

POTASSIUM ENHANCED GRAIN ZINC ACCUMULATION IN WHEAT GROWN ON A CALCAREOUS SALINE-SODIC SOIL

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Abstract

Zinc (Zn) and iron (Fe) dense wheat grains are required for malnourished populations groups. Nutrient and water imbalances under calcareous saline-sodic soils decrease mineral accumulation in wheat grains. On such soils, the influence of soil potassium (K) application was studied on the accumulation of native soil and foliar-applied minerals (Zn, Fe,) in wheat grains. To field-grown wheat, two K rates were applied (0 or 35 kg ha⁻¹) at sowing in combination with four levels of foliar micronutrient sprays (control, 2×0.25% Zn w/v, 2×0.25% Fe w/v or combined Zn + Fe) at anthesis. Zinc and Fe were increased with respective micronutrient sprays. Potassium application increased grain yield, grain concentration of K and Zn in the grain. Grain Zn concentration was maximum (33 mg Zn kg⁻¹) with combined soil-applied K + foliar-applied Zn. As compared to respective foliar treatments in plots not supplied with K, grain Fe concentration was increased with soil K application in plots sprayed with Zn alone. Conclusively, optimum soil K supply increased accumulation of native soil and foliar-applied Zn in wheat grains.

Key words: Calcareous salt-affected soils; Potassium nutrition; Spring wheat; Zinc biofortification.

Introduction

More than 50% of the world's total wheat is produced and consumed in the developing countries (FAO, 2018). Wheat grains, therefore, have fundamental importance in fulfilling daily energy and nutrition requirements of the people living in those countries. Most of the cultivated wheat varieties have lower grain concentration of zinc (Zn) and iron (Fe) than old cultivars (Fan *et al.*, 2008). This might be due to unconscious breeding for higher grain weights to meet yield targets (Hussain *et al.*, 2012b). Low nutrient availability from problem soils and imbalanced fertilization further decrease grain mineral densities in the produced wheat. In fact, the cereal consumption in the world correlates with micronutrient deficiency in human populations (Gregory *et al.*, 2017). As a result, there are more than 2 billion people in the world suffering from Zn and Fe malnutrition (Anon., 2017).

Cereal biofortification, by both genetic and agronomic means, is considered the most feasible solution to widespread micronutrient deficiencies in the countries where cereal grains are consumed in bulk. Therefore, the application of micronutrient fertilizers to low-status soils, both to soil and foliage, is known to increase grain mineral densities in cereals. As compared to soil applications, a significantly more grain Zn and Fe concentration can be achieved in wheat by foliar application of these minerals during flowering and grain development stages (Hussain, *et al.*, 2012b, Velu *et al.*, 2014). This is probably because the foliar-applied nutrients have to travel less and face less physiological resistance than soil-applied nutrients (White & Broadley, 2011). Additionally, as mentioned above, many soil conditions decrease bioavailability to plants and mobility in soil of the native and soil-applied minerals (Olsen & Palmgren, 2014).

Salt stress impairs many physiological and metabolic processes in crop plants (sheldon *et al.*, 2017; Ibrahim *et al.*, 2019). The widespread issues of soil salinity and sodicity in the world are hampering quantity and quality of produced cereal grains. Grain Zn concentration in wheat also negatively correlates with salts in soils (Maqsood *et al.*, 2015). An optimum supply of K regulates water relations in plant tissues and maintains ionic homeostasis by improving nutrient uptake by roots. It is well known that K mitigates the negative effects of drought and salt stress on crop plants by increasing rate of photosynthesis, stomatal conductance and other physiological processes (Wang *et al.*, 2013). However, the possible positive effects of K application on accumulation of Zn and Fe in grains of wheat grown in calcareous salt-affected soils have not been studied. The present field experiment was conducted on a calcareous salt-affected soil to study the accumulation of Zn and Fe in wheat grain with or without soil application of K and foliar applications of Zn and Fe.

