

Response of maize to soil applied ZnSO₄ and ZnEDTA

Rita KREMPER¹, Andrea BALLA KOVÁCS¹, Evelin JUHÁSZ¹,
Márk RÉKÁSI², Anita SZABÓ^{2*}, Péter CSATHÓ²

¹Institute of Agricultural Chemistry and Soil Science, Faculty of Agricultural and Food Sciences and Environmental Management, University of Debrecen, Böszörményi Street 138. H-4032, Debrecen, Hungary; kremper@agr.unideb.hu; kovacs@agr.unideb.hu; juhasz.evelin@agr.unideb.hu

²Institute for Soil Sciences, HUN-REN Centre for Agricultural Research, Fehérvári Street 144. H-1116, Budapest, Hungary; rekasi.mark@atk.hun-ren.hu; szabo.anita@atk.hun-ren.hu (*corresponding author); csatho.peter@atk.hun-ren.hu

Abstract

In this study the fertilizing effect of ZnEDTA (zinc ethylenediaminetetraacetate), and ZnSO₄·7H₂O (ZnSO₄) (zinc sulphate heptahydrate) was compared on calcareous loam soil. A pot experiment was set up with maize (*Zea mays* L. var. 'P37N01'). Plants were treated with basic NPK doses and with Zn at increasing rates: 0, 2.5, 5.0 and 10 mg kg⁻¹ Zn in ZnEDTA and in ZnSO₄ form, respectively. The ZnEDTA and ZnSO₄ treatments increased the dry matter production compared to control by an average of 16%. Applying 2.5 mg kg⁻¹ Zn, shoot Zn uptake was 1.3-fold greater by plants treated with ZnEDTA, than that of treated with ZnSO₄. In the case of higher rates (5.0 mg kg⁻¹ Zn and 10 mg kg⁻¹ Zn), Zn uptake from ZnEDTA was approximately twice as much as that of ZnSO₄. This result confirms that ZnEDTA as Zn complex is more effective than ZnSO₄ on calcareous soil. The increasing ZnEDTA and ZnSO₄ doses enhanced the soil's initial diethylenetriaminepentaacetic acid (DTPA) Zn concentration to 1.8, 3.0, 6.3 mg kg⁻¹ and 1.9, 3.5, and 6.5 mg kg⁻¹, respectively. Contrary to our expectations, DTPA soil extractant was not able to indicate the difference in Zn availability between soils treated with ZnEDTA and ZnSO₄. During the experiment, 0.6-1.7% of the added Zn doses were absorbed by the plants, and 48-59% of it could be measured back in the form of DTPA-Zn for both Zn fertilizers, the other half of the added Zn presumably converted to less available Zn forms.

Keywords: Hungary; maize; soil fertilizer; ZnSO₄; ZnEDTA

Introduction

Zinc (Zn) deficiency is a global problem both in human and crop nutrition (Yadav *et al.*, 2023). It can lead to stunted growth in children and increase the susceptibility to infectious diseases. Approximately 155 million children have Zn deficiency induced malnutrition globally (Balk *et al.*, 2019). Zinc deficiency of plants can be eliminated effectively by Zn fertilization, and as a consequence, Zn concentration in cereal grains can be increased to the optimum level (Prasad *et al.*, 2013). Zinc is an essential micronutrient for human health, playing key roles in immune function, growth, and cellular metabolism. Zinc deficiency affects an estimated 17% of the global population, particularly in developing countries where cereals are the primary staple food.

Received: 15 Jul 2024. Received in revised form: 15 Oct 2024. Accepted: 31 Oct 2024. Published online: 18 Dec 2024.

From Volume 49, Issue 1, 2021, Notulae Botanicae Horti Agrobotanici Cluj-Napoca journal uses article numbers in place of the traditional method of continuous pagination through the volume. The journal will continue to appear quarterly, as before, with four annual numbers.

Due to the low availability of zinc in cereals, increasing its levels through farming methods or breeding techniques can greatly improve human health, particularly in populations at risk (Cakmak, 2009). Rice, wheat, together with maize are the three most important grains of the human diet and approximately one-third of the calories are from these crops (Soto-Gómez and Pérez-Rodríguez, 2022). Of the three crops, maize is the most susceptible to Zn shortages, it is also known as a Zn deficiency indicator plant (Suganya *et al.*, 2020). Wheat is also sensitive to Zn deficiency when it is grown on calcareous soils as well as rice grown on soils that are flooded (Alloway, 2009; Saleem *et al.*, 2022).

