

## ZINC BIOFORTIFICATION OF MUNGBEAN (*VIGNA RADIATA* L.) CULTIVARS THROUGH ZINC CHEMO-PRIMING

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### Abstract

Among micronutrients, zinc (Zn) is one of the most vital elements required for the growth and development of human and plants. The deficiency of Zn in Bangladesh's soil is causing a huge population of the country malnourished especially due to low Zn content of pulses like mungbean. A two year's research was conducted during pre-monsoon season of 2022 and 2023 in the Department of Crop Botany, Bangladesh Agricultural University, Mymensingh to evaluate the seed priming for Zn biofortification of recently released eight varieties of mungbean (BARI Mug-5, BARI Mug-6, BARI Mug-7 and BARI Mug-8 were grown during 2022 year, the other four varieties named BARI Mug-2, BARI Mug-3, BARI Mug-4 and BINA Mug-8 were grown during the growth season of 2023). Mungbean seeds were soaked for 6 hours before sowing in water (hydropriming), or aerated Zn solution (as osmopriming) of 0.01M or 0.05M with 1:5 seed to liquid ratio. The results showed that seed priming with 0.01M Zn remained unmatched by improving the yield attributes including plant height and number of primary branches per plant and grain yield of mungbean. In addition, seed priming with water and 0.01M Zn solution increased the grain yield by 7 and 15% and grain Zn concentrations by 9 and 14%, respectively compared to control. Overall, BARI Mug-3, BARI Mug-5 and BARI Mug-6 surpassed rest of varieties in terms of grains Zn content. Although Zn enrichment in mungbean grain was observed from the crops grown with primed seed through 0.05M Zn solution, but the performance of plant growth, yield traits and grain yield were noticed significantly poor. Based on recorded findings, it may be recommended that mungbean seeds might be primed with low concentrated zinc solution like 0.01M to attain better stand development and yield attributes, grain yield and grain Zn content.

**Key words:** Biofortification, Cultivars, Malnutrition, Mungbean, Pulse, Seed priming, Zinc deficiency.

### Introduction

Micronutrient deficiency hampers human growth and body functioning when the diet does not contain the right amount and proportion of minerals. It is termed as hidden hunger as one normally appears to be healthy, but frequently encounters illness and infections. Out of 25 trace elements, zinc (Zn) is quite essential for human health because it is a component of organic compounds such as carbohydrates, fats, proteins and vitamins (Stein, 2010). The Zn scarceness in the daily diets results in reduced immunity, stunting, risk to diarrheal and respiratory diseases along with increased risk for both mothers and infants during childbirth (Yakoob *et al.*, 2011; Hussain *et al.*, 2022). Additionally, it is involved in activating over 200 enzymes system which maintains immunity and growth (Hotz & Brown, 2004; Saboor *et al.*, 2021; Ozturk *et al.*, 2023). Recently, Zn nutrition is strongly emphasized globally to boost human immune system against COVID-19 Pandemic (Singh *et al.*, 2021). Interestingly, Zn is the second only to Fe among the mineral elements for which a human alimentary sine qua non has been established (Kaur *et al.*, 2014). Zn shortcoming is a dreadful trace element deprivation for human well-being across-the-board, which hits hard on top of one-third of the mankind. The Zn scarceness is

exceedingly all whereabouts of Bangladesh people and principally related to cereal or rice (monocrop cereal) based diet or otherwise intake of inadequate quality of meal (Rahman *et al.*, 2016). The recommended daily allowance of Zn is 8 and 11 mg/day for adult females and males, respectively. However, most people in Bangladesh couldn't intake this allowance.

Like humans, photosynthetic activity of crop plants is seriously affected by Zn deficiency due to change in chloroplast pigments (Kosesakal & Unal, 2009). The Zn plays key role in stabilization of ribosomal proteins, auxin formation and establishment of dehydrogenase enzymes (Hafeez *et al.*, 2013) and improves the crops productivity (Ullah *et al.*, 2019). Thus scarcity of Zn nutrient is a ubiquitous stumbling block both in humans and crop production leading to economic impact of 2-3% of GDP per annum in Bangladesh (World Bank, 2013).

Among the legumes, mungbean is the main conventional pulse with relatively higher protein contents (Mensah & Ihenyen, 2009; Islam *et al.*, 2023a; Islam *et al.*, 2024) and considered as poor man's meat due to higher protein content in comparison to cereals (Islam *et al.*, 2023b; Mahajan *et al.*, 2023; Ro *et al.*, 2023). Moreover, it has roughly 0% more carbohydrates content along with substantial amount of phosphorus (367 mg/100 g of seed) and calcium (132 mg 100 g of seeds) as well

(Faruque *et al.*, 2000). Nevertheless, mungbean grain is inherently low in Zn concentration. Thus, mungbean production with Zn-biofortification approach may be a feasible course of action to overcome Zn malnutrition to a great extent. Mungbean is easier to digest than other legumes (Rizvi *et al.*, 2012) and its utilization as oral rehydration solutions have been successfully used to treat children between three months and five years with acute diarrhea (Bhan *et al.*, 1987). Mungbean flour can be mixed with wheat flour, skim milk and sugar to prepare a better nutrition mixture for children (Imtiaz *et al.*, 2011; Lian *et al.*, 2023).

