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The Effect of Zinc (Zn) Fertilization on Alleviating Cd Accumulation in Durum Wheat Grain

Faruk Özkutlu[1] and Şevket Metin Kara[2]

1. Department of Soil Science and Plant Nutrition, Faculty of Agriculture, Ordu University, Ordu 52200, Turkey

2. Department of Field Crops, Faculty of Agriculture, Ordu University, Ordu 52200, Turkey

Abstract: Cadmium (Cd) contamination of agricultural soils may pose severe risks and hazards for humans through food chain, as crop plants accumulate Cd in their edible tissues. Cd translocation from soil to plant is largely dependent on soil and plant type. Cd accumulation occurs much more in crop plants grown in soils with severe zinc (Zn) deficiency and durum wheat tends to accumulate more Cd in grain than the other cereals. The objective of this study was to evaluate the alleviating effect of Zn fertilization on Cd accumulation in durum wheat grain. A pot experiment including foliar Zn application of 0.3% w/v $ZnSO_4$ and soil Cd applications of 0, 0.2 and 1.0 mg/kg was carried out in a completely randomized design using a Zn-deficient soil. Grain Cd concentration of plants receiving 0 mg/kg Cd was 31 µg/kg, whereas with 0.2 mg/kg and 1.0 mg/kg Cd applications it increased to the levels of 215 µg/kg and 1,489 µg/kg, respectively. Along with 0.3% $ZnSO_4$ leaf applications, grain Cd concentrations decreased to 171 µg/kg and 754 µg/kg, by a reduction of 20.5% and 49.3%, respectively. In conclusion, it was determined that leaf applied Zn fertilizer might alleviate Cd accumulation in durum wheat grain in Zn deficiency conditions.

Key words: Cadmium, *Triticum turgidum* L. *durum*, Zn deficiency, Zn-deficient soil.

1. Introduction

Cadmium (Cd) is a heavy metal with no known essential biological functions in higher plants, animals and humans. Although Cd normally occurs in low concentrations in agricultural soils, it is highly toxic even at very low concentrations [1]. The limit of total Cd concentration in cultivated areas has been reported to be 3 mg/kg soil, while this limit was around 0.1 mg/kg soil for uncultivated areas [2]. Accumulation of Cd in agricultural soils has gained a great concern due to agricultural productivity, food safety, human health, environmental pollution and biodiversity [3]. In consequence of this, Cd transfer to human through food chain has also become a widespread challenge throughout the world. There is a need to reduce the accumulation of toxic metals such as Cd in edible plant parts. It has been stated that Cd accumulation in human body may lead to some major health problems in liver and lung [4, 5]. Since Cd primarily enters plant tissues from the soil and then transports to human body by virtue of food chain, an understanding of the factors affecting Cd transport from soil into plant is vitally important to predict and reduce Cd accumulation in edible plant tissues. Cd translocation from soil to plant is largely dependent on soil and plant type, and the amount of Cd accumulated by different plant species varies widely [3, 6].

Among major food sources, cereals are considered playing the most important role in human nutrition, with wheats (bread and durum wheat, *Triticum aestivum* and *T. durum* L.) in general are of particular concern. Durum wheat accumulates more Cd than the other cereals commonly grown and Cd accumulation increases in the order of rye < barley < oats < bread wheat < durum wheat [6]. Nutrient status of agricultural soils may show an important effect on Cd accumulation in cereal grains, in particular certain micronutrients such as zinc (Zn) is of a great concern and needs much more attention than the other

Corresponding author: Faruk Özkutlu, professor, research field: plant nutrition.

elements.

Zn is an essential trace element for plants, humans and animals, but may be toxic when present at high levels [7]. Cd and Zn have similar properties in periodic table. So, their chemical similarity can lead to interaction between Cd and Zn during plant uptake, transport from roots to the aerial parts and accumulation in edible parts [8, 9]. It is generally accepted that Zn status in soils and plants play an important role in Cd accumulation in crop plants [10, 11]. Several studies exposed that Zn applications decreased Cd uptake and accumulation in plant tissues [10, 12-14]. Zn deficiency appears to be the most widespread micronutrient problem worldwide. It is estimated that nearly half of the soils on which cereals are grown has levels of available Zn low enough to cause Zn deficiency [7]. Sensitive crops, such as durum wheat, grown on Zn-deficient, alkaline and calcareous soils show severe Zn deficiency [15]. Both soil and foliar applications of Zn are used to ameliorate Zn plant deficiency as well as increase Zn and decrease Cd content in edible plant parts [10, 11, 14]. Generally, durum wheat genotypes accumulate higher Cd in grain than bread wheat genotypes in Zn deficiency conditions [16]. There is much evidence that Cd may be accumulated in cereal grains grown on Zn deficient soils [17-19]. Interactions of Cd-Zn and their accumulation in plant have been reported in solution culture or in pot experiment [20-22]. The objective of this study was to evaluate the alleviating effect of Zn fertilization on Cd accumulation in durum wheat grain.

