.* 2019, Farouk and Al-Amri 2019, Bala *et al*. 2019 and Alloway 2009). Zn significantly impacted the development of lentils,output of grain, and Zn content of grain in current study. As anticipated, Zn application enhances setting of seed (Hussain *et al*. 2013, Marschner and Rengel 2012), which leads to inflated grain production (Pandey and Gautam 2009, Pandey *et al.* 2006). This is why both zinc treatments unveiled a noteworthy improvement in grain production when connected to the control(Table 2). Additionally, Zn treatment was shown to increase the pods numberper plant and grains per pod (Table 2). Nonetheless, notable genetic variations were seen between genotypes of lentils. When 25 ppm of zinc was applied to the lentil variety BARI Masoor-8, matched to the control, a roughly 29% escalation in grain production was seen (Table 2). Since pollen grains contain a large quantity of zinc, this increase in grain output may be the

**Table 1:** Average yield values (mean±SD) and associated traits of genotypes of lentils as affected by zinc application

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Parameters | Test weight (g plant-1)  | Number of pods per plant | Number of grains pod-1 | Grain yield (g plant-1) | Straw yield (g plant-1) |
| Genotypes | Zn application (ppm) | Zn application (ppm) | Zn application (ppm) | Zn application (ppm) | Zn application (ppm) |
|   | 0 | 12.5 | 25 | 0 | 12.5 | 25 | 0 | 12.5 | 25 | 0 | 12.5 | 25 | 0 | 12.5 | 25 |
| Ranjan | 21.14 | 21.14 | 21.145 | 54 | 56 | 63 | 1.5 | 1.5 | 1.51 | 1.44 | 1.57 | 1.86 | 4.7 | 4.92 | 5.23 |
| Subhendu | 21.1 | 21.11 | 21.13 | 61 | 63 | 70 | 1.58 | 1.6 | 1.63 | 1.62 | 1.79 | 1.99 | 4.95 | 5.1 | 5.3 |
| DPL-62 | 26.7 | 27.7 | 27.7 | 78 | 81 | 88 | 1.5 | 1.51 | 1.53 | 2.29 | 2.76 | 3.05 | 5.24 | 5.44 | 5.82 |
| Maitree | 23.98 | 23.99 | 24 | 103 | 106 | 112 | 1.66 | 1.66 | 1.68 | 3.49 | 3.62 | 3.96 | 5.07 | 5.27 | 5.67 |
| Subrata  | 22.72 | 22.72 | 22.72 | 85 | 88 | 93 | 1.72 | 1.72 | 1.72 | 2.89 | 2.98 | 3.16 | 4.89 | 5.05 | 5.27 |
| PusaVaibhab | 22.14 | 22.145 | 22.15 | 62 | 65 | 71 | 1.6 | 1.6 | 1.61 | 1.65 | 1.83 | 2.01 | 5.45 | 5.66 | 5.94 |
| Precoz | 21.07 | 21.09 | 21.1 | 50 | 53 | 58 | 1.5 | 1.5 | 1.51 | 1.38 | 1.42 | 1.59 | 5.22 | 5.43 | 5.81 |
| BARI Masoor 3 | 19.86 | 19.86 | 19.89 | 60 | 62 | 68 | 1.53 | 1.53 | 1.54 | 1.62 | 1.78 | 1.97 | 4.86 | 4.99 | 5.39 |
| BARI Masoor 4 | 21.08 | 21.1 | 21.11 | 68 | 71 | 77 | 1.53 | 1.53 | 1.53 | 1.81 | 1.99 | 2.21 | 4.92 | 5.03 | 5.4 |
| BARI Masoor 5 | 20.84 | 20.85 | 20.87 | 86 | 88 | 95 | 1.61 | 1.61 | 1.63 | 2.9 | 3.01 | 3.2 | 5 | 5.26 | 5.63 |
| BARI Masoor 8 | 23.76 | 23.76 | 23.76 | 108 | 111 | 118 | 1.66 | 1.67 | 1.69 | 3.74 | 3.91 | 4.53 | 5.72 | 6.01 | 6.5 |
| L-4076 | 27.54 | 27.55 | 27.55 | 59 | 62 | 68 | 1.43 | 1.44 | 1.44 | 1.57 | 1.69 | 1.86 | 5.61 | 5.8 | 6.26 |
| SEm(±) | 0.01 | 0.28 | 0.01 | 0.02 | 0.01 |
| CD (P=0.05) | NS | NS | NS | 0.05 | 0.04 |