Mungbean cultivation has been reported to improve soil fertility through subsuming crop residue and atmospheric nitrogen (Kaisher *et al.*, 2010; Islam *et al.*, 2020; Islam *et al.*, 2024). Owing to its quick growing and short duration personality, wide range of cultivars that released recently and stony-hearted uniqueness to photoperiod it can grow throughout the year (Chauhan & Williams, 2018; Mehandi *et al.*, 2019). It flourishes better in wet environment, however can also be seeded under water deficit condition (Ahmad *et al.*, 2015). Now-a-days, mungbean is cultivated after the harvest of transplanted *Aman* rice on the vast lands of salinity prone southern coastal reach of Bangladesh during dry season where the other crops are difficult to grow due to habitat brininess (BARI, 2016).

Biofortification of food crops can be a feasible option to overcome emerging extensive dietary deficiency of Zn in human health (Haider *et al.*, 2018a). To achieve this goal, agronomic management practices especially seed priming (pre-sowing seed soaking or treatment) must be studied to explore bio-economical Zn fortification techniques for pulses. Seed priming is a cost-effective (Mushtaq *et al.*, 2023), easiest (Choudhary *et al.*, 2023) and environmentally benign (Yasir *et al.*, 2023) approach of biofortification to enhance Zn content which can potentially increase the growth and yield of pulse crops (Johnson *et al.*, 2005; Kaya *et al.*, 2007; Masuthi *et al.*, 2009). The Zn priming have resulted in encouraging results in terms of robust growth, yield enhancement and improved tolerance against abiotic stresses in different crops (Ahmad *et al.*, 2023; Donia & Carbone, 2023; Khalili *et al.*, 2023), however, research gaps exist regarding its efficacy for mungbean crop. Thus, we hypothesized that chemo-priming of Zn in different doses might effectively increase growth, yield attributes and grains Zn content in mungbean cultivars. Therefore, this study was designed to evaluate the impact of seed priming with Zn solution on growth, yield traits, grain yield, and enrichment of grain Zn content in mungbean cultivars.

## Material and Methods

**Experimental site:** The experiment was conducted during pre-monsoon spring season in the Field of the Department of Crop Botany, Bangladesh Agricultural University, Mymensingh (24°71'N latitude and 90° 42'E longitude at the elevation of 18 meter above the sea level). The experimental site is situated in the sub-tropical monsoon climatic zone. The location is characterized by

dry weather with low rainfall during November through April and enough rainfall with moist climate during the remaining period of the year. The topography of the experimental field was medium high land with fairly leveled and belonging to the Sonatala Soil Series of gray flood plain soil type under the Agro-Ecological Zone-9 (AEZ-9), named Old Bramhaputra Flood Plain. The experimental soil is silt loam in texture with imperfectly to poorly-drained permeable having pH 6.32.

**Planting materials:** Four mungbean varieties named BARI Mug-5, BARI Mug-6, BARI Mug-7 and BARI Mug-8 were grown during 2022; the other four varieties named BARI Mug-2, BARI Mug-3, BARI Mug-4 and BINA Mug-8 were grown during the growth season of 2023. Seeds of BARI Mug varieties were collected from Pulses Research Centre & RARS, BARI, Ishurdi, Pabna and BINA Mug-8 from Bangladesh Institute of Nuclear Agriculture (BINA), Mymensingh.

**Priming treatments:** The experiment was conducted following a Randomized Complete Block Design (RCBD) with four treatments viz. (i) control (unpriming), (ii) hydropriming (with water), and two osmopriming like (iii) priming with 0.01M Zn solution and (iv) 0.05M Zn solution, while each treatment was replicated thrice. Mungbean seeds were soaked in water (hydropriming) or aerated solution containing 0.01M Zn and 0.05M Zn or unprimed. Seeds were put in water or aerated solution for 6 hours, at room temperature with 1:5 seed to liquid ratio. After 6 hours, seeds were removed from priming media and washed four times with ordinary tap water. Thereafter, seeds were directly transferred to the prepared plots in the experimental site for sowing.

**Land preparation and fertilization:** The land was ploughed and cross-ploughed several times with a power tiller followed by laddering until a good tilth was achieved. Weeds and stubbles were removed as far as possible from the field before the final land preparation. The unit plot size was 3m×2m. The plots were fertilized with recommended doses of manures and fertilizers except Zn. In combination of 4 priming treatments and 4 mungbean varieties, there were 16 treatment combinations in each year where a treatment represents a unit plot. The total experimental area comprised 3 blocks (each represents a set of replication) where each block represents 16 treatments combination. Distance between block to block was 100 cm and plot to plot within a block was 50 cm.

**Seed sowing and crop husbandry:** The mungbean seeds were sown in the well-prepared plots as per experimental design following N-S row orientation with 40 cm row and 10 cm inter plant distance. The sowing date was 23 February in both 2022 and 2023 years. Three healthy seeds were sown in a sowing point and one healthy plantlet was kept after emergence. Weeding, irrigation, disease and insect pest management were performed as and when required to optimize the growth and development of crops.