Materials and Methods

Soil and weather: This study was conducted in a saline-sodic field at Agricultural Farm, Bahauddin Zakariya University (30.258° N, 71.515° E), Multan (Pakistan). Before the experiment, random samples of soil were collected from the surface layer (0–15 cm depth) of the field for determination of selected soil physical and chemical properties (James & Wells, 1990). The soil of the field was silt loam having sand, silt and clay, respectively, 25, 52 and 23% as determined by Hydrometer method (Gee & Bauder, 1979). Soil pH 8.8 was measured in saturated soil paste and electrical conductivity 4.9 dS m⁻¹ was measured in saturated soil paste extract. Sodium adsorption ratio, calculated based on soluble Na⁺ and Ca²⁺+Mg²⁺, was 15.9 (mmol_c L⁻¹)^{1/2}.

Organic matter, as determined by wet oxidation method of Walkley-Black (Nelson & Sommers, 1996), was 0.8% and acid dissolvable (Loeppert *et al.*, 1982) free calcium carbonate (CaCO_3) was 5.1% of the soil. Ammonium acetate-extractable K in the soil was 109 mg kg^{-1} . The DTPA-extractable concentration of Zn and Fe in the soil were 0.4 and 4.4 mg kg^{-1} , respectively.

Multan is located in arid to semiarid climate with an average annual precipitation of 175 mm that is most received during summer in July-August. The mean daily temperature of the area during the wheat growing season (25 November 2015 to 30 April 2016) was 19°C and it ranged from 5°C to 34°C (Fig. 1). The total precipitation during the growing season (25 November 2015 to 30 April 2016) was 54 mm.

Experimental details: Wheat was grown under two levels of K application i.e. 0 or 35 kg K ha^{-1} and Zn and Fe were applied on foliage at the rate of 0.25% w/v alone and in combination while distilled water was sprayed in control plots. Potassium, Zn and Fe were applied as potassium sulfate (K_2SO_4), zinc sulfate heptahydrate ($\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$) and ferrous sulfate pentahydrate ($\text{FeSO}_4 \cdot 5\text{H}_2\text{O}$). Potassium was applied at sowing and micronutrient solutions (500 L ha^{-1}) were sprayed twice, first at anthesis stage and second nine days after anthesis. Control plots of micronutrient sprays received foliar distilled water. The experiment was laid out following randomized complete plot design with the factorial arrangement. The experimental area was divided into three blocks having subplots net size of $5.0 \text{ m} \times 1.2 \text{ m}$.

Crop husbandry: Pre-soaking irrigation of 10 cm was applied to create suitable conditions for sowing. At

workable moisture level, seedbed was prepared by cultivating and planking. A salt-resistant wheat cultivar, Faisalabad-2008 (Khaliq *et al.*, 2015), was sown on November 15, 2015 in 20 cm apart rows with single row hand drill using seed rate of 160 kg ha^{-1} . At sowing, all plots were uniformly supplied with $37 \text{ kg phosphorus (P) ha}^{-1}$ as di-ammonium phosphate and $60 \text{ kg nitrogen (N) ha}^{-1}$ as urea / di-ammonium phosphate. Potassium was applied according to treatment plan. Weeds were controlled by Bromoxynil + MCPA (ethyl hexyl ester) herbicide. Second split of 30 kg N ha^{-1} was used as urea with first irrigation. The crop was irrigated, as and when required, with tube-well water of good irrigation quality. The mature crop was harvested on April 30, 2016.

Sampling and analysis: At full maturity of the crop, one m^2 from the center of each plot was manually harvested (not including the outer rows on either side of the plot) by cutting the plants from a uniform height of 2.5 cm above the soil surface. Grains were separated by manual threshing and stored in labelled paper bags. Grain and straw samples were dried in a hot-air oven (Pol-Eko Aparatura, SLN 32 STD, Wodzislaw, Poland) at 65°C for 72 h to measure yields. Thousand grains were randomly selected from each experimental unit to measure 1000-grain weight.

A known weight of grain samples was digested in 2:1 solution of nitric and perchloric acids (Jones & Case, 1990). Zinc and Fe concentration in digests were determined on an atomic absorption spectrophotometer (SpectrAA 220, Varian Inc., Palo Alto, California, USA). Potassium in leaf digests and grain samples was measured on a flame photometer (PFP7, Jenway, UK).