**Table 2:** Effects of zinc treatment on zinc absorption, phytate content, grain zinc, and straw zinc in 12 genotypes of lentils

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Parameters | Straw Zinc (mg kg-1) | Grain Zinc (mg kg-1) | Grain Phytate (μg g-1) | Zinc Uptake (g ha-1) |
| Genotypes | Zn application (ppm) | Zn application (ppm) | Zn application (ppm) | Zn application (ppm) |
|   | 0 | 12.5 | 25 | 0 | 12.5 | 25 | 0 | 12.5 | 25 | 0 | 12.5 | 25 |
| Ranjan | 28.67 | 38.5 | 46.17 | 44.5 | 52.67 | 58 | 9.08 | 7.87 | 6.7 | 31.33 | 40.5 | 49 |
| Subhendu | 28.67 | 37.67 | 44.83 | 42.83 | 51 | 56.33 | 9.03 | 7.87 | 6.73 | 32.67 | 41.67 | 50.83 |
| DPL-62 | 29.17 | 38.83 | 46.67 | 45.42 | 53.5 | 60.5 | 8.28 | 7.1 | 5.83 | 34.67 | 44 | 53.5 |
| Maitree | 29.5 | 40.17 | 47.5 | 45.17 | 54.33 | 60 | 8.18 | 7 | 5.87 | 32 | 41.17 | 50.5 |
| Subrata  | 30 | 39.67 | 46.83 | 43.83 | 51.83 | 57.5 | 7.62 | 6.48 | 5.35 | 29 | 37.17 | 43.83 |
| PusaVaibhab | 29.67 | 39 | 46.5 | 44.83 | 53 | 58.83 | 6.95 | 5.9 | 4.77 | 29.17 | 38 | 46.67 |
| Precoz | 28.17 | 38.33 | 45.83 | 45.67 | 54.17 | 59.83 | 7.68 | 6.65 | 5.48 | 30.83 | 40 | 48.67 |
| BARI Masoor 3 | 29.17 | 38.33 | 45.67 | 47 | 55.33 | 60.67 | 7.95 | 6.82 | 5.67 | 30 | 39.67 | 48.83 |
| BARI Masoor 4 | 28.83 | 38.83 | 46.83 | 44 | 52.67 | 59 | 7.68 | 6.62 | 5.4 | 32.17 | 42.33 | 53.5 |
| BARI Masoor 5 | 30.17 | 40.17 | 48.17 | 48.5 | 57.33 | 63.67 | 7.45 | 6.33 | 5.13 | 33.17 | 43.5 | 53.33 |
| BARI Masoor 8 | 30.33 | 41 | 49.33 | 47 | 56 | 62.17 | 7.97 | 6.83 | 5.68 | 32.83 | 42.5 | 52.17 |
| L-4076 | 28.49 | 38.42 | 45.96 | 44.49 | 52.5 | 58.03 | 8.4 | 7.24 | 6.06 | 31.64 | 42.06 | 50.45 |
| SEm(±) | 0.11 | 0.14 | 0.02 | 0.13 |
| CD (P=0.05) | NS | NS | NS | 0.37 |

consequence of zinc's participation in the reproductive stage (such as pollen grain formation and fertilization) (Ali *et al*.2017, Reid *et al*.2011 and Jenik and Kathryn 2005). Several researchers have previously shown similar increases in lentil grain production with Zn treatment (Ali *et al.* 2017, Singh and Singh 2012 and Singh *et al*. 1995).

