

Effect of Zinc Activities on Shoot, Root Biomass and Phosphorus Uptake in Wheat Genotypes

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Abstract: An experiment was conducted to find out the variation in tolerance toward Zn deficiency among four different wheat genotypes using chelate-buffered nutrient solution of three Zn²⁺ activities. The genotypes (NRL-1027, NRL-1241, NRL-1242 and NRL-1243) were transplanted to nutrient culture solution in a net house for 45 days to find the impression of various levels of Zn²⁺ activity (2, 10 and 40 pecomolar (pM)) on shoot and root biomass. Among all the wheat genotypes, “NRL-1242” exhibited the highest tolerance to Zn deficiency and hence categorized as Zn-efficient, whereas “NRL-1243” proved to be Zn-inefficient. The remaining two genotypes were medium in efficiency on the above criteria. Efficient wheat genotype “NRL-1242” showed less reduction in growth and accumulated higher P at all lower Zn activities. Thus on the basis of survival in low Zn availability genotypes “NRL-1242” may be used by the researchers to plan their breeding experiments and to set genotype specific recommendations to Zn deficient soils.

Key words: Wheat • Zinc • Deficiency • Phosphorous • Biomass

INTRODUCTION

Wheat is a main staple food in Pakistan. It is mainly planted on calcareous soil which has low plant available Zn [1]. Diethylenetriaminepenta acetic acid (DTPA) extractable Zn in the calcareous soil averages as 0.37 $\mu\text{g g}^{-1}$, whereas a critical value of 0.46 mg kg^{-1} is required for optimal wheat growth [2]. The low level of Zn in soil has a negative effect on wheat yield and nutritional value [3]. In a global study initiated by FAO, it has been shown that about 30 % of the cultivated soil of the world contains low amount of Zn available to plant [4] and Zn deficiency is prominent in the crops grown on these soils.

Zinc acts as a functional, structural, or regulatory cofactor of over 300 enzymes and these are involved in cell division, nucleic acid and protein metabolism[5]. Zinc is component of plant CA and the presence of CA in the chloroplast of C3 plants was considered evidence for its involvement in maintaining the internal bicarbonate pool of the chlorophyll or, possibly, its association with ribose biphosphate carboxylase in CO₂ fixation.

In C4 plants, the CA present in the cytosol of mesophyll cells specifically catalyses the conversion of CO₂ to HCO₃⁻ which is then assimilated by carboxylase [6]. Even though wheat is classed as less sensitive to low available Zn and also showing[7] great genotypic variation to Zn deficiency but in Pakistan still extremely affected by this condition.

Soil Zn deficiency reduces both grain yield and quality [8] and may lead to human Zn deficiency, especially in developing countries where diets are abundant in cereal-based foods and deficient in animal protein [9,10]. Recent improvements in the technique of using a nutrient solution with a buffer chelates micronutrients free ionic format appropriate levels low, it mimics the situation on the ground, has enabled the study of genotypic differences in micronutrient intake in a more realistic, while still maintaining all the advantages of solution culture system [11-13]. Early experiments have shown that the chelate-buffered nutrient solution system can be used to distinguish definitely different levels of Zn efficiency of wheat genotypes [14].

Due to lack of information on the adaptation of the cultivars on Zn deficient soils, Zn deficient susceptible cultivars are cultivated which also intensify the problem of Zn deficiency in wheat. Wheat cultivars greatly differ in their adaptation to low Zn, therefore this study was initiated to screen four wheat genotype to classify it into various categories on the basis of their adaptation ability to varying zinc activities.

MATERIALS AND METHODS

An experiment was conducted to study effect of zinc activities on shoot, root biomass and phosphorus uptake in wheat genotypes in the net house of Nuclear Institute for Food and Agriculture (NIFA) Tarnab, Peshawar, Pakistan during the year 2012-2013. To select the “Zn-efficient and inefficient wheat genotypes”, seeds of 4 wheat genotypes (NRL-1027, NRL-1241, NRL-1242 and NRL-1243) were grown in chelate-buffered nutrient solution of three Zn²⁺ activities i.e. 2 (deficient), 10 and 40 (adequate) pM making concentration of 0.1, 0.5 and 2 µM. The chelate-buffered nutrient solution containing full strength of (Zn²⁺ and K₃HEDTA) and half strength of all macro nutrients and micro nutrients. After 10 days full strength solutions was used.