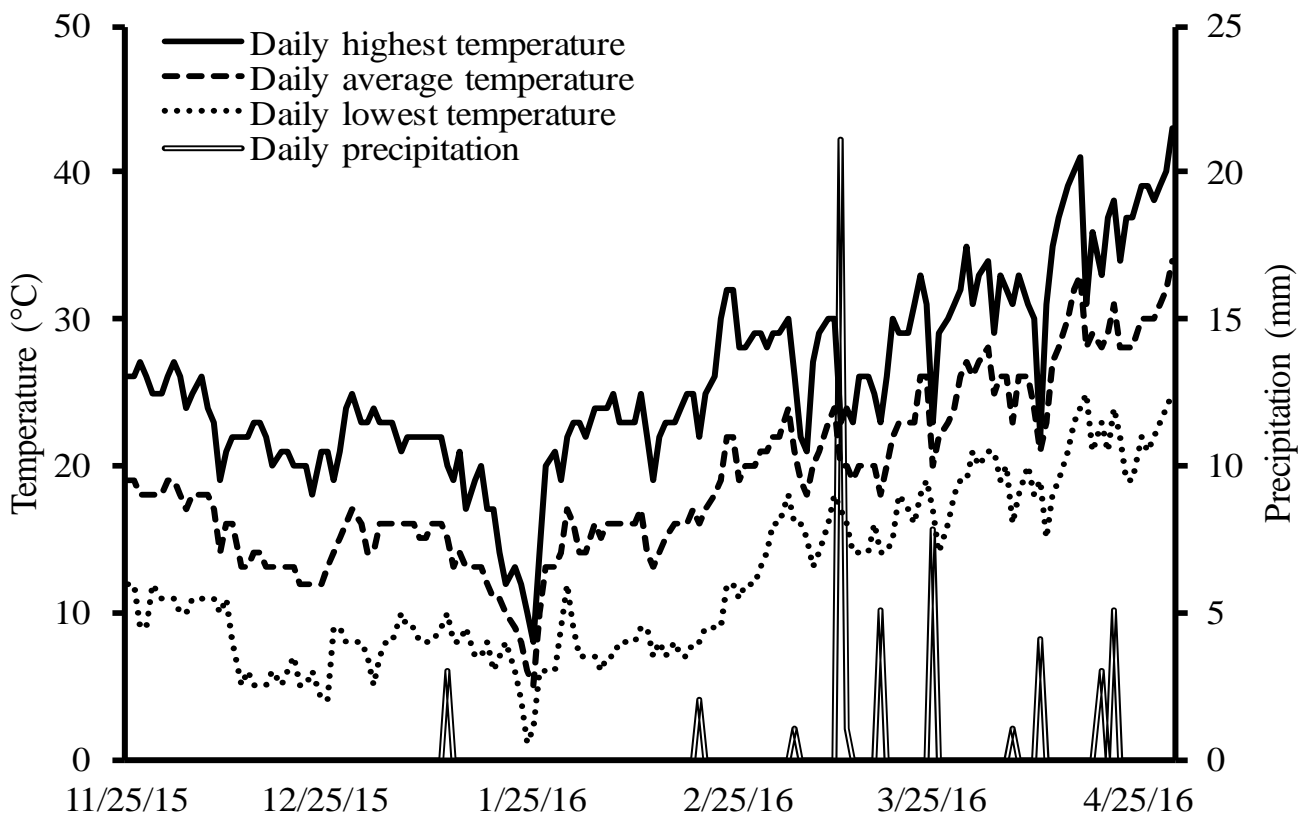


Fig. 1. Weather conditions (temperature and precipitation) of Multan during the experimental period.

Data analysis: Main and interactive effects of soil K application and micronutrient sprays were statistically analyzed with analyses of variance (ANOVA) test for two-factorial Randomize Complete Block Design (Quinn & Keough, 2002). Post-hoc test was Tukey's Honestly Significant Difference (HSD) test at $p \leq 0.05$.