**Zinc, Phytate Concentration and Zinc Uptake**

While certain genotypes displayed statistically insignificant changes with identical letters, Zn concentration [Zn] of straw indicates dissimilarities based on Zn application, genotypes, and their corresponding interactions (Table 3). With a 25 ppm Zn treatment, straw [Zn] was greater. BARI Masoor-8 had the highest concentration of zinc in straw (49.33 mg kg-1), but the lentil variety Subhendu had the lowest response. Grains[Zn] and plant components of genotypes of lentil is significantly impacted by zinc treatment. While certain genotypes were comparable to others, statistical data for grain [Zn] revealed variances based on treatments (Table 3). The range of grain [Zn] is 42.83-63.67 mg kg-1. The lentil genotype BARI Masoor-5 showed a higher value of grain [Zn] (63.67 mg kg-1) when 25 ppm of Zn was administered, while Subhendu showed the lowest value of grain [Zn] (42.83 mg kg-1) when control treatment was used. In lentil genotypes, it was shown that the content of zinc in straw and grain had a modest but favorable association.

Auxin and other hormones that are requisite for healthy plant improvement are activated by zinc (Begum *et al.* 2016). Prior research also showed that zinc treatment improved the content of zinc in grains (Bala *et al*. 2019, Hidoto *et al*. 2017, Kaya *et al*. 2009). Zinc application considerably raised zinc content of grain in genotypes of lentil in the current investigation. When zinc was applied at 25, the lentil variety BARI Masoor-5 showed aelevatedlevel of zinc in seed. According to Cakmak (2008a), increasing zinc levels might lead to improved grain biofortification performance (Cakmak and Kutman 2018). Since zinc is necessary for pollen grains and fertilization, the majority of zinc moved to grains during fertilization; hence, a reduced zinc supply led to grain zinc insufficiency (Ali *et al*. 2017, Reid et al.2011, Pandey and Gautam 2009). According to handful of studies, zinc deficit is corrected by the zinc application rates (Cakmak and Kutman 2018, Zhao *et al*.2014, Abid *et al.* 2013, Cakmak 2008b). According to several studies (Farooq *et al*. 2018, Cakmak and Kutman 2018, Gupta *et al.* 2016, Ram *et al.* 2015, Zou *et al.* 2012, Cakmak *et al.* 2010, White and Broadley 2009), the Zn fertilization approach is a successful method for biofortifying food crops with zinc. In order to biofortify the lentil crop in future breeding programs, a greater quantity of grain zinc is therefore required (Ali *et al*. 2017, Reid *et al.* 2011, Thavarajah *et al*. 2009).

Zn treatment to all lentil genotypes also had a substantial effect on content of grain phytate (Table 2). The concentration of phytate is considerably decreased by using zinc. With a Zn treatment of 25 ppm, the lowest grain phytate content (4.77 μg g-1) was determined in Pusa Vaibhab, whereas the highest phytate concentration (9.08 μg g−1) was recorded in Ranjan with control.

The availability of zinc is negatively impacted by high amounts of inorganic phosphorus (Perez-Novo et al.2011). Phytate, which is included in the majority of plant diets, is the most significant inhibitor of zinc absorption. As previously studied by researchers (Thavarajah *et al.* 2009, Erdal *et al.* 2002, Marschner and Cakmak 1986), Zn application in lentil genotypes decreased the content of phytate in grains (Table 3). Highest value of grain phytate level was noted in case when no Zn was administered.With a Zn treatment of 25 ppm, the maximum decrease in grain phytate content was determined in Pusa Vaibhab. This decrease in phytate content may be the result of growth dilution or of modifications brought about by zinc in taking up of P from root zone and their subsequent transport throughout the plant (Johnson and Thavarajah 2013, Thavarajahetal.2009, Huang et al. 2000). Zinc absorption and concentration in different plant sections are improved when inorganic P deficiency causes the overexpression of many genes (Khan *et al.* 2014, Misson *et al.* 2005). Because it forms compounds with minerals like zinc, phytotate, which is found in grains, limits the body's ability to absorb zinc.

According to the findings in Table 2, the solitary application of zinc at various development stages considerably enhanced the grain's absorption of zinc in lentils. DPL-62 and BARI Masoor-4 cultivars had the highest zinc uptake (53.5 gha-1). The Subrata type of lentil grain has the lowest zinc absorption (43.83 gha-1). Therefore, the grain zinc absorption of lentils is significantly impacted by the foliar spray of ZnSO4 (25 ppm) at 30 and 60 days after planting. All things considered, foliar applications of ZnSO4 (12.5 and 25 ppm) at 30 and 60 days after seeding are similarly efficient at increasing the absorption of zinc in lentil grains. The study's findings demonstrated that external supplementation significantly increases micronutrient (Zn) intake.