The seeds of 4 wheat genotypes were surface-sterilised and rinsed with double deionised water. The seeds were then submerged in double deionised water for three hours in separate Pyrex glass beakers with the cultivar name clearly marked. The soaked seeds were then placed on moist filter paper in Petri dishes. These Petri dishes were placed in incubator at 20 ± 1 °C till germination and transplanting.

Seven days after germination, the seedlings were large enough to be held in the holes of thermo pore sheet. From incubator two uniform seedlings were transplanted into foam plugged holes of white thermo pore sheet floating on continuously aerated 20 L filled chelate-buffered nutrient solution. The fresh mixture of nutrient culture solution was replaced after day 10, 18, 25, 30, 35, 40 following transplantation. The stock solutions were made in 1 L volumetric flask and were kept refrigerated. Metal chelates were prepared by mixing HEDTA and the metals on an equal molecular weight basis for a molar solution. Fresh working solutions were made up from these stock solutions as and when needed. A required quantity of stock solution for each element except Zn was taken in stainless steel container (pre-washed with 3% HCl and double-deionised water) and volume was made up to 20L. Zinc solution required for each treatment was

added directly to the individual container from the stock solution. The pH of the nutrient solution was monitored on daily basis and adjust with 0.1M HCl or 0.1M KOH at 6.0 ± 0.01.

The experiment was conducted in the net house with the conditions set up to operate at minimum 2°C and maximum 18°C with mean value of 10°C. After 45 days of transplantation plant were harvested; three lots of deionised water were used to root and shoot samples, than wash with double deionised water. The washed samples were air dried with tissue paper and record a fresh weight. The air dried sample were than dried for 48 hours in force drought oven at 80 ± 1 °C. The dried samples were stored in a desiccator. The oven dried samples of wheat (tops and roots) were then finely grinded in a mechanical grinder (MF 10 IKA, Werke, Germany) to pass through a 1 mm sieve. The grinded samples were then stored in paper bags labeled properly for further analysis. Total P was also determined by digesting the plant material in HNO₃:HClO₄ mixture prepared in 5:1 ratio. The digested material was analysed for total P by metavanadate yellow colour method as described by Jonathan [15]. Data were analyzed statistically using complete randomize block design and differences among treatments were evaluated [16].

RESULTS AND DISCUSSION

The significant ($P \leq 0.05$) effects of genotypes and Zn²⁺ activities were observed on dry weight of shoots and roots, length of shoots and roots, phosphorus and micronutrients (Zn, Fe, Cu and Mn) uptake in shoots and roots, Zn efficiency, growth reduction. The data are presented and discussed below:

Shoot and Root Dry Matter: Different Zn²⁺ activities in solution had a significant impact on the growth of wheat plants (Fig. 1). As Zn²⁺ activities in the culture solution were increased, the growth of the plants was increased and plants yielded higher dry matter production. Some genotypes showed better growth at 2pM Zn²⁺ activity than others and even some produced more dry matter at 40 pM Zn²⁺ than others. This variation in the growth of genotypes at different Zn²⁺ activities was exploited for the calculation of Zn efficiency. Higher dry weight of shoot (7.6 g pot⁻¹) is observed in genotype “NRL-1243” at 40pM Zn²⁺ activity and the lowest shoot dry matter yield (2.0 g pot⁻¹) was produced by the same genotype at 2pM Zn²⁺ activity. Due to low shoot dry matter production at Zn-deficiency conditions (2pM Zn²⁺),

