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The Effect of Foliar Application of Different Amino Acids (L-Histidine, Methionine) on Cadmium and Zinc Uptake of Wheat

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A R T I C L E I N F O

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A B S T R A C T

In this study, the effect of foliar L-Histidine and Methinonin amino acid applications on grain cadmium (Cd) and zinc (Zn) uptake on durum wheat grown in soil contaminated with Cd was investigated. The research was carried out according to the randomized blocks design as a pot experiment in greenhouse conditions. In the experiment, Cd doses were applied as control (0 mg Cd kg^{-1}) and (3 mg Cd kg^{-1}). L-Histidine and Methionine amino acids were applied 7 times from the leaf after the start of flag leaf formation in wheat at 0.5 mM doses and harvested after the grain maturity was completed. According to the results obtained, the wheat grain dry matter yield in 3 mg Cd kg^{-1} polluted soil was 44.5 mg grain^{-1} in the control, while the dry matter weight was increased with the application of L-Histidine and Methionine from the leaves being 48 and 50 mg grain^{-1}, respectively. It was determined that there were differences in grain Cd and Zn concentrations with the application of amino acids from the leaves at the dose of 3 mg kg^{-1} cadmium. When the grain Cd and Zn intakes were compared with the control, it was found that the grain Cd concentration decreased, and the Zn concentration increased as a result of the application of amino acids. The high amount of Cd in wheat can make it to the human body through the food chain and is known to cause serious health problems after a certain amount of accumulation. As a result of this study, it is seen that L-Histidine and Methionine are effective in reducing Cd in the grain, and these amino acids probably form a complex with Cd, resulting in less transport.

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Introduction

Zinc (Zn) deficiency is a global nutritional problem and is called “hidden hunger” (Stein, 2010, Korkmaz et al., 2021). Studies have reported that one sixth of the world's population is faced with insufficient Zn intake (Wessellsand and Brown, 2012). Zinc deficiency is usually caused by eating foods with low zinc content. Corn and wheat are the most important sources of zinc (Welch and Graham., 2004;). FAO (2016) has declared that wheat is the most basic food source for humans and its worldwide production is increasingly >740 Mt. The deficiency of Zn in soils and factors such as high pH, high calcium carbonate, clay, iron and aluminum oxide content, as well as low moisture content limit the usefulness of Zn causing low concentrations of Zn to be transported to wheat grains (Alloway, 2008; Cakmak, 2008). It has been reported that more than 30% of the world's agricultural soils are Zn deficient and therefore Zn concentration will be low in plants grown in these areas (Alloway., 2008). As well as the importance of Zn deficiency in agricultural soils, its contamination with other heavy metals is a serious threat to crop production worldwide (Adrees., 2015a; Rizwan et all, 2016a; Korkmaz et al., 2017; Korkmaz et al.,2018). Cd, which is among the heavy metals and an important environmental pollutant, is not an essential element in the nutrition of plants, animals, and humans. Although in very low amounts in agricultural soils, Cd can enter the soil from various sources. Some of the sources that lead to the introduction of Cd into the soil result from anthropogenic sources through the atmosphere, through the application of sewage sludge to agricultural lands, and through the use of fertilizers. The permissible Cd concentration in agricultural soils is 3 mg kg^{-1}, and the Cd level in soils is generally around 0.1 mg kg^{-1} (Alloway, 1995). In addition to the low amount of Cd in the soil, when Cd enters from various sources, Cd in wheat grown in these soils can be easily taken up by the roots and accumulated in the grain. Common health problems in humans with excess cadmium are lung, liver and kidney disorders, visual impairment, anemia, and high blood pressure (Järup et al., 1998; Aikens and Rouse, 2005). WHO and FAO (1993) declared that the maximum allowable cadmium concentration in cereal-based foods should be at the level of 100 µg kg^{-1}.

According to the World Health Organization, the amount of Cd that an adult person can take daily is between 60-70 µg. Although cadmium is not an essential nutrient for plant growth, Zn is an essential element for plants, animals, and humans. In case of Cd accumulation in the environment under zinc deficiency conditions, more Cd can enter the plant (Hart, et al., 2005; Özkutlu et al., 2009; Korkmaz et al., 2010; Brennan and Bolland, 2014; Özkutlu and Kara, 2018; Özkutlu and Erdem, 2018; Korkmaz and Turkis, 2021). While Zn in high concentration has a positive effect on human health in wheat grain, Cd is harmful to human health. For this reason, in recent years, researches on obtaining new varieties that accumulate higher amounts of Zn in the grain and reducing Cd in the grain have been increasing rapidly since it has a positive effect on human health. In this study, the effects of L-Histidine and Methionine amino acid applications on Cd and Zn uptake in wheat grain were investigated.

Material and Method

Material

The research was carried out as a pot experiment in greenhouse conditions. Harran-95, a durum wheat variety, was used in the experiment. Some physical and chemical properties of the soil used in the experiment are as follows; pH 8.06, soil lime content 12%, organic matter 1.08%, available phosphorus (P) concentration 4.13 mg kg^{-1}, potassium (K) concentration 244 mg kg^{-1}, extractable Zn concentration in DTPA 0.11 mg kg^{-1}, Cd concentration 0.005 mg kg^{-1}.