**Characterization of Lentil Genotypes**

All evaluated lentil genotypes' grain Zn concentration and yield were impacted by zinc treatment. However, as compared to the control treatment, genotypes differed considerably in response to administered zinc (Table 3). With applied zinc treatments, the following lentil genotypes-Precoz, Ranjan, L-4076, BARI Masoor-3, Subhendu, Pusa Vaibhab, and BARI Masoor-4produced yield of grainless than 2.61 g plant-1 and received lowest index score (Table 3). The genotypes Maitree and BARI Masoor-8 produced grain yields more than 3.66 g in a single plant, earning them a better grade (3). For grain yield, the other three genotypes were categorized as middling scorers (2), with results ranging 2.61 to 3.66 g plant-1.25 ppm Zn considerably increased grain Zn content. The grain zinc content of the two lentil genotypes, Subhendu and Subrata, was less than 57.83 mg kg-1, resulting in a minimal index score (1). More than 60.18 mg kg-1 of zinc was generated by the four genotypes DPL-62, BARI Masoor-3, BARI Masoor-8, and BARI Masoor-5. The grain zinc concentrations of the remaining six genotypes-Ranjan, L-4076, Pusa Vaibhab, BARI Masoor-4, Precoz, and Maitree ranged from 57.83 to 60.18 mg kg-1.

**Table 3:** Low, medium, and high scoring genotypes of lentils are categorized based on their index scores of several criteria. The population mean (μ) and standard deviation (SD) for each parameter are used to classify each genotype depending on how well the zinc treatment performed in comparison to its control.

|  |  |  |  |
| --- | --- | --- | --- |
| Parameter | Low (Score 1) | Medium (Score 2) | High (Score 3) |
| Grain yield (g plant-1) | <2.61 Precoz< Ranjan, L-4076 < BARI Masoor-3 <Subhendu<PusaVaibhab< BARI Masoor-4 | 2.61-3.66 DPL-62 < Subrata < BARI Masoor-5 | >3.66 Maitree< BARI Masoor-8 |
| Grain Zinc (mg kg-1) | <57.83Subhendu< Subrata | 57.83-60.18 Ranjan < L-4076 <PusaVaibhab< BARI Masoor-4 <Precoz<Maitree | >60.18 DPL-62 < BARI Masoor-3 < BARI Masoor-8 < BARI Masoor-5 |

Based on Zn content and grain output, current classification divides lentil genotypes into many groups. Different genotypes of lentils exhibit varying performance, achieving high, medium and lowscores (Table 2) (Bilal *et al*. (2018); Aziz *et al.* (2011)). Precoz, Ranjan, L-4076, BARI Masoor-3, Subhendu, PusaVaibhab, and BARI Masoor-4 had poor scores because of their low grain yields, but Maitree and BARI Masoor-8, two genotypes of lentils, generated high producing genotypes and received high scores. BARI Masoor-8 is categorized as a Zn-efficient genotype because to its high grain yield and highest grain Zn content (Table 3). Due to its poor yield and decreased zinc content, the genotype Subhendu is categorized as inefficient (Table 3). Because of its low grain zinc score, the lentil genotype Subhendu is classified as Zn non-responsive. Additionally, greater grain zinc concentrations were noted in DPL-62, BARI Masoor-3, 5 and 8 (Table 3).Due to its lower grain zinc content, the lentil genotype Subhendu has a lower score and is graded non-efficient. Likewise, DPL-62, BARI Masoor-3, BARI Masoor-8, and BARI Masoor-5 were classified as Zn responsive genotypes (Table 3) due to their greater grain Zn content and high score. Although, genetic potential of newly crafted biofortified genotypes to imbibeample zinc or to transfer into individual plant parts, especially grains, at beneficial levels may not be completely expressed under scanty supply of available zinc from soil, the classification of lentil genotypes can be used to successfully breed zinc-fortified crops (Cakmak and Kutman 2018, Gupta *et al*. 2016, Cun *et al.* 2014, Cakmak *et al*. 2010).