**Data collection:** Data pertaining to plant height and number of primary branches per plant were recorded directly from 10 randomly selected plants in field condition at full bloom stage. Plant height was measured from ground level of stem to the tip or apex of the plant with a graduated meter scale. Primary branch indicates the branch that emerged directly from the main stem of plant having at least a distinct trifoliate leaf. Due to indeterminate types of flowering habit periodic pod harvest was performed. The mungbean plants were harvested on physiological maturity of crops when 80 percent pods become ripen. After sun-drying, the harvested plants threshed manually to compute the grain yield per plot.

Number of pods per plant was recorded as the average of that of 10 randomly selected plants from each plot. Fifty randomly selected pods from a replicated plot were considered to estimate the pod length and the number of grains per pod. Five samples of 1000 seeds from a plot's seed lot were taken and weighed to record the 1000-grain weight. Grain yield adjusted to 10 percent moisture condition and computed as ton per hectare.

**Analysis of grain Zn content:** The Zn content of experimented mungbean seeds were estimated after digestion with a perchloric and nitric- di-acid mixture (HClO<sub>4</sub> + HNO<sub>3</sub>; 1:2 ratio) solution (Jones & Case, 1990). The 500 mg seed samples were digested in the said acid mixture at 360°C for 1-2 hours until the digest became clear and colorless. After digestion, it was diluted to 50 ml with distilled water. After proper dilution of the extract, the Zn concentration in the extract was estimated by an Atomic Absorption Spectrophotometer (Perkin Elmer, CA, USA).

**Statistical analysis of recorded data:** The recorded data were subjected to two-way analysis of variance technique for determining the significance of employed treatments. Thereafter, Duncan's Multiple Range Test (DMRT) was put to use for the estimation of significance among treatment means at  $p < 0.01$  using statistical package of Statistix 8.1 version.

## Results

**Plant height and number of primary branches per plant:** Data for plant height and numbers of primary branch per plant are shown in (Tables 1 and 2), whereby the effects of various priming treatments on those vegetative growth traits was found statistically significant (at  $p < 0.01$ ). Although the seed priming either with water or 0.01M Zn solution produced taller plant with greater number of primary branches per plant during both the years studied. But the vegetative growth in terms of plant height and number of primary branches per plant was found poor when the crops were grown from the seeds primed with 0.05M Zn solution as compared to no priming.

Irrespective of the mungbean varieties during both years of 2022 and 2023, seed priming with 0.01M Zn solution significantly enhanced the vegetative growth as reflected by the taller plant with greater number of

primary branches per plant (Table 3). Irrespective of the priming treatments on the other hand, the effect of varieties was significant for plant height ( $p < 0.01$ ) but insignificant for the number of primary branches per plant in 2022 (Table 4). During the second year of experiment (2023), the varietal effect was insignificant for plant height but significant for the production of number of primary branches per plant ( $p < 0.05$ ) while BARI Mug-3 variety produced the higher number of primary branches followed by the BINA Mug-8 variety (Table 4).

**Yield traits:** During 2022, the effect of seed priming treatments on the all yield components of four varieties of mungbean was found significant except the grain size i.e. 1000-seed weight of BARI Mug-7 (Table 1). During 2023, the priming effects on the yield components of four varieties of mungbean differed significantly except the number of pods per plant of BINA Mug-8, pod length of BARI Mug-4 and the grain size of all the mungbean varieties (Table 2).

Irrespective of the varieties sown in 2022, priming treatments showed significant effect on the yield traits (Table 3). During the 2023 year, seed priming treatments also showed significant effect on the yield traits except 1000-grain weight. The performances of yield traits were found better and worst when the crops grown with the seeds subjected to priming with 0.01M Zn and 0.05M Zn solutions, respectively. The performances of yield traits were also found better when the seeds were primed only with water (i.e. hydropriming) as compared to no priming (control).

Irrespective of the priming treatments, the mungbean varieties exhibited significant but differential responses for the yield traits (Table 4). For example, BARI Mug-6 and BARI-8 produced higher number of pods per plant, BARI Mug-7 produced larger sized pods and grains with higher number of grains per pod in 2022. During the 2023 year, BARI Mug-2 and BARI-3 produced higher number of pods per plant but BARI Mug-4 and BINA Mug-8 ranked in higher position to exhibit higher number of grains per pod, BINA Mug-8 showed bigger grains. Irrespective of the years BARI Mug-2, BARI Mug-3, BARI Mug-4 and BARI Mug-8 varieties produced smaller sized grains as compared to the grained yielded by the other varieties.

**Grain yield:** Grain yield exhibited significant difference for the mungbean varieties and seed priming treatments (Tables 1 and 2). BARI Mug-7 and BINA Mug-8 varieties subjected to 0.01M Zn priming treatment produced the higher grain yield followed by crops grown with hydropriming and no priming during 2022 and 2023 years, respectively. Irrespective of the varieties in both years, higher grain yield was obtained from the crops when the seeds were primed with 0.01M Zn solution followed by the hydropriming (Table 3) (Fig. 1). The grain yield was found poor when the grains yielded from the crops grown with 0.05M Zn solution priming treatment. The varieties BARI Mug-7 and BINA Mug-8 produced significantly ( $p < 0.01$ ) higher grain yield followed by BARI Mug-5 and BARI Mug-6 while BARI Mug-2 variety produced the lower yield of grains (Table 4).