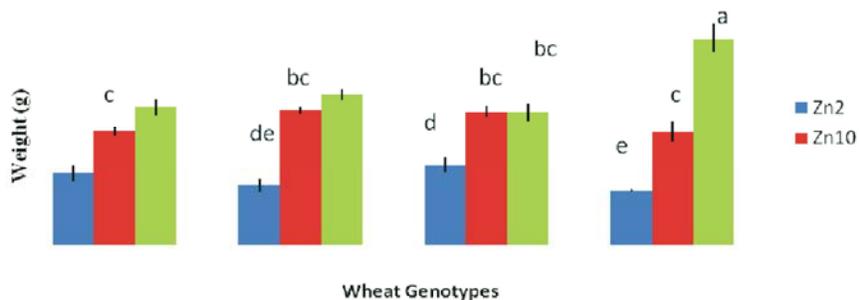


Fig. 1: Effect of Zn²⁺ activity on shoot dry matter

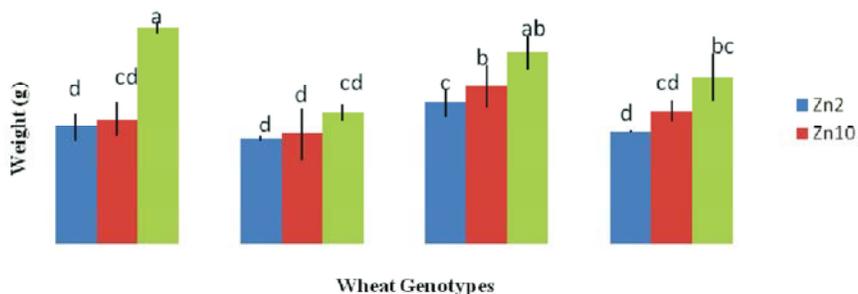


Fig. 2: Effect of Zn²⁺ activity on root dry matter

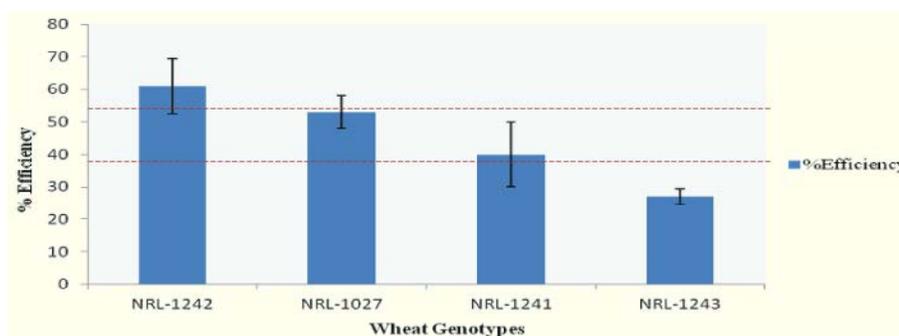


Fig. 3: Effect of Zn²⁺ activity on Zinc-efficiency

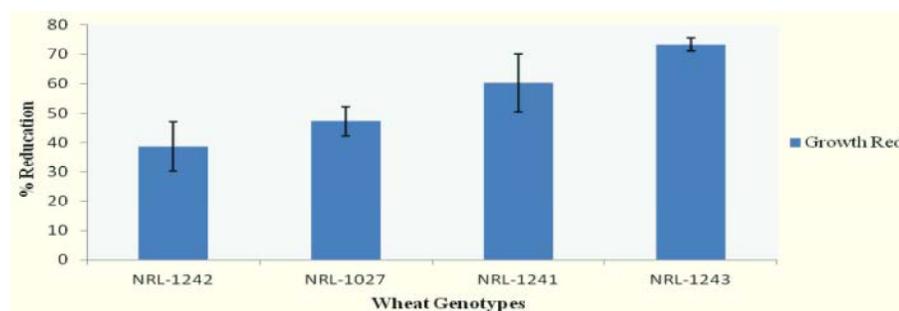


Fig. 4: Effect of Zn²⁺ activity on growth reduction

genotype “NRL-1243” proved to be Zn-inefficient and genotype “NRL-1242” showed higher dry matter production at 2pM Zn²⁺ activity, proved to Zn-efficient. The “NRL-1027” and “NRL-1241” has been considered

Zn-medium genotypes. As with the shoot dry matter, Zinc activity in the solution also affected the production of root dry matter by different genotypes, as Zn activity in solution was increased, the root dry matter production by

Table 1: Shoot and root Phosphorus uptake in different wheat genotypes

Genotypes	P (mg pot ⁻¹) Shoot			P (mg pot ⁻¹) Root		
	2pM	10pM	40pM	2pM	10pM	40pM
NRL-1027	28.0 abcd	37.9 a	21.5 cd	2.8 bcd	5.6 a	1.2 d
NRL-1241	28.1 abcd	28.8 abcd	23.4 bcd	3.6 b	1.5 d	1.3 d
NRL-1242	29.8 abcd	37.3 ab	19.9 d	3.5 b	4.3 ab	1.2 d
NRL-1243	28.3 abcd	34.2 abc	32.4 abcd	2.7 bcd	3.4 bc	1.8 cd
LSD(<0.05)	14.20			1.7		

Means with different letter(s) in rows and columns are statistically significant at $P \leq 0.05$

the genotypes increased significantly (Fig. 2). Zinc application increased the dry matter production of wheat genotypes, however dry matter production varied from cultivar to cultivar [17]. This variation in dry matter production of cultivars was used to differentiate Zn²⁺ efficiencies of cultivars [18]. Carbonic anhydrase is an enzyme which is involved in the photosynthetic process and under the condition of Zn deficiency its activity in wheat plant is reduced [19], resulting lower dry matter production.