Economic analysis: Economic analyses were performed to assess the economic feasibility of soil K application and foliar micronutrient sprays. Total expenditures incurred for wheat production included soil cultivation and crop sowing, land rent, seed cost, irrigation, fertilizer, herbicide, harvesting and threshing of crop. Net income was assessed from the difference in outputs at harvesting and total inputs. All calculations, in Pakistani rupees, were based on per hectare basis. Moreover, benefit to cost ratio (BCR) was calculated as the ratio of gross income to total expenditure.

Results

Averaged across foliar treatments, soil K application increased grain yield, straw yield, harvest index and 1000-grain weight respectively by 18, 9, 5 and 11% over control K (Table 1). Similarly, K accumulation in leaves and grains was affected only by K application. On average, the increase in concentration and contents of grain K was respectively 26 and 48% with K than without K application.

As compared to control (0 kg K ha⁻¹), concentration and contents of Zn in wheat grains were significantly ($p \leq 0.05$) increased with K application at respective foliar treatments (Fig. 2). Grain Zn concentration ranged from 16 to 33 mg kg⁻¹ in different combinations of soil K application and foliar micronutrient treatments. The increase with than without K application was 23, 38, 47 and 39% respectively at distilled-water control, Fe only, Zn only and Zn + Fe spray. At both K levels, grain Zn

concentration was significantly ($p \leq 0.05$) increased (41–42% at control K and 61–70% at applied K) with Zn spray than the treatments with no Zn spray. Similarly, grain Zn contents were significantly ($p \leq 0.05$) increased, 39–40% at control K and 62–69% at applied K, with Zn spray than the treatments with no Zn spray. Compared to other treatments, grain concentration and contents of Zn were significantly ($p \leq 0.05$) more at combined soil K application and foliar Zn treatments.

Only at control K level, sole Zn spray decreased grain Fe concentration by 6% and grain Fe contents by 7% than distilled-water control (Fig. 3). Grain Fe concentration was significantly ($p \leq 0.05$) increased with K application than control K only at sole Zn spray. Foliar treatments having Fe significantly ($p \leq 0.05$) increased grain concentration and contents of Fe over the respective foliar treatments with no Fe at two soil K levels. This increase at control and applied K rates was respectively 13 to 15% and 11 to 12% in grain Fe concentration, and 10 to 13% and about 11% in grain Fe contents. At combined soil K application and foliar Fe treatments, grain Fe concentration ranged from 38 to 39 mg kg⁻¹ and did not differ significantly ($p \leq 0.05$) among the treatments. Grain Fe contents were increased significantly ($p \leq 0.05$) with K application than control K in respective treatments of micronutrient sprays.

Maximum net return of PKRs. 84,414 was achieved with soil K application at control level of foliar micronutrient sprays (Table 2). It was 33% more than no K application at control level of foliar micronutrient sprays. At both rates of soil K application, foliar application of Zn and Fe (alone or combined) decreased net returns compared at control level of foliar spray. However, benefit-to-cost ratio (BCR) remained 1.7 at four levels of micronutrient sprays albeit higher than respective foliar treatment at control level of K application.

Table 1. Effect of soil potassium (K) application on yield and K accumulation in wheat grown in a calcareous saline-sodic soil.