Table 1. Growth, yield traits and grain yield, and grain Zn content of four varieties of mungbean as affected by seed priming treatments during 2022.

Variety × Seed priming interaction	Plant height (cm)	No. of primary branches/plant	No. of pods/plants	Pod length (cm)	No. of grains/pod	1000-grain weight (cm)	Grain yield (t ha <sup>-1</sup> )	Grain Zn conc. (µg g <sup>-1</sup> )
<b>BARI Mug-5×</b>								
No priming	36.7 b	3.48 c	17.0 b	6.37 c	9.87 c	39.4 c	1.27 c	34.5 d
Hydropriming	37.1 b	3.50 b	17.3 b	6.70 b	10.47 b	40.3 b	1.42 b	37.3 c
Osmopriming_0.01M Zn	39.7 a	3.51 a	18.1 a	7.17 a	11.10 a	40.6 a	1.58 a	39.6 a
Osmopriming_0.05M Zn	32.9 c	3.22 d	14.3 c	5.82 d	9.03 d	37.2 d	1.16 d	38.7 b
α (n = 12)	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
<b>BARI Mug-6×</b>								
No priming	39.8 ab	3.87 c	19.0 c	7.30 d	9.53 c	42.1 c	1.49 c	35.1 d
Hydropriming	41.1 ab	3.93 b	19.8 b	7.60 b	9.93 b	43.2 b	1.56 b	37.9 c
Osmopriming_0.01M Zn	43.6 a	4.00 a	21.1 a	7.96 a	10.37 a	43.4 a	1.70 a	40.8 a
Osmopriming_0.05M Zn	37.3 b	3.61 c	14.4 c	6.57 c	8.77 d	40.9 c	1.34 d	39.0 b
α (n = 12)	0.05	0.01	0.01	0.01	0.01	0.01	0.01	0.01
<b>BARI Mug-7×</b>								
No priming	46.7 c	3.87 c	15.2 b	8.30 c	10.83 b	46.1	1.76 c	32.1 d
Hydropriming	48.2 b	3.95 b	15.8 ab	8.64 b	10.90 ab	46.4	1.84 b	36.0 c
Osmopriming_0.01M Zn	50.2 a	4.05 a	16.9 a	8.80 a	11.03 a	46.9	1.93 a	37.1 a
Osmopriming_0.05M Zn	43.7 d	3.48 d	13.5 c	7.80 d	10.13 c	45.4	1.60 d	36.7 b
α (n = 12)	0.01	0.01	0.05	0.01	0.05	NS	0.01	0.01
<b>BARI Mug-8×</b>								
No priming	43.5 c	3.70 c	18.2 c	5.77 c	9.70 c	27.6 c	1.27 c	32.1 d
Hydropriming	44.6 b	3.78 b	19.5 b	5.94 b	9.83 b	27.7 bc	1.42 b	35.9 c
Osmopriming_0.01M Zn	45.2 a	3.88 a	20.0 a	6.20 a	10.17 a	28.0 a	1.51 a	38.1 a
Osmopriming_0.05M Zn	39.7 d	3.41 d	17.8 d	5.31 d	8.87 d	26.9 d	1.18	37.3 b
α (n = 12)	0.01	0.01	0.01	0.01	0.01	0.05	0.01	0.01
LSD for all interactions (n = 48)	9.440	0.041	0.826	0.100	0.100	3.193	0.001	0.596

For a mungbean variety, the figures followed by a similar smaller letter do not differ significantly at 1% or 5% level of probability as per Duncan's Multiple Range Test (DMRT); NS = Not significant at  $p < 0.05$ ; α = Level of significance; LSD = Least significant difference at  $p < 0.01$ ; n = Number of observation

Table 2. Growth, yield traits and grain yield, and grain Zn content of four varieties of mungbean as affected by seed priming treatments during 2023.