Zn Efficiency (%) and Growth Reduction (%): Zinc efficiency (%) was calculated as the ratio of shoot dry matter produced under zinc deficiency (2pM Zn²⁺) to Zinc adequate (40 pM Zn²⁺) by method of Torun *et al.* [18]. Zinc efficiencies among tested genotypes varied from 27% to 61% (Fig. 3). The genotypes which produced a lower reduction in growth had a higher Zn efficiency. The genotype “NRL-1242” proved to be highly Zn-efficient having Zn efficiency (61%), while “NRL-1243” had 27%. According to this classification “NRL-1242” genotype was Zn-efficient, “NRL-1243” Zn-inefficient and the rest of two genotypes are medium in Zn efficiency. Significant variation was observed in growth reduction within genotypes as presented in Fig. 4. Zn-efficient genotypes showed a lower reduction in growth and Zn-inefficient genotypes showed higher reduction in growth. Zinc efficiency was determined as explained by Rengel and Graham [7] that ability of a genotype to grow and yield well under Zn²⁺ deficiency condition, all the wheat genotypes were classed as Zn-efficient and Zn-inefficient. Zinc efficiency of wheat genotypes is related to carbonic anhydrase activity as discussed by Rengel and Graham [7]. The Zn-efficient cultivars maintain higher Carbonic anhydrase activity than Zn-inefficient genotypes under Zn²⁺ deficient condition and high carbonic anhydrase activity in Zn-efficient genotypes maintain high rate of photosynthesis which lead to more yield. The genotypes proved as Zn-inefficient showed greatest reduction in dry matter yield as also observed by Cakmak *et al.* [20].

Phosphorus Uptake: The different Zn²⁺ activities have significant effect on shoots P uptake by wheat plants. Phosphorus uptake was higher in the Zn-inefficient than Zn-efficient genotype (average of three Zn²⁺ activities). It was observed from the data (Table 1) that P uptake was increased statistically with increasing Zn treatment from 2 to 10 pM Zn²⁺ and further increase in Zn treatment decreased P uptake significantly. The high P uptake 37.9 mg pot⁻¹ was observed in genotype “NRL-1027” and lowest in Zn-efficient genotype “NRL-1242” which is 19.9 mg pot⁻¹ at 40pM Zn²⁺ activity. The shoot P uptake varied between genotypes from 19.9 to 37.9 mg pot⁻¹. The average P uptake in the roots of the plants is lower in genotype not tolerant to Zn compare to Zn-efficient genotypes. The P uptake varied between cultivars from 1.2 to 5.6 mg pot⁻¹ (Table 1). These results are in agreement to those of Zeinab *et al.* [21], who observed that P uptake was higher in Zn-sufficient plants compare with Zn-deficient plant and the lack of enhanced P uptake may have been due to a period of Zn²⁺ deficiency to which the plants had been exposed. In Zn deficient plant high uptake of P is caused by improper mechanisms to control the release of P from root cells into the xylem and the distribution of P from shoots to roots affected and lead to higher accumulation or toxic concentrations of P in the leaves of plant showing Zn deficiency [22].

CONCLUSIONS

Significant interaction among genotypes and Zn activity for dry matter production and Zn contents indicated large genetic variations which can be exploited to select more Zn efficient wheat genotypes. The cultivar “NRL-1242” proved to be Zn efficient which accumulated higher Zn and more biomass production at Zn deficient level compared to the Zn-inefficient genotypes. It is recommended that Zn efficient genotypes may be cultivated which grow and yield well under Zn deficient condition and reduce the requirement for Zn fertilizer.