Worldwide, nearly 50% of the arable soils contain inadequate available Zn (Alloway, 2008). About 33% of Hungarian soils are also Zn deficient (Várallyay *et al.*, 2010). The zinc supply of the plant is most often provided by soil or foliar zinc fertilizers but organic fertilizers or composts can also have a high Zn content (Ragályi and Kádár, 2012). The application of different Zn fertilizers continues to receive great interest. Starter fertilizers for plants with high Zn demand such as maize often contain Zn. Nutrients applied in starter fertilizers provide the direct nutrient supply of the germinating, rooted plant. Even if the soil is not Zn deficient high yield can be connected with intensive use of soils, thus nutrients such as Zn are to be replaced (Gabryszuk *et al.*, 2020).

The most commonly used Zn fertilizer is (ZnSO_4), this is followed by ZnEDTA which is a chelated source of Zn (Alloway, 2008). The application of zinc-oxide (ZnO) is not so common, but according to Mazhar *et al.* (2023) its use as nanoparticle fertilizer is very promising. ZnEDTA as a soil fertilizer can be more effective on certain soils than that of ZnSO_4 , however it is also more expensive (Zhao *et al.*, 2016). Naik and Das (2008) compared the effect of ZnSO_4 vs. ZnEDTA as soil fertilizer in a rice cultivating experiment (soil properties: DTPA-Zn = 0.78 mg kg^{-1} , pH = 7.2). As much as 0.5 kg ha^{-1} Zn dose as ZnEDTA resulted in a higher yield increase than 20 kg ha^{-1} Zn as ZnSO_4 . The effectiveness ratios of ZnEDTA to ZnSO_4 can vary greatly depending on the soil properties, the plant species, the fertilizer application methods, and the degree of Zn deficiency.

The purpose of this study was to compare the effect of soil applied ZnEDTA and ZnSO_4 on the dry matter (DM) production, Zn concentration, and Zn uptake of maize shoots obtained in a pot experiment set up on Chernozem soil with 2.9% CaCO_3 content. Observations include the Zn content of (DTPA) soil extracts and of the maize shoots, and the impact of the treatments on the photosynthetic and mineral composition of the leaves and shoots.

Materials and Methods

Description of the study site

A greenhouse experiment was set at the University of Debrecen (Hungary). The soil sample was collected from the surface (0-20 cm) soil layer in Nádudvar (GPS 47.464968, 21.233613). The most important soil characteristics were the following: soil type according to the World Reference Base classification system was Chernozem, $\text{pH}_{\text{H}_2\text{O}} = 7.28$, the texture was loam, soil organic matter (SOM) = 3.2%, CaCO_3 content = 2.9%. The ammonium-lactate (AL) extractable element concentrations were measured according to Egnér *et al.* (1960) AL $\text{P}_2\text{O}_5 = 186 \text{ mg kg}^{-1}$, AL $\text{K}_2\text{O} = 242 \text{ mg kg}^{-1}$. The NPK supply of the studied soil was determined based on the Hungarian fertilizer advisory system (MÉM-NAK, 1979). Original soil N and K supplies were good, while the P supply was medium. DTPA-Zn concentration was 0.7 mg kg^{-1} , and it was measured according to Lindsay and Norvell (1978). Deficiency symptoms in the studied soil on maize were observed under field conditions.

Treatments and design of the study

The experimental soil was dried, crushed then passed through a 10 mm diameter sieve. Twenty-eight pots were filled with 2.4 kg of air-dry soil. The test plant was maize (*Zea mays L.*, var. P37N01). Plants were grown in 2.5 L polypropylene pots (17.5 cm upper diameter, 12 cm lower diameter, and 15.5 cm high).

Seven treatments were replicated four times. Zinc rates were the following: 0, 2.5, 5.0 and 10 mg kg⁻¹ Zn as ZnEDTA and ZnSO₄ (Table 1). The NPK rates were uniform for all pots: 100 mg kg⁻¹ N as NH₄NO₃, 100 mg kg⁻¹ of P₂O₅ in the form of KH₂PO₄ and 100 mg kg⁻¹ of K₂O as KCl and KH₂PO₄. The applied fertilizer doses were dissolved in 50 cm³ water, mixed thoroughly with the soil in a bowl, then the soil was poured back into the pot. The NPK rates were set taking into account the specific nutrient requirements of maize and the soil original NPK supply as well.

Table 1. Zinc treatments of the experiment

Treatments	Zn rate (mg kg ⁻¹)	Zn fertilizer form	Code
1	0	-	control
2	2.5	ZnEDTA	2.5 ZnEDTA
3	5.0		5.0 ZnEDTA
4	10		10 ZnEDTA
5	2.5	ZnSO ₄	2.5 ZnSO ₄
6	5.0		5.0 ZnSO ₄
7	10		10 ZnSO ₄

Four maize seeds were sown on April 30th. Pots were irrigated to 60% of water holding capacity. After germination two plants were removed and the two healthy seedlings remained in the pots until harvest. At eight leaves plant growth stages (46 days after sowing) SPAD (Soil Plant Analysis Development) values of the 7th leaves from base were measured with SPAD-502 meter (Minolta Corporation, Ramsey, NJ). SPAD values relate to the relative chlorophyll content of the plant. Maize plants at 71 days after sowing were harvested, the above ground part of the plants was cut, chopped, air-dried then grounded.