Potassium rate (kg K ha ⁻¹)	Foliar micronutrient sprays	Grain yield (Mg ha ⁻¹)	Straw yield (Mg ha ⁻¹)	Harvest index (%)	1000-grain weight (g)	Grain K concentration (g kg ⁻¹)	Grain K content (kg ha ⁻¹)
0	Control	4.2 ± 0.1 b	8.6 ± 0.2 ab	33 ± 0 a	35.4 ± 1.3 bcd	4.3 ± 0.2 b	18 ± 1 b
	Zn	4.1 ± 0.1 b	8.2 ± 0.3 b	33 ± 1 a	35.1 ± 1.0 cd	4.3 ± 0.2 b	18 ± 1 b
	Fe	4.1 ± 0.1 b	8.5 ± 0.3 ab	33 ± 1 a	35.4 ± 1.2 cd	4.2 ± 0.2 b	17 ± 1 b
	Zn + Fe	4.1 ± 0.1 b	8.4 ± 0.6 ab	33 ± 2 a	34.7 ± 0.6 d	4.2 ± 0.3 b	17 ± 1 b
35	Control	4.8 ± 0.1 a	9.4 ± 0.2 a	34 ± 0 a	39.4 ± 1.2 a	5.3 ± 0.3 a	26 ± 2 a
	Zn	4.8 ± 0.1 a	8.9 ± 0.3 ab	35 ± 2 a	38.8 ± 2.5 abc	5.4 ± 0.2 a	26 ± 2 a
	Fe	4.8 ± 0.2 a	9.3 ± 0.1 a	34 ± 0 a	39.2 ± 2.4 ab	5.2 ± 0.2 a	25 ± 1 a
	Zn + Fe	4.9 ± 0.1 a	9.0 ± 0.4 ab	35 ± 1 a	38.9 ± 0.5 abc	5.6 ± 0.1 a	27 ± 0 a

Values are means ± standard deviations; For each parameter, means sharing different letters are significantly ($p \leq 0.05$) different based Tukey's HSD test.

Table 2. Economic analysis of potassium application and micronutrient sprays on wheat crop.

Potassium rate (kg K ha ⁻¹)	Foliar micronutrient sprays	Total expenses (cost) (PKRs.)	Gross return (benefit) (PKRs.)	Net return (PKRs.)	Benefit to cost ratio (BCR)
0	Control	114,898	178,333	63,435	1.6
	Zn	115,783	173,267	57,484	1.5
	Fe	115,783	176,667	60,884	1.5
	Zn + Fe	116,418	174,083	57,665	1.5
35	Control	119,869	204,283	84,414	1.7
	Zn	120,754	200,317	79,563	1.7
	Fe	120,754	203,850	83,096	1.7
	Zn + Fe	121,389	203,150	81,761	1.7