Variety × Seed priming interaction		Plant height (cm)	No. of primary branches/ plant	No. of pods/ plants	Pod length (cm)	No. of grains/ pod	1000-grain weight (cm)	Grain yield (t ha <sup>-1</sup> )	Grain Zn conc. (µg g <sup>-1</sup> )
<b>BARI Mug-2×</b>									
No priming		42.4 c	3.46 c	19.7 ab	5.87 c	9.67 c	23.9	1.10 c	33.1 c
Hydropriming		44.2 b	3.56 b	20.3 a	6.13 b	9.90 b	24.0	1.18 b	35.0 b
Osmpriming_0.01M Zn		46.3 a	3.85 a	20.6 a	6.77 a	10.23 a	24.3	1.26 a	36.1 a
Osmpriming_0.05M Zn		40.1 d	3.22 d	18.2 c	5.39 d	8.90 d	23.7	1.01 d	36.2 a
α (n = 12)		0.01	0.01	0.05	0.01	0.05	NS	0.01	0.01
<b>BARI Mug-3×</b>									
No priming		42.5 c	3.97 c	19.5	5.34 c	10.53 c	26.0	1.15 c	35.1 d
Hydropriming		43.8 b	4.08 b	20.0	5.37 b	10.63 b	27.0	1.23 b	37.6 b
Osmpriming_0.01M Zn		44.5 a	4.11 a	20.4	5.53 a	10.73 a	27.3	1.36 a	38.2 a
Osmpriming_0.05M Zn		38.5 d	3.69 d	18.5	4.88 d	9.40 d	25.4	1.04 d	37.5 c
α (n = 12)		0.01	0.01	NS	0.01	0.01	NS	0.01	0.01
<b>BARI Mug-4×</b>									
No priming		40.5 c	3.63 c	16.5 a	5.99	10.60 c	30.7	1.23 c	29.1 d
Hydropriming		42.2 b	3.82 b	16.8 a	6.07	10.77 b	31.0	1.31 b	32.4 c
Osmpriming_0.01M Zn		43.0 a	3.93 a	17.3 a	6.16	10.93 a	31.2	1.40 a	34.2 a
Osmpriming_0.05M Zn		36.5 d	3.24 d	15.0 b	5.99	10.07 d	28.3	1.15 d	33.7 b
α (n = 12)		0.01	0.01	0.05	NS	0.01	NS	0.01	0.01
<b>BINA Mug-8×</b>									
No priming		38.9 c	3.94 c	15.6	8.16 c	10.63 ab	41.2	1.72 c	30.5 d
Hydropriming		40.5 b	3.99 b	16.1	8.49 b	10.87 a	41.3	1.82 b	33.2 c
Osmpriming_0.01M Zn		43.8 a	4.15 a	16.7	8.60 a	11.13 a	41.8	1.94 a	34.8 a
Osmpriming_0.05M Zn		34.8 d	3.54 d	15.1	7.55 d	9.53 c	40.1	1.61 d	34.4 b
α (n = 12)		0.01	0.01	NS	0.01	0.01	NS	0.01	0.01
LSD for all interactions (n = 48)		5.405	0.026	1.185	0.055	0.178	2.876	0.001	0.300
LSD for interaction considering both years <sup>y</sup> (n = 96)		8.951	0.041	1.106	0.082	0.173	3.065	0.001	0.431

For a mungbean variety, the figures followed by a similar smaller letter do not differ significantly at 1% or 5% level of probability as per Duncan's Multiple Range Test (DMRT); NS = Not significant at p<0.05; α = Level of significance; LSD = Least significant difference at P<0.01; n = Number of observation; <sup>y</sup>Considered the data in Tables 1 and 2

Table 3. Growth, yield traits and grain yield, and grain Zn content of mungbean as affected by seed priming treatments during 2022 and 2023.

Seed priming treatment (irrespective of mungbean variety)	Plant height (cm)	No. of primary branches/plant	No. of pods/plants	Pod length (cm)	No. of grains/pod	1000-grain weight (cm)	Grain yield (t ha <sup>-1</sup> )	Grain Zn conc. (µg g <sup>-1</sup> )
<b>a) 2022 year</b>								
No priming	41.7 c	3.73 c	17.4 c	6.94 c	9.98 c	38.8 c	1.45 c	33.5 d
Hydropriming	42.7 b	3.79 b	18.1 b	7.22 b	10.28 b	39.4 b	1.56 b	36.8 c
Osmopriming_0.01M Zn	44.7 a	3.86 a	19.1 a	7.53 a	10.67 a	39.7 a	1.68 a	38.9 a
Osmopriming_0.05M Zn	38.4 d	3.43 d	15.0 d	6.38 d	9.20 d	37.6 d	1.32 d	37.9 b
α (n = 12)	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
<b>b) 2023 year</b>								
No priming	41.1 c	3.75 c	17.8 c	6.34 c	10.36 c	30.4	1.30 c	31.9 d
Hydropriming	42.7 b	3.86 b	18.3 b	6.51 b	10.54 b	30.8	1.39 b	34.6 c
Osmopriming_0.01M Zn	44.4 a	4.01 a	18.8 a	6.77 a	10.76 a	31.1	1.49 a	35.9 a
Osmopriming_0.05M Zn	37.5 d	3.42 d	16.7 d	5.92 d	9.48 d	29.4	1.20 d	35.5 b
α (n = 12)	0.01	0.01	0.05	0.01	0.01	NS	0.01	0.01
LSD considering both years (n = 24)	2.825	0.015	0.471	0.016	0.063	0.996	0.001	0.055

For a year, the figures followed by a similar smaller letter do not differ significantly at 1% or 5% level of probability as per Duncan's Multiple Range Test (DMRT); NS = Not significant at  $p < 0.05$ ; α = Level of significance; LSD = Least significant difference at  $p < 0.01$ ; n = Number of pooled observations irrespective of mungbean varieties.

Table 4. Growth, yield traits and grain yield, and grain Zn content of mungbean as affected by variety during 2022 and 2023.