**CONCLUSION**

Plant development, yield of grain, phytate content and zinc concentration were all enhanced by a 25-ppm zinc treatment. Regarding how the lentil genotypes responded to additional zinc in soil, a significant genotypic dissimilarity was found. Genotype BARI Masoor-8 was classified as a high-yielding and zinc-efficient genotype due to its greater grain production and zinc concentration, whereas the Subhendu genotype was classified as a low-yielding and zinc-inefficient genotype due to its lower yielding and lower zinc concentration. Future breeding initiatives and the promotion of lentils in traditional agricultural systems may benefit from the available data.

**Disclaimer (Artificial intelligence)**

We hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

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**REFERENCE**

Abid M, Ahmed N, Qayyum MF, Shaaban M and Rashid A (2013): Residual and cumulative effect of fertilizer zinc applied in wheat–cotton production system in an irrigated aridisol. Plant Soil Environ 69:505–510. https://doi.org/10.17221/313/2013-PSE

Ali A, Ahmad B, Hussain I, Ali A and Shah FA (2017): Effect of phosphorus and zinc on yield of lentil. Pure Applied Biology 6(4):1397–1402. https://doi.org/10.19045/bspab.2017.600150

Alloway BJ (2008): Zinc in soils and crop nutrition, 2nd edn. International Fertilizer Industry Association, Paris

Alloway BJ (2009): Soil factors associated with zinc deficiency in crops and humans. Environ Geochem Health 31:537–548. https://doi.org/10.1007/s10653-009-9255-4

Aziz T, Rahmatullah, Maqsood MA, Sabir M and Kanwal S (2011) Categorization of brassica cultivars for phosphorus acquisition from phosphate rock on basis of growth and ionic parameters. J Plant Nutr 34:522–533. https://doi.org/10.1080/01904167.2011.538114

Bala R, Kalia A and Dhaliwal SS (2019): Evaluation of efficacy of ZnO nanoparticles as remedial zinc nanofertilizer for rice. Journal of Soil Science and Plant Nutrition 19:379–389. https://doi.org/10.1007/s42729-01900040-z

Begum MC, Islam M, Sarkar MR, Azad MAS, Huda AN and Kabir AH (2016): Auxin signaling is closely associated with Zn efficiency in rice (Oryza sativa L.). J Plant Interact 11:124–129. https://doi.org/10.1080/17429145.2016.1220026

Bilal HM, Aziz T, Maqsood MA, Farooq M and Yan G (2018) Categorization of wheat genotypes for phosphorus efficiency. PLoS One 13(10):e0205471. https://doi.org/10.1371/journal.pone.0205471

Bouis HE and Saltzman A (2017) Improving nutrition through biofortification: a review of evidence from HarvestPlus, 2003 through 2016. Global Food Security 12:49–58. https://doi.org/10.1016/j.gfs.2017.01.009

Brown KH, Wuehler SE and Peerson JM (2001) The importance of zinc in human nutrition and estimate on of the global prevalence of zinc deficiency. Food Nutr Bull 22:113–125. https://doi.org/10.1177/156482650102200201

Cakmak I (2008a) Enrichment of cereal grains with zinc: agronomic or genetic biofortification? Plant Soil 302:1–17. https://doi.org/10.1007/s11104-007-9466-3

Cakmak I (2008b). Zinc deficiency in wheat in Turkey. In: Micronutrient deficiencies in global crop production. Springer, Dordrecht, pp 181–200

Cakmak Iand Kutman UB (2018) Agronomic biofortification of cereals with zinc: a review. Eur J Soil Sci 69(1):172–180. <https://doi.org/10.1111/ejss.12437>

Cakmak I, Pfeiffer WH and McClafferty B (2010) Biofortification of durum wheat with zinc and iron. Cereal Chem 87:10–20. https://doi.org/10.1094/CCHEM-87-1-0010

Cun P, Sarrobert C, Richaud P, Chevalier A, Soreau P, Auroy P (2014) Modulation of Zn/Cd P(1B2)–ATPase activities in Arabidopsis impacts differently on Zn and Cd contents in shoots and seeds. Metallomics 6:2109–2116. https://doi.org/10.1039/x0xx00000x

Dueñas M, Hernández T, Estrella I (2002) Phenolic composition of the cotyledon and the seed coat of lentils (Lens culinaris L.). Eur Food Res Technol 215:478-483. https://doi.org/10.1007/s00217-0020603-1