Soil and plant analysis

The whole shoots were dried at 60 °C and the DM production was determined, then the above-ground part of the plant was digested with HNO₃-H₂O₂ mixture according to the European standard (EN14084, 2003). The Zn, Ca, Mg, and Mn concentration of the plant were measured by Flame Atomic Absorption Spectrometry (FAAS) technique (Varian Spectr. AA-20), and the K concentration with a flame emission photometer (UNICAM SP95B). The P concentration of the plants was determined from sulphuric acid digestion with ammonium molybdenate vanadate UV-VIS spectrophotometry method. The Zn, Ca, Mg, Mn, and P uptake of plants were calculated by multiplying the shoot masses and the element concentrations. The DTPA Zn concentration of soil samples was also measured following the harvest.

The Zn fertilizer use efficiency was expressed in percentage and calculated by the following formula:

$$\text{Zn fertilizer use efficiency} = \frac{\text{Zn uptake of Zn fertilized plants} - \text{Zn uptake of control plants}}{\text{added Zn}} \times 100$$

The statistical analyses were conducted with SPSS Statistics 20 software. One way ANOVA analysis and Tukey's post hoc test at $P \leq 0.05$ were performed to compare the effects of the treatments. Significant differences between the treatments were indicated with different letters. The data in the figures and tables are presented as the mean \pm 2 SEM (standard error of the means).

Results

The ZnEDTA and ZnSO₄ treatments increased the DM production significantly compared to the control (averaged across Zn doses,) (Figure 1). However, the effect of the separate treatment could not be proved statistically in each treatment, due to the high standard deviations. The increment was significant in the case of 5 mg kg⁻¹ ZnEDTA and 2.5 mg kg⁻¹ ZnSO₄. In the average of the Zn doses, the effect of the two Zn forms was equal, i.e., the ZnSO₄ form resulted in 1% higher DM production as compared to the ZnEDTA form.

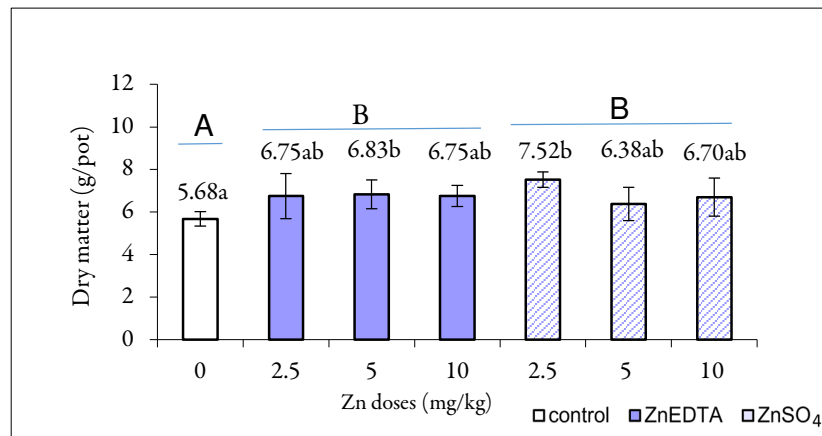


Figure 1. The effect of ZnEDTA and ZnSO₄ rates on maize shoot dry matter production. Different capital letters indicate significant differences between treatments averaged across Zn forms. Different small letters indicate significant differences between the individual fertilization treatments.

The effects of Zn treatment on the Zn concentration of plants were much more considerable than that on DM production (Figure 2). As an effect of 2.5 mg kg⁻¹ ZnEDTA and ZnSO₄ application, shoot Zn concentrations increased by 130 and 60%, respectively, compared to the control. The maize Zn concentration by ZnEDTA treatments was nearly twice as much as that by ZnSO₄ treatment.