Mungbean variety (irrespective of seed priming treatment)	Plant height (cm)	No. of primary branches/plant	No. of pods/plants	Pod length (cm)	No. of grains/pod	1000-grain weight (cm)	Grain yield (t ha <sup>-1</sup> )	Grain Zn conc. (µg g <sup>-1</sup> )
<b>2022 year</b>								
BARI Mug-5	36.6 b	3.43	16.7 b	6.52 c	10.12 b	39.4 ab	1.36 c	37.5 a
BARI Mug-6	40.4 ab	3.85	18.6 a	7.36 b	9.65 c	42.4 a	1.52 b	38.2 a
BARI Mug-7	47.2 a	3.84	15.4 c	8.38 a	10.73 a	46.2 a	1.78 a	35.5 b
BARI Mug-8	43.2 ab	3.69	18.9 a	5.80 d	9.64 c	27.6 c	1.35 d	35.8 b
α (n = 12)	0.01	NS	0.01	0.01	0.01	0.01	0.01	0.01
<b>2023 year</b>								
BARI Mug-2	43.2	3.52 d	19.7 a	6.04 b	9.68 c	24.0 d	1.14 d	35.1 b
BARI Mug-3	42.3	3.96 a	19.6 a	5.28 c	10.33 b	26.4 c	1.20 c	37.1 a
BARI Mug-4	40.5	3.66 c	16.4 b	6.02 b	10.59 a	30.3 b	1.27 b	32.4 d
BINA Mug-8	39.5	3.91 b	15.9 c	8.20 a	10.54 a	41.1 a	1.77 a	33.2 c
α (n = 12)	NS	0.05	0.01	0.01	0.05	0.01	0.01	0.01
LSD considering both years (n = 24)	9.896	0.032*	0.541	0.091	0.139	2.213	0.001	0.400

For a year, the figures followed by a similar smaller letter do not differ significantly at 1% or 5% level of probability as per Duncan's Multiple Range Test (DMRT); NS = Not significant at  $p < 0.05$ ; α = Level of significance; LSD = Least significant difference at  $p < 0.01$  or  $p < 0.05$  (\*); n = Number of pooled observations irrespective of priming treatments

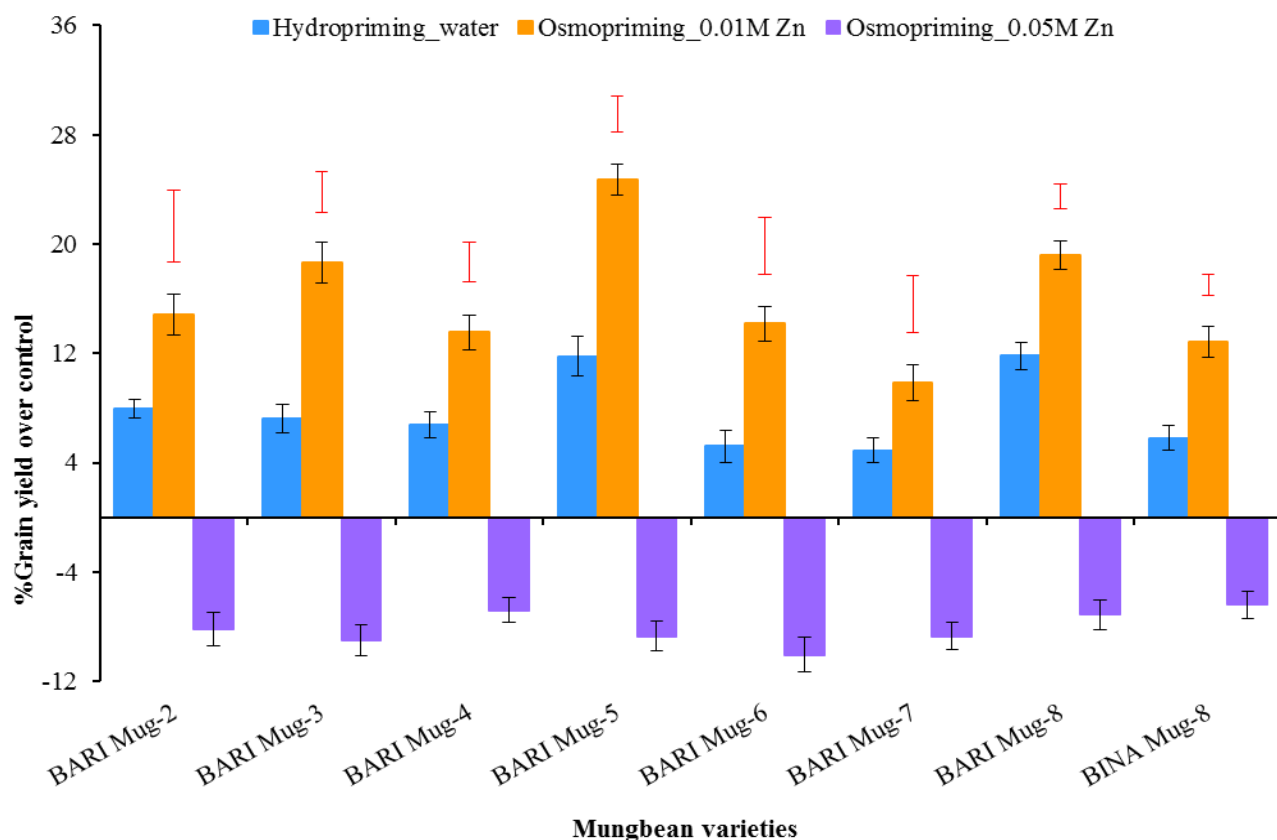


Fig. 1. Percentage of grain yield over control in eight varieties of mungbean. Attached vertical bars represent the standard deviation ( $\pm$ ) of meanwhile the detached vertical bar indicates the Least Significant Difference (LSD) for a variety at 1% level of probability.