2. Materials and Methods

Seeds of durum wheat (*T. turgidum* L. *durum*) variety Balcalı-2000 were sown in plastic pots (3.5 L) containing 2.70 kg dry soil and the wheat plants were grown under greenhouse conditions at Agricultural Faculty of the University of Cukurova Adana/Turkey. The soil used was Zn-deficient containing 0.10 mg DTPA-extractable Zn per kg soil and obtained from the Central Anatolia where Zn deficiency is a severe constraint to wheat production [14, 18]. The soil used for the pot experiment was a clay loam in texture with organic matter content of 1.04%. DTPA-extractable Fe, Mn and Cu concentrations of the soil were 3.51, 5.81 and 0.87, respectively. Total Zn and Cd concentrations of the soil were 55 mg Zn/kg and 0.24 mg Cd/kg, measured after wet ashing as described by Jackson [23], while DTPA-extractable [24] concentrations of Zn and Cd were 0.10 mg Zn/kg and > 0.005 mg Cd/kg, respectively. The analysis of all chemical and physical properties of the soils was carried out using standard methods given by Jackson [23].

A pot experiment including foliar Zn application of 0.3% w/v $ZnSO_4$ and soil Cd applications of 0, 0.2 and 1.0 mg/kg was carried out in a completely randomized design with three replications. In the experiment, 12 seeds were sown in each pot. After one week of emergence, the number of seedlings in each pot was reduced to four. Before potting, the soils were treated with three levels of Cd (0, 0.2 and 1.0 mg/kg soil) in the forms of $CdSO_4$. Since the soil used in the study was Zn-deficient, soil fertilization of 1.0 mg/kg $ZnSO_4$ was carried out in order to prevent the diminishing effect of Zn deficiency on grain production. The basal fertilizer of 200 mg N/kg soil as $Ca(NO_3)_2$, 200 mg P/kg soil as KH_2PO_4 and 2.5 mg Fe/kg soil as Fe-EDTA were applied to each pot. All nutrients were mixed homogenously with the soil before sowing. The wheat plants were treated with 0% and 0.3% (w/v) $ZnSO_4$ from the leaf, four times starting at stem elongation stage.

The plants were grown under greenhouse conditions until the grains were harvested and concentrations of Zn and Cd were determined along with grain dry weight. The grain samples were ground and digested in 2 mL 30% H_2O_2 and 5 mL 65% HNO_3 [14] in sealed vessels of a microwave apparatus. Zn and Cd were measured by inductively coupled plasma-atomic emission spectrometry (ICP-AES, Jobin Yvon, Paris), and all sampling and measurements were carried out by using three independent replications.

3. Results and Discussion

Increasing soil Cd application showed a negative effect on grain dry weight. The grain dry weight in the plants not treated with Cd was found as 1.78 g/plant, but it decreased significantly to 1.19 g/plant, with the soil application of 1.0 mg/kg Cd (Table 1). On the other hand, application of foliar 0.3% (w/v) $ZnSO_4$ cleared away the decreasing effect of soil applied Cd and significantly increased grain dry weight of durum wheat in all soil applications of Cd. With the application of foliar 0.3% (w/v) $ZnSO_4$, the grain dry weight increased to 3.94, 3.86 and 3.42 g/plant in 0, 0.2 and 1.0 mg/kg Cd soil applications, respectively.

As expected, the durum wheat grain showed higher Cd accumulation with increasing Cd doses, irrespective of Zn application (Table 2). Grain Cd concentration of the plants receiving 0 mg/kg Cd was 31 µg/kg, whereas with 0.2 mg/kg and 1.0 mg/kg Cd applications it reached to the levels of 215 µg/kg and 1,489 µg/kg, respectively. Foliar application of 0.3% (w/v) $ZnSO_4$, however, significantly decreased grain Cd concentration. In comparison to the control without Zn fertilization, the foliar Zn treatment decreased grain Cd content to 171 µg/kg and 754 µg/kg in Cd soil applications of 0.2 mg/kg and 1.0 mg/kg, respectively.

In the case of grain Zn concentration, a slight increase occurred with the application of 0.2 mg/kg Cd as compared to the control, but with no additional increase of 1.0 mg/kg Cd (Table 3). Foliar application of 0.3% (w/v) $ZnSO_4$ affected positively grain Zn concentration, and led to an increase of grain Zn to 41, 45 and 57 µg/kg, compared to the control without Zn. With the foliar application of 0.3% (w/v) $ZnSO_4$, as compared to the control without Zn, respective increases of 95%, 73% and 119% in grain Zn concentration were recorded.

Consistent with the previous studies, the findings of the present study indicated that increasing Cd addition to the soil resulted in a decrease in grain dry weight,

Table 1 Grain dry weight (g/plant, mean ± std) of durum wheat subjected to soil applied Cd and leaf applied Zn.