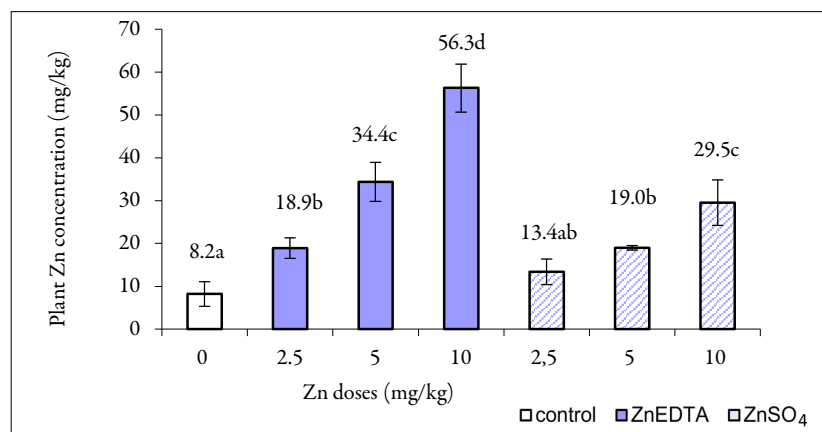


Figure 2. The effect of ZnEDTA and ZnSO₄ rates on maize shoot Zn concentration. Different small letters indicate significant differences between the individual fertilization treatments.

The DTPA available Zn gradually increased with the applied Zn rates (Figure 3). As an effect of 2.5 mg kg⁻¹ ZnEDTA and ZnSO₄ application, DTPA soluble soil Zn concentrations increased by 200 and 220%,

respectively, compared to the control. There were no significant differences between the extracted Zn concentrations of ZnEDTA and ZnSO₄ treatments with the same Zn dose.

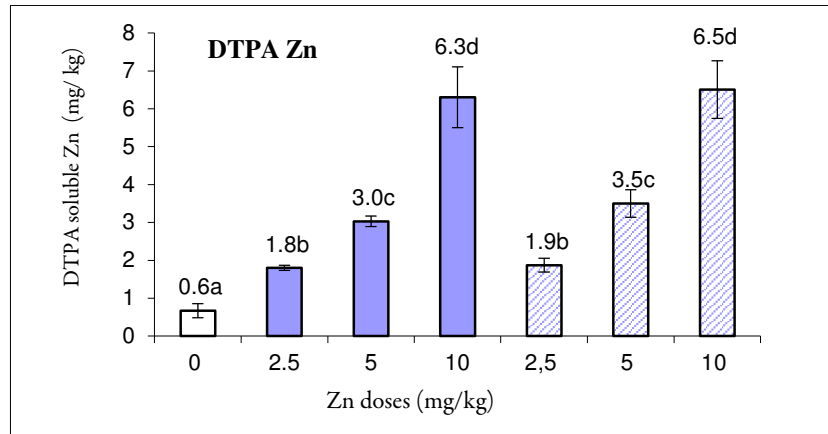


Figure 3. The changes in DTPA soluble Zn concentration of the soil after harvesting the experiment. Different small letters indicate significant differences between the individual fertilization treatments.

The Zn uptake showed a similar trend to Zn concentration (Table 2). In the average of the Zn doses, ZnSO₄ resulted in only 56% shoot Zn uptake as compared to the effect of ZnEDTA. During the experiment at most 0.6 to 1.7% of the added Zn doses were absorbed by the plants. The highest Zn utilization value (1.7%) was obtained in 5 mg kg⁻¹ ZnEDTA treatment. Zinc balances were strongly positive in the Zn fertilized pots and increased with Zn rates. Only 48 to 59% of the added Zn can be measured back in the form of DTPA-Zn. The net DTPA Zn recoveries (Zn fertilized – control), measured after harvest increased together with the Zn rates (Table 2).

Table 2. Zn turnover of the pots, depending on the added Zn fertilizer forms and rates

Zn rate	Added Zn	Zn uptake	Zn fertilizer use efficiency	Zn balance	Final DTPA Zn	$\frac{\Delta(\text{DTPA Zn})}{\text{added Zn}} \cdot 100$
(mg kg ⁻¹)	(mg pot ⁻¹)		%	(mg pot ⁻¹)		%
control	0	0.046	-	-0.046	1.44	-
2.5 ZnEDTA	6	0.13	1.40	5.87	4.32	48
5.0 ZnEDTA	12	0.247	1.68	11.75	7.20	48
10 ZnEDTA	24	0.379	1.39	23.62	15.12	57
2.5 ZnSO ₄	6	0.101	0.92	5.90	4.56	52
5.0 ZnSO ₄	12	0.121	0.62	11.88	8.40	58
10 ZnSO ₄	24	0.199	0.64	23.80	15.60	59

$\Delta(\text{DTPA Zn}) = \text{DTPA Zn of Zn fertilized pots} - \text{DTPA Zn of the control pots}$

The zinc doses and forms had no significant effect on the K concentration and uptake of plants, which ranged from 2.89 to 3.59% (Table 3). These values are considered as good K supplies. The P concentration of the control plants (0.26% P) was significantly larger than those of the Zn treated plants. Lower P concentrations were obtained in the ZnEDTA treatments (0.17 to 0.19% P) than in the ZnSO₄ treatments (0.19 to 0.22% P).