Method

In the experiment, 2.65 kg of soil sifted through a 4 mm sieve was placed in each pot. Before planting, Cd doses and basic fertilizers were mixed into the soil. As the application doses, two different doses of Cd (0 mg Cd kg^{-1} and 3 mg Cd kg^{-1}) were applied. In addition to the cadmium application, as the basic fertilization, 300 mg of nitrogen (N) in the form of $(CaNO_3)_2.4H_2O$, 100 mg of P in the form of KH_2PO_4, 2.5 mg of iron (Fe) from the form of Fe-EDTA and 1 mg of Zn from the form of $ZnSO_4$ were applied per 1 kg of soil. 0.5 mM L-Histidine and Methionine amino acids from the leaf were applied 7 times at 5-day intervals in wheat, 1 week after the flag leaf formation. The plants were harvested at the grain maturity period (Day 96) and the grain weights were determined after the wheat grains were separated from the ear. Grain samples were burned in a H_2O_2 -HNO_3 acid mixture in a microwave oven according to the wet burning method, and Cd and Zn concentrations in the obtained filtrates were measured in an Inductively Coupled Plasma-Optical Emission Spectrophotometer (ICPOES -Varian Vista).

Results and Discussion

Dry Matter Weight

Studies so far have proven that amino acids can directly or indirectly affect physiological activities in plant growth and yield (Mohamed, 2006). Apart from these effects, the effects of amino acids on metals are also of great interest. The variety used in determining the grain yield of wheat and the yield of wheat is affected by many factors in which nitrogenous fertilizers and amino acids play important roles. In this study, it was determined that application of L-Histidine and methionine amino acids from the leaves caused differences in dry matter weight. While the dry matter weight was 42.7 mg $grain^{-1}$ in the control pots where Cd was not applied, the weight was 48.7 mg $grain^{-1}$ with the foliar application of L-Histidine leading to an increase of 14%. A similar increase was obtained with the application of Methionine from the leaves, as 11%. In wheat grown under cadmium stress, improvement in grain yield was achieved by foliar application of amino acids. For example, it was determined that the grain yield of wheat grown under 3 mg kg^{-1} soil application of Cd was 41.1 mg $grain^{-1}$ in the control without amino acid application, while it increased to 48 and 50 mg $grain^{-1}$ level as a result of foliar L-Histidine and Methionine applications (Table 1). It is thought that L-Histidine and Methionine amino acids probably form complexes with Cd and affect the Cd uptake of plants directly or indirectly on plant development and growth regulation. This effect is due to the availability of amino acids in the rhizosphere solution, as plants can take up nitrogen in the form of amino acids, both through stimulatory effects on plant growth and indirectly through reduced Cd uptake (Gramlich et al., 2013; Kaewchangwat et al., 2017). Another study supporting our research results was carried out by Mousavi et al. (2021), in which it was reported that Cd-Methionine complex was formed with the application of Methionine amino acid, and this had a positive effect on plant growth. Another study by Azimi et al. (2013) determined that the application of foliar amino acids increased wheat yield and quality. Other studies supporting our results were also reported by Sonia et al. (2013) and Salwa and Osama (2014). The importance of amino acids derives from their widespread use for the biosynthesis of a wide variety of non-protein nitrogenous materials such as pigments, vitamins, coenzymes, purine, and pyrimidine bases. According to many studies in the literature, it has been proven that amino acids can directly or indirectly affect physiological activities in plant growth and yield. In this study, we determined that the application of L-Histidine and Methionine from the leaves directly affects the grain yield and quality.

Table 1. Dry Matter Weight of Foliar L-Histidine and Methionine Amino Acid Applications

Applications		Dry Matter Weight (mg $grain^{-1}$)
Soil	Leaf	
0 mg Cd kg^{-1}	Control	42.7 ± 5.4
0 mg Cd kg^{-1}	0.5 mM L-Histidine	48.7 ± 2.4
0 mg Cd kg^{-1}	0.5 mM Methionin	47.2 ± 2.4
3 mg Cd kg^{-1}	Control	41.1 ± 4.8
3 mg Cd kg^{-1}	0.5 mM L-Histidine	48.0 ± 3.6
3 mg Cd kg^{-1}	0.5 mM Methionin	50.0 ± 4.2

Table 2. Grain Cd and Zn Concentration of Foliar L-Histidine and Methionine Amino Acid Applications

Applications		Cd	Zn
Soil	Leaf	(µg kg^{-1})	(mg kg^{-1})
0 mg Cd kg^{-1}	Control	62 ± 9	10.4 ± 1.4
0 mg Cd kg^{-1}	0.5 mM L-Histidine	57 ± 8	13.3 ± 2.0
0 mg Cd kg^{-1}	0.5 mM Methionin	53 ± 7	12.9 ± 1.5
3 mg Cd kg^{-1}	Control	9826 ± 216	10.8 ± 2.1
3 mg Cd kg^{-1}	0.5 mM L-Histidine	7559 ± 170	13.6 ± 2.3
3 mg Cd kg^{-1}	0.5 mM Methionin	8846 ± 177	14.1 ± 1.7