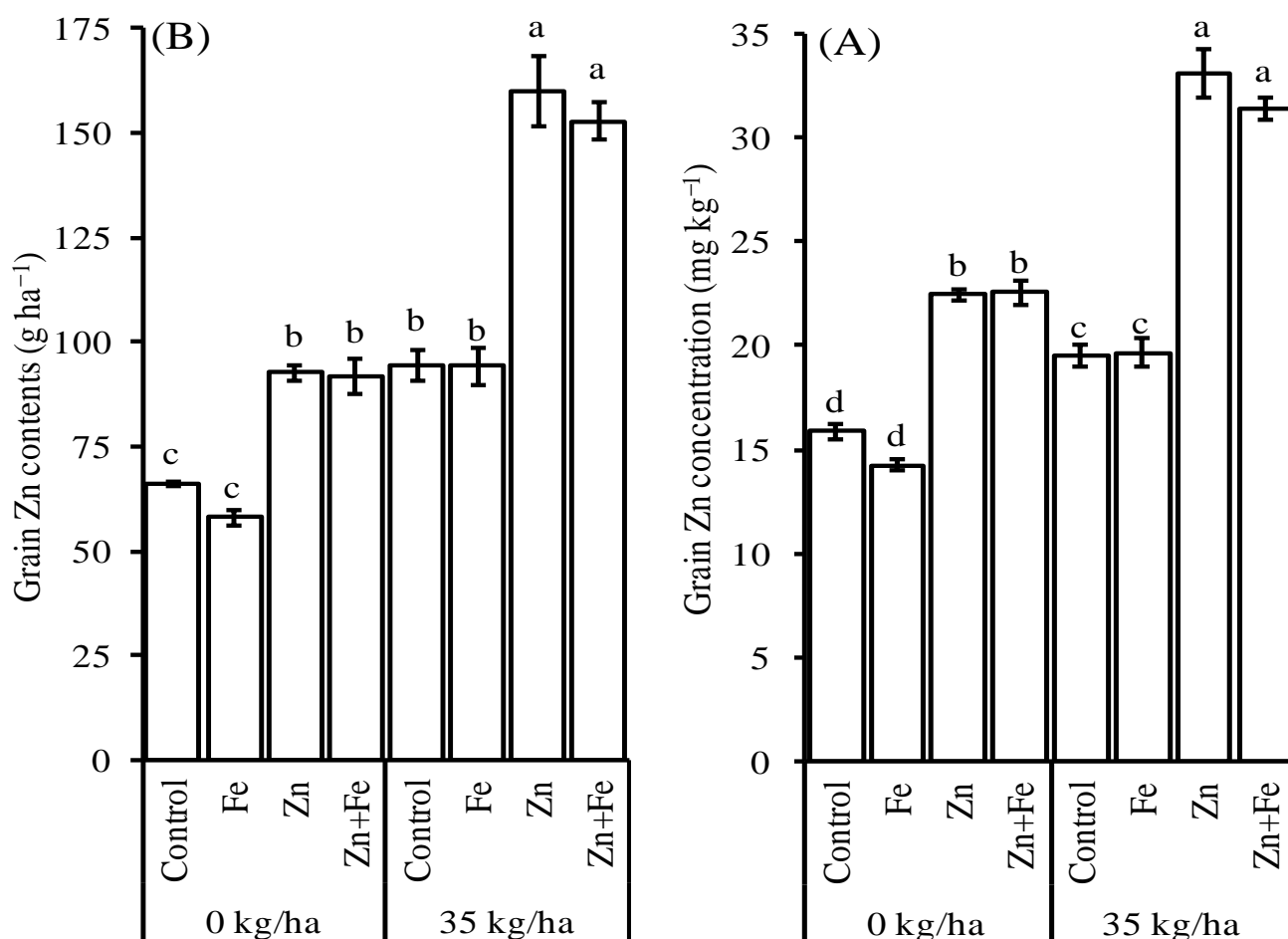


Fig. 2. Effect of soil potassium (K) application and foliar micronutrient sprays on concentration (A) and contents (B) of zinc (Zn) in grains of wheat grown in a calcareous saline-sodic soil. Different letters on the bars indicate significant ($p \leq 0.05$) difference based on Tukey's HSD test. Error bars are of standard deviations.

Discussion

Sodium and other cations present in the soil solution of saline-sodic soils compete for root uptake and decrease K uptake by roots (Benito *et al.*, 2014). The toxic levels of Na in plants further suppress normal physiological functioning of K in plants. To mitigate Na stress and to improve K nutrition, therefore, routine soil K applications are recommended for low K saline-sodic soils. In present experiment too, K application significantly ($p \leq 0.05$) increased grain and straw yield of wheat grown in a medium K saline-sodic soil (Table 1). In plants, K mitigates the negative effects of stressed environments (Anschütz *et al.*, 2014, Zhang *et al.*, 2016). Therefore, the increase in yield was correlated with improved K accumulation in plant tissues with soil K application (Table 1).

Mitigation of salinity stress by K application was also evident with increased net return and BCR (Table 2) due to increase in grain and straw yield (Table 1). However, foliar application of Zn and Fe decreased net return at both K rates and BCR at control level of K (Table 2). This seems to relate with a non-significant decrease in grain and straw yield with foliar micronutrient sprays (Table 1). In fact, the strengths of applied foliar sprays (0.25% w/v Zn and 0.25% w/v Fe) were more than generally recommended for wheat biofortification with micronutrients (Shahzad *et al.*, 2014).

At control K rate, grain Zn concentration was increased from 16 mg kg⁻¹ at distilled-water control to 22 mg kg⁻¹ at

sole foliar Zn application (Fig. 2A). Combined soil K application and foliar Zn spray increased grain Zn concentration to 33 mg kg⁻¹. Increase in grain Zn concentration with foliar Zn sprays is well documented (Hussain *et al.*, 2012a, Zhao *et al.*, 2014, Ram *et al.*, 2016). The positive influence of soil K application in increasing grain Zn concentration is relatively a new topic (Naeem *et al.*, 2018). The increase in grain Zn concentration with soil K application to medium K saline-sodic soil (Fig. 2A) might relate with improved water relations in the presence of optimum K. Proper K availability, both in normal and salt-affected soils, improve plant nutrition by positively interacting with several minerals (Hafsi *et al.*, 2014). These interactions of K are more important under saline-sodic soils.