Table 3. Effect of Zn doses and forms on the element concentration, uptake and SPAD values of plants

Zn rate (mg kg ⁻¹)	Nutrient concentration					SPAD
	K (%)	P (%)	Ca (%)	Mg (%)	Mn (mg kg ⁻¹)	
	0.079 ns	0.010*	0.014*	0.000***	0.000***	
Control	3.59 ± 0.51	0.26 ± 0.03 b	0.73 ± 0.09 b	0.44 ± 0.02 b	78.3 ± 6.6 b	26.7 ± 2.6
2.5 ZnEDTA	2.89 ± 0.37	0.17 ± 0.01 a	0.62 ± 0.04 ab	0.35 ± 0.00 a	60.8 ± 5.2 a	29.1 ± 2.6
5.0 ZnEDTA	2.91 ± 0.49	0.19 ± 0.01 a	0.62 ± 0.04 ab	0.35 ± 0.02 a	57.6 ± 5.3 a	30.8 ± 2.0
10 ZnEDTA	2.89 ± 0.36	0.17 ± 0.01 a	0.64 ± 0.06 ab	0.36 ± 0.02 a	53.8 ± 2.4 a	29.7 ± 1.4
2.5 ZnSO ₄	3.09 ± 0.32	0.22 ± 0.01 ab	0.61 ± 0.03 ab	0.37 ± 0.03 a	64.8 ± 3.5 a	29.6 ± 2.7
5.0 ZnSO ₄	3.56 ± 0.54	0.19 ± 0.01 a	0.57 ± 0.04 a	0.34 ± 0.02 a	56.3 ± 2.8 a	28.2 ± 2.5
10 ZnSO ₄	3.04 ± 0.24	0.20 ± 0.03 a	0.61 ± 0.03 a	0.34 ± 0.02 a	59.8 ± 5.7 a	29.3 ± 2.0
Zn rate (mg kg ⁻¹)	Nutrient uptake					
	K (mg pot ⁻¹)	P (mg pot ⁻¹)	Ca (mg pot ⁻¹)	Mg (mg pot ⁻¹)	Mn (µg pot ⁻¹)	
	0.348 ns	0.061 ns	0.182 ns	0.123 ns	0.058 ns	
Control	203 ± 37	14.30 ± 2.56	41.3 ± 4.4	25.4 ± 1.1	447 ± 29	
2.5 ZnEDTA	195 ± 41	11.95 ± 1.98	42.6 ± 6.0	24.6 ± 3.8	414 ± 68	
5.0 ZnEDTA	208 ± 48	13.70 ± 1.04	44.7 ± 6.9	25.6 ± 3.1	414 ± 71	
10 ZnEDTA	196 ± 32	11.95 ± 0.60	43.3 ± 1.4	24.4 ± 0.3	363 ± 35	
2.5 ZnSO ₄	232 ± 31	15.81 ± 1.20	46.0 ± 2.3	27.9 ± 3.2	487 ± 13	
5.0 ZnSO ₄	230 ± 78	11.50 ± 1.12	36.2 ± 3.4	21.9 ± 3.8	360 ± 58	
10 ZnSO ₄	202 ± 14	15.65 ± 3.78	41.0 ± 7.0	22.7 ± 2.4	404 ± 86	

* P ≤ 0.05; ** P ≤ 0.01; *** P ≤ 0.001; ns = not significant; Different small letters indicate significant differences between the individual fertilization treatments within a column.

In our pot trial, the Ca and Mg concentration ranged from 0.57 to 0.73% and from 0.34 to 0.44% respectively (Table 3). In the case of Mn the concentration values ranged between 54 to 78 mg kg⁻¹. Generally, the K, P, Ca, Mg and Mn uptake did not change significantly with increasing Zn doses. The SPAD values of the upper developed leaves in control treatment were 26.7, while in the ZnEDTA treatments, they varied between 29.1 to 30.8 and in the ZnSO₄ treatments, between 28.2 and 29.6.

Discussion

The effect of increasing ZnEDTA and ZnSO₄ doses was reflected in the plant DM production, the Zn concentration, and uptake and the DTPA Zn concentration of soil, but the magnitude of the effect differed for each variable. While the DM production had a very slight increment with increasing Zn doses, the plant Zn concentration and Zn uptake responded to a great extent. From Zn concentration values of plant, the Zn supply related to certain treatments can be evaluated.