Erdal I, Yilmaz A, Taban S, Eker S, Torun B, Cakmak I (2002) Phytic acid and phosphorus concentrations in seeds of wheat cultivars grown with and without zinc fertilization. J Plant Nutr 25:113–127. https://doi.org/10.1081/PLN-100108784

FAO (2009) Global agriculture towards 2050. How to feed the world2050. Rome 12-13 October. http://www.fao.org/fileadmin/templates/wsfs/docs/Issues\_papers/HLEF2050\_Global\_Agriculture.pdf

FAO (2017) The future of food and agriculture - trends and challenges.Food and Agriculture Organization of the United Nations Rome.http://www.fao.org/3/a-i6583e.pdf

FAO/WHO (2004) Zinc. In: Vitamin and mineral requirements in human nutrition, 2nd edn. Food and Agriculture Organization and World Health Organization, Rome, pp 230-245

Farooq M, Ullah A, Rehman A, Nawaz A, Nadeem A, Wakeel A, Nadeem F, Siddique KH (2018) Application of zinc improves the productivity and biofortification of fine grain aromatic rice grown in dry seeded and puddled transplanted production systems. Field Crops Res 216:53–62. https://doi.org/10.1016/j.fcr.2017.11.004

Farouk S, Al-Amri SM (2019) Exogenous zinc forms counteract NaClinduced damage by regulating the antioxidant system, osmotic adjustment substances, and ions in canola (Brassica napus L. cv. Pactol) plants. J Soil Sci Plant Nutr:1–13. https://doi.org/10.1007/s42729-019-00087-y

Ganesan K, Xu B (2017) Polyphenol-rich lentils and their health promoting effects. Int J Molecular Sci 18(11):2390. https://doi.org/10.3390/ijms18112390

Gupta N, Ram H, Kumar B (2016) Mechanism of Zinc absorption in plants: uptake, transport, translocation and accumulation. Rev Environ Sci Bio 15(1):89–109

Haider MU, Farooq M, Nawaz A, Hussain M (2018) Foliage applied zinc ensures better growth, yield and grain biofortification of mungbean. Int J Agri Biol 20:2817-2822. https://doi.org/10.17957/IJAB/15.0840

Haug W, Lantzsch H (1983) Sensitive method for the rapid determination of phytate in cereals and cereal products. J Sci Food Agric 34:1423–1424. https://doi.org/10.1002/jsfa.2740341217

Hidoto L, Worku W, Mohammed H, Bunyamin T (2017) Effects of zinc application strategy on zinc content and productivity of chickpea grown under zinc deficient soils. J Soil Sci Plant Nutr 17:112–126. https://doi.org/10.4067/S0718-95162017005000009

Hotz C, Brown KH (2004) Assessment of the risk of zinc deficiency in populations and options for its control. Food Nutr Bull 25:S91-S204

Huang C, Barker SJ, Langridge P, Smith FW, Graham RD (2000) Zinc deficiency up-regulates expression of high-affinity phosphate transporter genes in both phosphate-sufficient and -deficient barley roots. Plant Physiol 124:415–422. https://doi.org/10.1104/pp.124.1.415

Hussain S, Maqsood MA, Aziz T, Basra SMA, (2013) Zinc bioavailability response curvature in wheat grains under incremental zinc applications. Arch Agron Soil Sci 59(7):1001–1016

Hussain S, Maqsood MA, Rahmatullah (2011) Zinc release characteristics from calcareous soils using diethylenetriaminepentaacetic acid and other organic acids. Commun Soil Sci Plant Anal 42:1870-1881. https://doi.org/10.1080/00103624.2011.587571

Hussain S, Qaswar M, Ahmad F (2018) Zinc application enhances grain zinc density in genetically-zinc-biofortified wheat grown on a lowzinc calcareous soil. J Sci Agric 2:107–110

Jenik PD, Kathryn BM (2005) Surge and destroy: the role of auxin in plant embryogenesis. Development 132(3):577–585. https://doi.org/10.1242/dev.01952

Johnson CR, Thavarajah P (2013) The influence of phenolic and phytic acid food matrix factors on iron bioavailability potential in 10 commercial lentil genotypes (Lens culinaris L.). J Food Comp Anal 31:82–86. https://doi.org/10.1016/j.jfca.2013.04.003