	Grain dry weight (g/plant)	
	Leaf application control (deionized water)	Leaf application 0.3% (w/v) $ZnSO_4$
0	1.78 ± 0.1	3.94 ± 0.3
0.2	1.70 ± 0.1	3.86 ± 0.4
1.0	1.19 ± 0.1	3.42 ± 0.5

Std: standard deviation.

Table 2 Grain Cd concentration (µg/kg, mean ± std) of durum wheat subjected to soil applied Cd and leaf applied Zn.

	Grain Cd concentration (µg/kg)	
	Leaf application control (deionized water)	Leaf application 0.3% (w/v) $ZnSO_4$
0	31 ± 2.9	24 ± 1.6
0.2	215 ± 25	171 ± 20
1.0	1,489 ± 171	754 ± 56

Std: standard deviation.

Table 3 Grain Zn concentration (mg/kg, mean ± std) of durum wheat subjected to soil applied Cd and leaf applied Zn.

	Grain Zn concentration (mg/kg)	
	Leaf application control (deionized water)	Leaf application 0.3% (w/v) $ZnSO_4$
0	21 ± 2.1	41 ± 2.5
0.2	26 ± 2.3	45 ± 5.0
1.0	26 ± 1.6	57 ± 4.6

Std: standard deviation.

but an increase in the concentration of Cd in grain of durum wheat grown in Zn-deficient soil [3, 9, 21]. The soil application of 1.0 mg/kg Cd reduced grain dry weight by 33%, whereas resulted in a 48-fold increase in the concentration of grain Cd, compared to the control without Cd and Zn. The negative effect of increasing Cd addition to soil was much more pronounced in terms of grain Cd concentration. As foreseen by previous investigators, foliar 0.3% (w/v) $ZnSO_4$ application healed the negative effect of Cd on dry weight and Cd concentration of durum grain [10, 14, 17, 25, 26]. Grain dry weight increased 2.21, 2.27 and 2.87-fold with leaf Zn treatment in all Cd soil applications, while grain Cd concentration decreased by 22.5%, 20.5% and 49.3%, respectively. The results also showed that Cd/Zn ratio in durum wheat grain was 1.47 in the control pots receiving no Cd and Zn and increased to 8.27 and 57.27 with the application of 0.2 mg/kg and 1.0 mg/kg Cd, respectively. Foliar Zn treatment resulted in a respective decrease at all levels of soil Cd as 0.58, 3.8 and 13.23.

The relatively high increases in dry weight and decreases in Cd concentration of durum wheat grain indicate the importance of Zn and its deficiency. A few studies have revealed that, durum wheat is known to be very sensitive to Zn deficiency [27, 28], possibly due to a low release rate of Zn-mobilizing phytosiderophores from roots [29]. Compatible with this work, several studies reported that Zn application decreased Cd concentration in wheat [30-33]. In a compatible study in barley, it has also been indicated that increasing rate of Zn application from 0 kg/ha to 3 kg/ha decreased flag leaves Cd from 12.9 mg/kg to 5.5 mg/kg and grain Cd from 12.0 mg/kg to 4.0 mg/kg [34]. As compatible with the results of the present study, it has been suggested that foliar application of 0.3% Zn in the form of $ZnSO_4$ has effectively ameliorated the adverse effects of Cd and decreased grain Cd concentration in wheat grown in Cd contaminated soil [35]. The similar results obtained in several studies carried out on Zn fertilization to abate Zn deficiency in crop plants on soil with low Zn availability and for the purpose of agronomic Zn biofortification of cereal grains [36, 37] and when grain Zn concentration increases, Cd concentration decreases in grain. On the other hand, the Joint FAO/WHO Expert Committee on Food Additives (JECFA) has established a provisional tolerable level for Cd as 25 µg Cd intake per kg body weight per month [38]. Furthermore, interactions among trace elements can also influence the extent to which metals and micronutrients are taken up and transported to plant tissues. Previous studies have reported both competitive and non-competitive interactions may occur among toxic heavy metals and micronutrients such as Zn, Fe and Mn [11, 39]. Soil deficiency in Mn and Fe can affect the availability of Cd to plant, as in the case of Zn deficiency. It has been stated that there is a competition between Zn, Fe and Cu for uptake and translocation in plant and at low levels of these micronutrients the adverse effects of Cd was much more apparent [14].

4. Conclusions

The interaction effect regarding Zn and Cd uptake was determined as the result of this study. Cd application increased the Cd/Zn ratio whereas Zn application reduced the Cd/Zn ratio of durum wheat grain. The Cd concentration in the grain of durum wheat was largely dependent on the Zn level. Significant inhibitory effect of Zn on Cd concentration in the grains occurred at all Cd levels and Cd concentration in grain decreased with increasing foliar Zn application. The present study demonstrated that the foliar application of 0.3% (w/v) $ZnSO_4$ could effectively increase dry weight and decrease Cd concentration of grain of durum wheat plants grown on Cd contaminated soil.

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