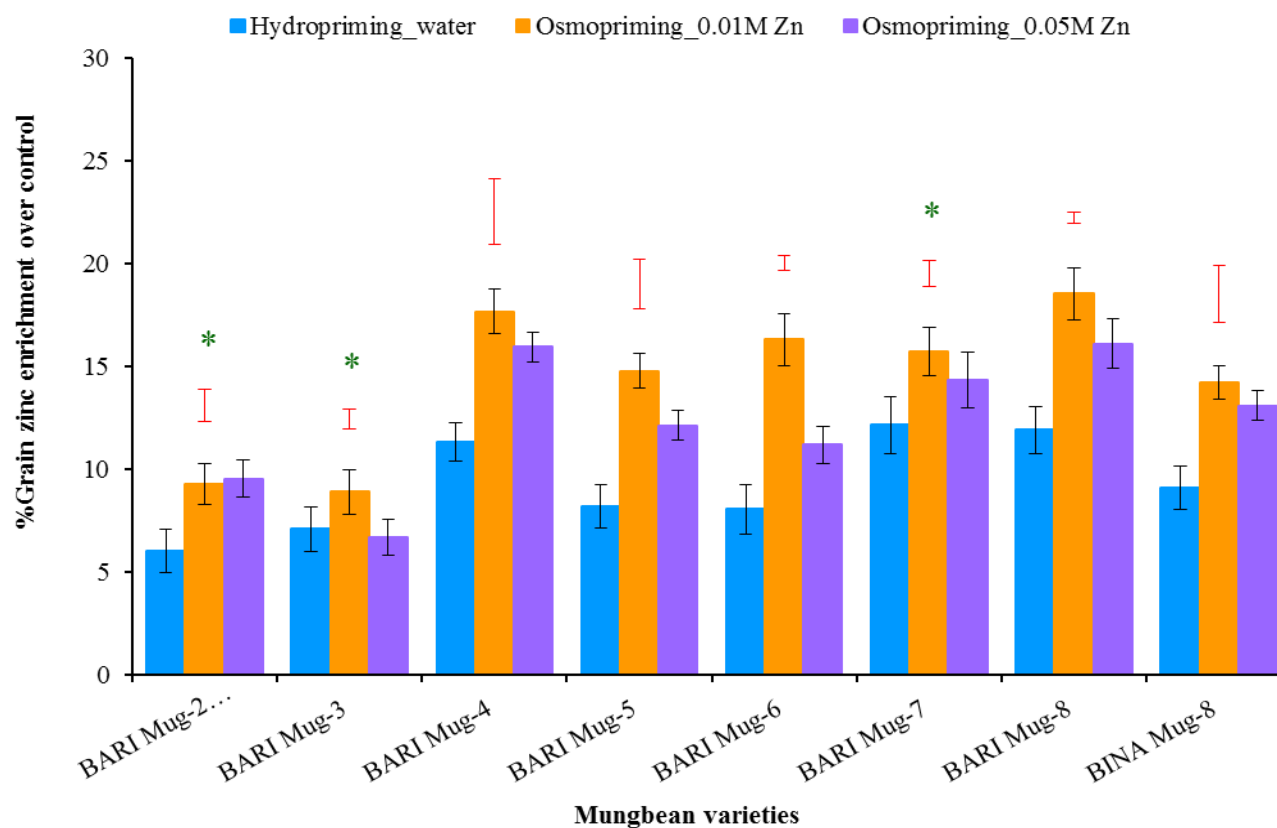


Fig. 2. Percentage of grain Zn increase over control in eight varieties of mungbean. Attached vertical bars represent the standard deviation ( $\pm$ ) of meanwhile the detached vertical bar indicates the Least Significant Difference (LSD) for a variety at 1% or 5% (\*) level of probability.

**Grain Zn concentration:** The effect of seed priming treatments on grain Zn enrichment for different varieties of mungbean was found significant (Tables 1 and 2). Higher amount of Zn accumulation was noticed in the grains from all varieties of mungbean crops during both the years studied when the seeds were primed in the solution media as compared to the unprimed crops (Fig. 2). Satisfactory Zn accumulation in grain was noticed when the seeds were primed with 0.01M Zn solution followed by the 0.05M Zn solution and water (i.e. hydropriming) while crops grown with unprimed seed attracted lower content of Zn in the seeds (Table 3). Lower grain Zn content was noticed as  $29.1 \mu\text{g g}^{-1}$  from BARI Mug-4 while higher Zn found as  $40.8 \mu\text{g g}^{-1}$  from BARI Mug-6 crops grown with unprimed seeds and the seeds primed through 0.01M Zn solution, respectively. Irrespective of the priming treatments, grains produced from BARI Mug-3, BARI Mug-5 and BARI Mug-6 crops attracted higher amount of Zn followed by the grains produced by the BARI Mug-7 variety (Table 4).