Kaya M, Zeliha K, Albrahin E (2009) Phytase activity, phytic acid, zinc, phosphorus and protein contents in different chickpea genotypes in relation to nitrogen and zinc fertilization. Afr J Biotechnol 8(18):4508–4513

Khan GA, Bouraine S, Wege S, Li Y, de Carbonnel M, Berthomieu P, Poirier Y, Rouached H (2014) Coordination between zinc and phosphate homeostasis involves the transcription factor PHR1, the phosphate exporter PHO1, and its homologue PHO1;H3 in Arabidopsis. J Exp Bot 65:871–884. https://doi.org/10.1093/jxb/ert444

Liu ZH, Wang HY, Wang XE, Zhang GP, Chen PD, Liu DJ (2006) Genotypic and spike positional difference in grain phytase activity, phytate, inorganic phosphorus, iron, and zinc contents in wheat (Triticum aestivum L.). J Cereal Sci 44:212–219. https://doi.org/10.1016/j.jcs.2006.06.001

Lönnerdal B (2000) Dietary factors influencing zinc absorption. J Nutr 130(5):1378S–1383S

Manaf A, Raheel M, Sher A, Sattar A, Ul-Allah S, Qayyum A, Hussain Q (2019) Interactive effect of zinc fertilization and cultivar on yield and nutritional attributes of canola (Brassica napus L.). J Soil Sci Plant Nutr:1–7. https://doi.org/10.1007/s42729-019-00067-2

Maqsood MA, Hussain S, Naeem MA, Ahmad M, Aziz T, Raza HA, Kanwal S, Hussain M (2015) Zinc indexing in wheat grains and associated soils of Southern Punjab. Pak J Agric Sci 52:429–436

Marschner H, Cakmak I (1986) Mechanism of phosphorus-induced zinc deficiency in cotton. II. Evidence for impaired shoot control of phosphorus uptake and translocation under zinc deficiency. Physiol Plant 68:491–496. https://doi.org/10.1111/j.1399-3054.1986.tb03387.x

Marschner P, Rengel Z (2012) Nutrient availability in soils. In: Marschner P (ed) Marschner’s mineral nutrition of higher plants, 3rd edn. Academic Press, London, pp 315–330

Miller DD, Welch RM (2013) Food system strategies for preventing micronutrient malnutrition. Food Policy 42:115-128. https://doi.org/10.1016/j.foodpol.2013.06.008

Misson J, Raghothama KG, Jain A, Jouhet J, Block MA, Bligny R, Ortet P, Creff A, Somerville S, Rolland N, Doumas P, Nacry P, HerrerraEstrella L, Nussaume L, Thibaud MC (2005) A genome-wide transcriptional analysis using Arabidopsis thaliana Affymetrix gene chips determined plant responses to phosphate deprivation. Proc Natl Acad Sci U S A 102:11934–11939. https://doi.org/10.1073/pnas.0505266102

Pandey N, Pathak GC, Sharma CP (2006) Zinc is critically required for pollen function and fertilisation in lentil. J Trace Elements Medi Biol 20:89–96. https://doi.org/10.1016/j.jtemb.2005.09.006

Pandey SN, Gautam S (2009) Effects of zinc supply on its uptake, growth and biochemical constituents in lentil. Indian J Plant Physio 14(1):67–70

Pandey, S., Chandra , S., Raghuvanshi , R., Singh , G. A., Kumar , A., & Pandey , S. (2024). Screening of Lentil Genotypes Against Wilt of Lentil (Lens culinaris Medik L.) Caused by Fusarium oxysporum F. SP. Lentis in Glasshouse Condition. Journal of Scientific Research and Reports, 30(5), 772–777. https://doi.org/10.9734/jsrr/2024/v30i51996

Rahman MH, Wajid SA, Afzal M, Ahmad A, Awais M, Irfan M, Ahmad AUH (2013) Performance of promising lentil (Lens CulinarisMedik.) cultivars at different nitrogen rates under irrigated conditions of Faisalabad, Pakistan. CercetariAgronomiceî