According to Singh and Banerjee (1987) the critical value for Zn concentration for maize (70th day, shoot) is 9.5 mg kg⁻¹. The Zn concentration of the shoots reached adequate values approximately by 2.5 mg kg⁻¹ ZnEDTA and 5 mg kg⁻¹ ZnSO₄. Converting this value to kg ha⁻¹ we get 6 kg ha⁻¹ Zn for ZnEDTA and 12 kg ha⁻¹ Zn for ZnSO₄ (the bulk density of the soil was 1.2 g cm⁻³ and the soil layer depth was 20 cm). 10 mg kg⁻¹ Zn rates were still not toxic for the plant, as the shoot Zn concentrations were in the range of proper supply.

Maize is sensitive to Zn deficiency but not sensitive to excessive Zn. Baran (2013) stated that maize Zn concentration at which 20% yield decline occurs was 745 mg kg⁻¹. In accordance with this no negative effect of even extremely high concentrations of Zn could be demonstrated on different soil microbial parameters (Szécsy *et al.*, 2011).

Zinc uptake values of maize are suitable to compare the efficiency of the ZnEDTA to ZnSO₄. According to it the efficiency of 2.5 mg kg⁻¹ Zn in ZnEDTA form is slightly larger than that of 5 mg kg⁻¹ Zn in ZnSO₄ form (Table 2). Due to the soil high CaCO₃ content (2.9%) presumably one part of ZnSO₄ was fixed, as the presence of CaCO₃ causes the precipitation of Zn-carbonate and Zn-hydroxide or calcium zincate (Rico *et al.*, 1996; Aljumaily *et al.*, 2022). At pH > 6 Fe oxides also adsorb Zn in an irreversible way (Rengel, 2015). Zinc in ZnEDTA form was prevented from forming precipitation, therefore, it was a more effective Zn source of plants. Recena *et al.* (2021) found that the soil DTPA-Zn characterizes the plant Zn uptake better than the applied ZnSO₄ dose.

The critical level of DTPA Zn is 0.8 mg kg⁻¹ and 0.6 mg kg⁻¹ according to Lindsay and Norvell (1978), and Srivastava and Gupta (1996), respectively. At the same time, Zare *et al.* (2009) obtained a 1.5 mg kg⁻¹ threshold value for DTPA Zn on calcareous Iranian soils, which calcium carbonate content were higher than 30%.

The DM production was 14% lower in the case of control soil with 0.6 mg kg⁻¹ DTPA Zn than that on Zn fertilized soil with 1.8 mg kg⁻¹ DTPA Zn. Thus, it can be stated that the control soil was Zn deficient and the DTPA critical value is larger than 0.6 mg kg⁻¹ on the studied soil. As an effect of Zn fertilization, the soil DTPA Zn concentration (1.8 mg kg⁻¹) exceeded even Zare's critical value (1.5 mg kg⁻¹), and the DM production results confirmed that the soil with 1.8 mg kg⁻¹ DTPA Zn was not Zn deficient. However, the adequate DTPA-Zn concentration of soil does not always guarantee a good Zn supply for all soils. Zhao *et al.* (2011) found that the application of 7.5 kg Zn ha⁻¹ as ZnSO₄ increased the soil DTPA soluble Zn concentration by 270% after 7 days, but despite this, the Zn fertilization had no effect on the wheat grain concentration on calcareous soil. In our experiment, we observed that even though DTPA Zn concentrations of ZnEDTA and ZnSO₄ treatments with the same Zn dose were similar (3 and 3.5 mg kg⁻¹ respectively, at 5 mg kg⁻¹ Zn dose) the Zn uptake values related to them were significantly different (247 and 121 mg Zn / pot). The plant Zn concentration and / or uptake characterizes plant available Zn supply much more sensitively than DTPA soluble Zn concentration.

The sum of the final DTPA Zn and the plant Zn uptake is lower than the added Zn (Table 2). It suggests the fact that during the experiment a major part (around half) of the added Zn was converted into less available forms. Contrary to our expectations there were no significant differences between Zn concentration of DTPA extracts of soils treated with ZnEDTA and ZnSO₄. According to Sharma *et al.* (2004) DTPA primarily extracts Zn from organic fraction and exchange sites of coarse clays. Less mobile forms of Zn include the above-mentioned precipitates (Zn-carbonate and Zn-hydroxide or calcium zincate), Zn phosphate, Zn forms adsorbed by iron-oxides, manganese oxides and Zn specifically adsorbed to clay minerals. According to González-Costa *et al.* (2017), clay minerals are the main sorbent surfaces of Zn in soil.