In wheat plants supplied with excessive P, soil-applied K enhanced grain Zn accumulation by influencing post-anthesis Zn uptake by roots and remobilization of pre-anthesis Zn reservoirs into grains (Naeem *et al.*, 2018). The increase in concentration and contents of Zn in grains with soil-applied K (Fig. 2) suggests a similar increase in root uptake, stem translocation and shoot remobilization of Zn in wheat grown on medium K saline-sodic soil. A significant ($p \leq 0.05$) increase in grain Zn concentration by soil K application in treatments receiving foliar Zn suggests a positive role of K in uptake of foliage applied Zn and its remobilization towards grains. This seems evident as K regulates stomatal conductance and water relations in plants (Andres *et al.*, 2014, Martineau *et al.*, 2017).

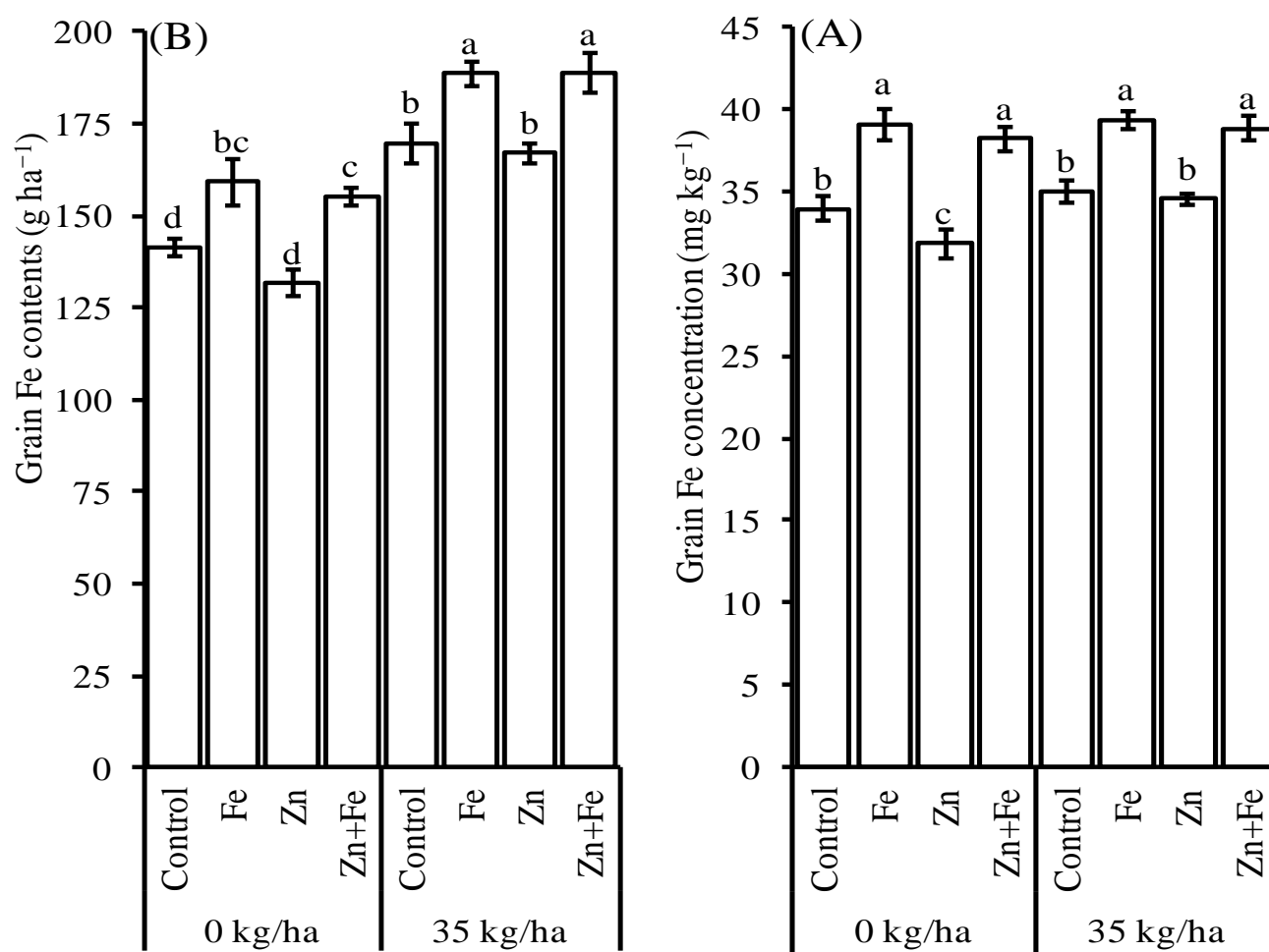


Fig. 3. Effect of soil potassium (K) application and foliar micronutrient sprays on concentration (A) and contents (B) of iron (Fe) in grains of wheat grown in a calcareous saline-sodic soil. Different letters on the bars indicate significant ($p \leq 0.05$) difference based on Tukey's HSD test. Error bars are of standard deviations.

As expected, grain concentration and contents of Fe were increased with foliar Fe application at both soil K rates (Fig. 3). However, there was relatively a lesser variation in grain Fe accumulation with foliar Fe sprays than the observed variation in grain Zn accumulation with foliar Zn sprays (Figs. 2 and 3). The extent of an increase in grain mineral accumulation with foliar spray depends on the nutrient supply from the soil (Hussain *et al.*, 2012b, Kutman *et al.*, 2012). Therefore, the limited plant-available Zn in the soil (0.4 mg DTPA-extractable Zn kg⁻¹) and the optimum plant-available Fe (4.4 mg DTPA-extractable Fe kg⁻¹) might explain this varied behavior of two micronutrients in grain accumulation in wheat (Figs. 2 and 3). Differential leaf absorption and remobilization of nutrients are other factors determining the accumulation of foliar-applied nutrients in grains.