## Discussion

The results of this two year's field study reflect that the mungbean's eight varieties in response to 0.05M Zn primed and un-primed seeds had poor stand establishment in terms of plant height and number of primary branches per plant, yield contributing traits and grain yield as compared to these of the crops grown with seed priming through 0.01M Zn solution or simply water. The underlying reason still remains unclear but it might be attributed to slow movement and utilization of higher quantities of applied Zn internally. However, seeds primed with Zn at 0.01M solution achieved the high and mighty vegetative growth, yield components, grain yield and enhanced the grain-Zn concentration. These might be attributed due to the involvement of Zn in better root growth and functioning with foremost resource acquisition, and thus higher cell division, photosynthesis, protein synthesis, retaining membrane structure and providing the resistance against pathogen (Misra *et al.*, 2005; Alberto *et al.*, 2008; Samreen *et al.*, 2017; Ji *et al.*, 2022). Such breakthrough might be attributed to accessibility of Zn indispensable for the biosynthesis of carbohydrates, lipids, nucleic acids and protein (Khan *et al.*, 2022) which are inevitable for the vigorous growth of crop plants and grain yield.

The highest grain yield from all the varieties was documented from the mungbean crops when grown with primed seeds with 0.01M Zn solution which is a proof of forwarding in growth and yield contributing traits like the number pods per plant and seeds per pod, and grain size or weight. In fact, these are the main yield attributes of most of pulse crops and any furtherance on those components tend to enhance the grain yield as evident from these recorded findings during both years. Haider *et al.*, (2020) and Haider (2021) recently reported that mungbean seed priming with a low concentration of Zn like 0.01M Zn solution remarkably enhanced seedling development, crop growth, yield attributing traits and grain yield. Similar growth and yield benefits due to seed

priming with Zn in the other crops have also been extensively documented (Harris *et al.*, 2007, 2008; Imran *et al.*, 2013, 2015; Reis *et al.*, 2018; Choukri *et al.*, 2022).

As seed priming is a partial hydration technique that enables pre-metabolic activities without radicle protrusion (Farooq *et al.*, 2012), thus different cultivars responded differently to chemo-priming. The better performance and quality improvement after priming seeds with Zn by mungbean cultivars might be attributed to variation in their genetic make-up which caused difference in their potential to uptake and utilize Zn. Previously, it was reported that crops cultivars tend to respond differently to Zn priming initially owing to genetic variations and latterly due to agro-botanical traits increase this differential response and thus crop-specific and variety-specific studies must be performed to ascertain the effectiveness of Zn chemo-priming (Tabasum *et al.*, 2010; Sen & De, 2017; Haider *et al.*, 2021; Mwangi *et al.*, 2021).

Although the crops grown with seed priming through 0.05M Zn solution showed better Zn enrichment in grain but it resulted in lower grain yield by all varieties under investigation. In contrast, seed priming with 0.01M Zn solution not only increased the grain yield and but also enhanced the grain Zn concentration which in reality is a good line of action to produce mungbean grains having plentiful load of zinc. This would be very helpful to reduce malnutrition owing to Zn deficiency in human population where dearth of zinc is run-of-the-mill as like in Bangladesh. There are some other agronomic approaches to apply zinc in crops including mungbean like soil and foliar applications (Haider *et al.*, 2018a, b). However, our research findings confirm that seed priming technique may offer a very simple or easy and cost effective way that might not only be used for boosting grain yield with higher economic returns but also for Zn biofortification in mungbean grains. Thus, improved Zn concentration in mungbean grains harvested in this study indicated that seed priming through low concentration of Zn solution might be developed as a viable option to reduce Zn malnutrition in humans under changing climate and increasing food security threats.

## Conclusion

Variation of mungbean cultivars in terms of growth and yield considerations were observed in this two-year trial in response to seed priming treatments with Zn. All eight genotypes of mungbean responded positively to seed priming through water (i.e. hydro-priming) or 0.01M Zn solution (osmopriming). These seed priming treatments improved the vegetative growth in terms of plant height and number of primary branches per plant, yield traits and grain yield than the crops grown with 0.05M Zn primed and unprimed seed (control or no priming). Nonetheless, grain zinc concentration was enhanced by seed priming with 0.01M Zn solution as compared to the crops grown with hydropriming and control treatment. Although zinc biofortification was noticed in the mungbean crops grown with higher concentration of zinc like 0.05M Zn priming solution



but growth and grain yield were drastically reduced. Thus, seed priming with low concentration of Zn solution might be an efficient option to enhance grain yield and higher accumulation of Zn in mungbean grains, and this might help to reduce Zn malnutrition in human population in Bangladesh and other South Asian countries like India and Pakistan.

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