Grain Cd and Zn Concentrations

Although studies so far have not identified a specific membrane transporter for Cd, it is generally believed that Cd is taken up into cells in ionic form via low-affinity membrane transporters that are involved in the uptake of Ca, Fe, Mg, Cu and Zn based on their divalent and chemical similarity (Ismael et al., 2019). It has been frequently encountered in the literature that Zn, which is chemically similar to Cd, has both antagonistic (Grant et al., 2002; Özkutlu and Erdem, 2018; Özkutlu and Kara, 2018; Erdem et al., 2012) and synergistic (Zhongren et al., 2002) effects on Cd uptake. It is thought that there is a relationship between amino acids and metal uptake. On the other hand, Khan et al. (2019) reported that L-Histidine and Methionine are amino acids that are associated with metal uptake and transport in plants. Methionine is a sulfur-containing amino acid that can form strong stable complexes with Cd, which is effective in the biosynthesis of plant growth hormones. These amino acids are thought to have an active role in Cd and Zn transport. In our research, it was determined that the Cd and Zn deposits accumulated in the grain made a difference as a result of the application of L-Histidine and Methionine amino acids from the leaves. Interestingly, with the application of these amino acids, it was determined that while the Zn concentration in the grain increased, the Cd accumulation decreased. For example, while the grain Cd concentration was 62 µg kg^{-1} in the control without foliar amino acid application, the Zn concentration was determined as 10.4 mg kg^{-1} (Table 2). In both foliar L-Histidine and Methinonin applications, it was found that the grain Cd concentration decreased to 57 and 53 µg kg^{-1}, respectively. It is seen that this situation is associated with an increase in the Zn concentration in the grain and is due to the antagonistic relationship between Cd and Zn. Even more striking results were obtained when the size of Cd contamination in the soil increased. In the experiment, while the Cd concentration was 9826 µg kg^{-1} in the control, where 3 mg kg^{-1} of Cd was applied and no amino acid application was made from the leaf, as a result of the improvement in the plant with the L-Histidine application, the grain Cd concentration decreased by 30% to 7559 µg kg^{-1} (Table 2). Despite the decrease in Cd in the grain, it was determined that the Zn concentration increased from 10.8 to 13.6 mg kg^{-1}. It was determined that this situation prevented the transport of Cd by transporting more Zn to the grain due to the decrease in the stress of the plants with the application of L-Histidine from the leaves. As a result of the application of the other amino acid, Methionine, it was determined that there was a similar tendency to decrease in grain Cd concentration. However, it was determined that the reduction of grain Cd concentration by Methionine application was at a level of 11% which is more limited compared to L-Histidine. According to the results obtained, it was determined that Zn had a blocking / limiting effect on Cd uptake and transport, indicating that Zn had an inhibitory effect on Cd uptake through roots. Possibly, it is concluded that Zn and Cd, which are chemically very similar to each other, compete for their absorption points (carrier proteins, ion channels) on the plasma membranes and this competition turns in favor of Zn. Inside a cell, only a small fraction of a metal is determined as a free cation, and metal ions that are not bound to proteins or peptides are mostly bound to low molecular weight organic acids and amino acids (Dalir et al., 2017).

The results obtained are probably related to the fact that plants under Cd stress take up more Zn to tolerate Cd toxicity, and alternatively, by foliar application of amino acids, the Zn adsorbed especially in the cell walls of the plant is more uptaken by plants, resulting in less granulation of Cd. resulted in relocation. These findings showed that L-Histidine and Methionine applications promoted more uptake of Zn and had a reducing effect on Cd transport in the plant. Similar results to this situation are supported by several researchers. For example, it was explained by Hart et al. (2002) that there is a common carrier protein for both Zn and Cd on plasma membranes, and that Zn and Cd compete for binding sites on this protein during absorption. In addition, Gomez et al. (2002) suggested that the uptake and transport of Cd in yeast cells is controlled by the Zn-specific carrier protein on the plasma membranes (Gomez et al., 2002). In addition to this, according to a study conducted in wheat plant, it was found that the transport of Cd from the old leaves to the young parts of the plant decreased with increasing Zn nutrition (Cakmak et al., 2000). As seen from these studies, it has been determined that L-Histidine and Methionine have a special role in reducing Cd transport into the grain.

Conclusion

Our results showed that foliar amino acid applications had a huge effect on wheat grain Cd uptake and both amino acids limited the grain Cd uptake. However, it has been demonstrated that L-Histidine has a greater capacity to inhibit Cd transport than the amino acid Methionine. According to the findings obtained from these results, it was obtained that both amino acids can form complexes with Cd. Apart from this effect, it has been determined that it also promotes growth and quality in plants.

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