Zinc fertilization can affect the concentration and uptake of other elements in the plant (Table 3). P induced Zn deficiency is a well-known limiting factor of Zn availability. In our experiment, 100 mg kg⁻¹ P₂O₅ rate was applied in each pot because of the medium P supply of the initial soil. The P/Zn ratio of plants was 317 in the control treatment. Csathó *et al.* (2019) observed P induced Zn deficiency when the P/Zn ratio exceeds 200 in the case of flowering stage maize leaves. As a result of Zn fertilization, the P/Zn ratio decreased gradually with Zn rates and varied between 54 and 100. Therefore, in Zn fertilized pots the applied P doses did not hinder the plant Zn uptake.

In our experiment, the P concentration of the plants treated with Zn was significantly lower than that of the control plants (Table 3). On average, there was a 27% decrease. Similarly to our observation Sánchez-Rodríguez *et al.* (2021) also observed significant decrease of P concentration of maize as an effect of Zn fertilization in a greenhouse experiment. This can be explained mainly by the dilution effect due to the larger biomass. The DM production increased by 16%, on average, compared to the control, therefore the extent of dilution effect cannot be more than that. At the same time, P-Zn antagonism also contributed to the decrease in P concentration. The added Zn could also hinder P uptake, and a reduction in transport of P to the plant tops can be assumed as well.

Although 0; 2.5; 5 and 10 mg kg⁻¹ Zn (0, 6, 12 and 24 kg ha⁻¹ Zn) applied in a pot experiment may be relatively high Zn doses, the 100 mg kg⁻¹ (240 kg ha⁻¹) P₂O₅ dose probably provided almost sufficient P to the plant, since no visible P deficiency symptoms occurred, although P concentrations were below the critical P value (8-leaf stage), which is 0.25%.

Based on the Ca and Mg concentration of the plants, the Ca and Mg supply was adequate as sufficiency levels range from 0.25% to 0.8% and from 0.15% to 0.6% related to Ca and Mg respectively (upper mature leaf). The relatively high CaCO₃ content of soil did not shift Ca/Mg ratio unfavorably. Zinc fertilization reduced Ca and Mg concentration compared to the control, which can be explained by the dilution effect (Table 3). The fact that the Ca and Mg uptake of plants did not change with Zn fertilization confirms this assumption. Comparing the plants' Mn concentration (54-78 mg kg⁻¹) to the sufficient range of Mn (40-100 mg kg⁻¹) it can be stated that the Mn supply of the plants was also adequate. The decrease of Mn concentration of Zn treated plants can be attributed to the dilution effect and Mn-Zn antagonism as well.

The SPAD values averaged across ZnEDTA treatments increased significantly compared to the control (26.7a vs. 29.85b) but this tendency can not be proved statistically for the separate treatments due to the high standard deviations. Zn acts as a cofactor of enzymes involved in pigment biosynthesis. Wang *et al.* (2009) and Tavallali (2017) also found, that Zn deficiencies cause a reduction in photosynthesis. It is reflected in our results as well.

Conclusions

The studied soil with 0.7 mg kg⁻¹ DTPA Zn was deficient in Zn, as the applied Zn fertilization increased the DM production. According to the shoot Zn concentrations, we concluded that either 2.5 mg kg⁻¹ Zn as ZnEDTA or 5 mg kg⁻¹ Zn as ZnSO₄ is the most favorable rate to eliminate Zn deficiency on the studied calcareous chernozem soil. Maize Zn uptake results also proved that ZnEDTA was twice as effective as ZnSO₄ under the studied circumstances. This result confirms the advantages of ZnEDTA as Zn complex on calcareous soil. After the experiment, 48-59% of the applied Zn could be measured back in the form of DTPA-Zn for both Zn fertilizers. The soil DTPA Zn concentration was approximately the same in the case of ZnEDTA and ZnSO₄ treatments, DTPA soil extractant could not indicate the difference in bioavailability of the inorganic and chelated Zn forms.

Authors' Contributions

R.K. and P.Cs. designed the experiments, wrote the manuscript. A.B.K. and E.J. made the greenhouse experiment and the analytical measurements. M.R. and A.Sz. did editing and made the statistical analysis. All authors read and approved the final manuscript.

Ethical approval (for researches involving animals or humans)

Not applicable.