REFERENCES

1. Siddiqui, S. and R.A. Khattak, 2010. Trace elements fraction atom in calcareous soils of Peshawar-Pakistan. *Soil Environ.*, 29(2): 148-158.
2. Yang, X.E., W.R. Chen and Y. Feng, 2007. Improving human micronutrient nutrition through biofortification in the soil-plant system: China as a case study. *Environmental Geochemical Health*, 29(5): 413-428.
3. Liu, Z., 1994. Regularities of content and distribution of Zn in soils of China. *Scientia Agricultura Sinica*, 27(1): 30-37.
4. Sillanpaa, 1982. Micronutrients and Nutrients Status of Soils: a Global Study. FAO Soil Bulletin No. 48. FAO, Rome.
5. Marschner, H., 1986. Mineral Nutrition of Higher Plants. New York: Academic Press.
6. Hacisalihoglu, G., J.H. Jonathan, Y. Wang, I. Cakmak and L.V. Kochian, 2003. Zinc efficiency is correlated with enhanced expression and activity of Zn-requiring enzymes in wheat. *Plant Physiology*, 131: 595-602.
7. Rengel, Z. and R.D. Graham, 1995. Importance of seed Zn contents for wheat growth differing in Zn efficiency. *Physiol. Plant*, 95: 604-612.
8. Lombnas, P. and B.R. Singh, 2003. Varietal tolerance to Zn deficiency in wheat and barley grown in chelator-buffered nutrient solution and its effect on uptake of Cu, Fe and Mn. *Journal of Plant Nutrition and Soil Science*, 166(1): 76-83.
9. Cakmak, I., M. Kalayci and H. Ekiz, 1999. Zinc deficiency as a practical problem in plant and human nutrition in Turkey: A NATO-science for stability project. *Field Crops Research*, 60(1-2): 175-188.
10. Ackland, M.L. and A. Michalczyk, 2006. Zinc deficiency and its inherited disorder-a review. *Genes and Nutrition*, 1(1): 41-50.
11. Chaney, R.L., P.F. Bell and B.A. Coulombe, 1989. Screening strategies for improved nutrient uptake and utilization by plants. *Horticulture Science*, 24: 565-72.
12. Norvell, W.A. and R.M. Welch, 1993. Growth and nutrient uptake by barley studies using an N-(2-hydroxyethyl) ethylenedinitrioltri-acetic acid-buffered nutrient solution technique (I. Zinc ion requirements). *Plant Physiology*, 101: 619-25.
13. Welch, R.M. and W.A. Norvell, 1993. Growth and nutrient uptake by barley studies using an N (2-hydroxyethyl) ethylenedinitrioltri-acetic acid-buffered nutrient solution technique. II. Role of zinc in the uptake and root leakage of mineral nutrients. *Plant Physiology*, 101: 627-31.
14. Rengel, Z. and R.D. Graham, 1993. Physiological basis of differential Zn efficiency among wheat genotypes. In: Abstracts of the 33rd meeting of Australian Society of Plant Physiologists, Perth, pp: 34.
15. Jonathan, J.H., 1998. Characterization of Zn uptake, Binding and translocation in intact seedlings of bread and durum wheat cultivars. *Plant physiology*, 118(1): 219-226.
16. Ambler, J.E. and J.C. Brown, 1969. Cause of differential susceptibility to Zn deficiency in two varieties of navy beans (*Phaseolus vulgaris* L.). *Journal of Agronomy*, 61(1): 41-43.
17. Imtiaz, M., B.J. Alloway, P. Khan, M.Y. Memon, S.U. H. Siddiqui, M. Aslam and S.K.H. Shah, 2006. Zinc deficiency in selected cultivars of wheat and barley as tested in solution culture. *Communications in Soil Science and Plant Analysis*, 37(11-12): 1703-1721.
18. Torun, B., G. Bozbay and I. Gültekin, 2000. Differences in shoot growth and Zn concentration of 164 bread wheat genotypes in a Zn-deficient calcareous soil. *Journal of Plant Nutrition*, 23(9): 1251-1265.
19. Rengel, Z. and R.D. Graham, 1995a. Wheat genotypes differ in Zn efficiency when grown in chelate-buffered nutrient solution, I. Growth. *Plant and Soil*, 176(2): 307-316.
20. Cakmak, I., H. Ekiz and A. Yilmaz, 1997. Differential response of rye, triticale, bread wheat and durum wheats to Zn deficiency in calcareous soils. *Plant and Soil*, 188(1): 1-10.
21. Zeinab, A.S. and M.E. Fouly, 2008. Evaluation the efficiency of some Egyptian wheat (*Triticum aestivum* L). Cultivars to Zn deficiency through Peroxidase activity and Protein Profile techniques. *Not. Bot. Hort. Agrobot. Cluj*, 36(2): 42-46.
22. Loneragan, J.F., D.L. Grunes and R.M. Welch, 1982. Phosphorus accumulation and toxicity in leaves in relation to Zn supply. *Soil Science Society of American Journal*, 46(2): 345-352.