Excepting the plots sprayed with Zn only, soil K application only non-significantly ($p \leq 0.05$) influenced grain Fe concentration (Fig. 3). However, grain Zn concentration was significantly ($p \leq 0.05$) influenced at all foliar treatments (Fig. 2). It seems that soil-applied K is more important in increasing the grain density of the micronutrient posing deficiency stress to plants. In fact, K mitigates a number of biotic and abiotic stresses in plants (Wang *et al.*, 2013). On the other hand, the increase in grain Fe contents with K application than control K (Fig.

3) might simply because of increase in grain yield (Table 1) by mitigation of negative effects of salt stress.

Only at control rate of K (0 kg K ha⁻¹), foliar Zn significantly ($p \leq 0.05$) decreased grain Fe concentration (Fig. 3). Non-significant to significant ($p \leq 0.05$) decrease in grain Fe concentration is reported with soil and foliar Zn application (Li-cheng *et al.*, 2010, Zhang *et al.*, 2010, Hussain *et al.*, 2012b). At two K rates, however, grain Zn and Fe concentration at combined Zn + Fe sprays were statistically at par with respective sole Zn or Fe sprays (Figs. 2 and 3). Hence, combined Zn + Fe sprays are more suitable for increasing mineral densities in cereal grains.

Conclusions

Foliar Zn sprays were relatively more effective in increasing grain Zn concentration than foliar Fe sprays for increasing grain Fe concentration. Foliar Zn decreased grain Fe concentration only in plots not supplied with K. Optimum supply of K in saline-sodic soil increased not only the yield and economic returns but also the grain Zn density in wheat. Conclusively, soil K application may increase grain accumulation of native soil- and foliar-applied Zn in wheat grown on calcareous saline-sodic soils.

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