Ram H, Sohu VS, Cakmak I, Singh K, Buttar GS, Sodhi GPS, Gill HS, Bhagat I, Singh P, Dhaliwal SS, Mavi GS (2015) Agronomic fortification of rice and wheat grains with zinc for nutritional security. Curr Sci 109:1171–1176 https://www.jstor.org/stable/24905830

Reid DE, Ferguson BJ, Hayashi S, Lin YH, Gresshoff PM (2011) Molecular mechanisms controlling legume auto regulation of nodulation. Annals of Bot 108:789–795. https://doi.org/10.1093/aob/mcr205

Rochfort S, Panozzo J (2007) Phytochemicals for health, the role of pulses. J Agric Food Chem 55:7981-7994. https://doi.org/10.1021/jf071704w

Seth A, Sarkar D, Masto RE, Batabyal K, Saha S, Murmu S, Das R, Padhan D, Mandal B (2018) Critical limits of Mehlich 3 extractable phosphorous, potassium, sulfur, boron and zinc in soils for nutrition of rice (Oryza sativa L.). J Soil Sci Plant Nutr 18:512-523. https://doi.org/10.4067/S0718-95162018005001601

Singh D, Singh H (2012) Effect of phosphorus and zinc nutrition on yield, nutrient uptake and quality of chickpea. Annals Plant Soil Res 14:71–74

Singh V, Kumar V, Karawasra SPS (1995) Interaction of S and Zn on dry matter yield, concentration and uptake of S in green gram. Crop Res 9:32–41

Singh, S. K., Panwar, J. D. S., Abbas, S., Ram, S. and Sirohi, G. S. (2005). Effects of zinc supply on its uptake, growth and biochemical constituents in lentil. Indian Journal of Plant Physiology, 31(4): 418-422.

Swain PS, Rao SB, Rajendran D, Dominic G, Selvaraju S (2016): Nano zinc, an alternative to conventional zinc as animal feed supplement:a review. Animal Nutr 2(3):134-141. https://doi.org/10.1016/j.aninu.2016.06.003

Thavarajah P, Thavarajah D and Vandenberg A (2009): Low phytic acid lentils (Lens culinaris L.): a potential solution for increased micronutrient bioavailability. J Agri Food Chem 57(19):9044–9049. https://doi.org/10.1021/jf901636p

Torun B, Bozbay G, Gultekin I, Braun HJ, Ekiz H and Cakmak I (2000): Differences in shoot growth and zinc concentration of 164 bread wheat genotypes in a zinc-deficient calcareous soil. J Plant Nutr 23(9):1251–1265. https://doi.org/10.1080/01904160009382098

Ullah A, Farooq M, Hussain M, Ahmad R and Wakeel A (2019): Zinc seed coating improves emergence and seedling growth in desi and kabuli chickpea types but shows toxicity at higher concentration. Int J Agric Biol 21:553-559. https://doi.org/10.17957/IJAB/15.0928

United Nations, Department of Economic and Social Affairs, Population Division (2015): World Population Prospects: The 2015 Revision, Key Findings and Advance Tables. Working Paper No. ESA/P/WP.241

United States Department of Agriculture (USDA) (2015): USDA Agricultural Projections to 2024. Office of the Chief Economist, World Agricultural Outlook Board, Long-term Projections Report OCE-2015-1. February

White PJ and Broadley M (2005): Biofortifying crops with essential mineral elements. Trends Plant Sci 10(12):586–593

White PJ and Broadley MR (2009): Biofortification of crops with seven mineral elements often lacking in human diets-iron, zinc, copper, calcium, magnesium, selenium and iodine. New Phytol 182:49–84.https://doi.org/10.1111/j.1469-8137.2008.02738.x

Zarcinase, R., Carias, L. and Reid, M. S. (1987): Estimation of zinc with the help of atomic spectroscopy Journal of Biological Chemistry 80: 549-54.

Zhao AQ, Tian XH, Cao YX, Lu XC and Liu T (2014): Comparison of soil and foliar zinc application for enhancing grain zinc content of wheat when grown on potentially zinc–deficient calcareous soils. J Sci Food Agric 94:2016–2022. https://doi.org/10.1002/jsfa.6518

Zou CQ *et al*. (2012): Biofortification of wheat with zinc through zinc fertilization in seven countries. Plant Soil 361:119–130. https://doi.org/10.1007/s11104012-1369-2a