Acknowledgements

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

References

- Aljumaily M, Al-Hamndi H, Jamal Al-obaidi M, Al-Zidan R (2022). The effect of calcium carbonate content on the zinc quantity-intensity relationship in some soils of Mosul, Irak. *Ciencia & Tecnología Agropecuaria* 23. https://doi.org/10.21930/rcta.vol23_num1_art:2373
- Alloway BJ (2008). Micronutrients and crop production: An introduction. In: *Micronutrient deficiencies in global crop production*. pp 1-39. Springer.
- Alloway BJ (2009). Soil factors associated with zinc deficiency in crops and humans. *Environmental Geochemistry and Health* 31(5):537-548. <https://doi.org/https://doi.org/10.1007/s10653-009-9255-4>
- Balk J, Connorton J, Wan Y, Lovegrove A, Moore K, Uauy C, Sharp P, Shewry P (2019). Improving wheat as a source of iron and zinc for global nutrition. *Nutrition Bulletin* 44(1):53-59. <https://doi.org/https://doi.org/10.1111/nbu.12361>
- Baran A (2013). Assessment of *Zea mays* sensitivity to toxic content of zinc in soil. *Polish Journal of Environmental Studies* 22(1).
- Cakmak I (2009). Enrichment of fertilizers with zinc: An excellent investment for humanity and crop production in India. *Journal of Trace Elements in Medicine and Biology* 23(4):281-289. <https://doi.org/https://doi.org/10.1016/j.jtemb.2009.05.002>
- Csathó P, Árendás T, Szabó A, Sándor R, Ragályi P, Pokovai K, Tóth Z, Kremper R (2019). Phosphorus-induced zinc deficiency in maize (*Zea mays* L.) on a calcareous chernozem soil. *Agrokémia és Talajtan* 68(1):40-52. <https://doi.org/https://doi.org/10.1556/0088.2018.00016>
- Egnér H, Riehm H, Domingo W (1960). Investigations on chemical soil analysis as the basis for estimating soil fertility. II. Chemical extraction methods for phosphorus and potassium determination. *Kungliga Lantbrukshögskolans Annaler* 26:199-215.
- EN14084 (2003). Determination of trace elements, determination of lead, cadmium, zinc, copper and iron by atomic absorption spectrometry (AAS) after microwave digestion.
- Gabryszak M, Barszczewsk J, Kuźnicka E, Sakowski T (2020). Effect of long-term fertilization of the permanent dry meadow on the zinc content in soil and meadow sward. *Journal of Water and Land Development* (47). <https://doi.org/https://doi.org/10.24425/jwld.2020.135032>
- González-Costa JJ, Reigosa, MJ, Matías JM, Fernandez-Covelo E (2017). Analysis of the importance of oxides and clays in Cd, Cr, Cu, Ni, Pb and Zn adsorption and retention with regression trees. *Public Library of Science* 12(1):e0168523. <https://doi.org/https://doi.org/10.1371/journal.pone.0168523>
- Lindsay WL, Norvel, W (1978). Development of a DTPA soil test for zinc, iron, manganese, and copper. *Soil Science Society of America Journal* 42(3):421-428. <https://doi.org/https://doi.org/10.2136/sssaj1978.03615995004200030009x>
- Mazhar Z, Akhtar J, Alhodaib A, Naz T, Zafar MI, Iqbal MM, Fatima H, Naz I (2023). Efficacy of ZnO nanoparticles in Zn fortification and partitioning of wheat and rice grains under salt stress. *Scientific Reports* 13(1):2022. <https://doi.org/10.1038/s41598-022-26039-8>
- MÉM-NAK (1979). Fertilization guidelines and calculation methods (in Hungarian) *Mezőgazdasági Kiadó*.
- Naik SK, Das DK (2008). Relative performance of chelated zinc and zinc sulphate for lowland rice (*Oryza sativa* L.). *Nutrient Cycling in Agroecosystems* 81:219-227. <https://doi.org/https://doi.org/10.1007/s10705-007-9158-7>
- Prasad R, Shivay YS, Kumar D (2013). Zinc fertilization of cereals for increased production and alleviation of zinc malnutrition in India. *Agricultural Research* 2:111-118. <https://doi.org/https://doi.org/10.1007/s40003-013-0064-8>
- Ragályi P, Kádár I (2012). Effect of organic fertilizers made from slaughterhouse wastes on yield of crops. *Archives of Agronomy and Soil Science* 58(1):S122-S126.

- Recena R, García-López AM, Delgado A (2021). Zinc uptake by plants as affected by fertilization with Zn sulfate, phosphorus availability, and soil properties. *Agronomy* 11(2):390. <https://doi.org/https://doi.org/10.3390/agronomy11020390>
- Rengel Z (2015). Availability of Mn, Zn and Fe in the rhizosphere. *Journal of Soil Science and Plant Nutrition*, 15(2):397-409.
- Rico MI, Alvarez JM, Mingot JI (1996). Efficiency of zinc ethylenediaminetetraacetate and zinc lignosulfonate soluble and coated fertilizers for maize in calcareous soil. *Journal of Agricultural and Food Chemistry* 44(10):3219-3223.
- Saleem MH, Usman K, Rizwan M, Al Jabri H, Alsafran M (2022). Functions and strategies for enhancing zinc availability in plants for sustainable agriculture. *Frontiers in Plant Science* 13:1033092. <https://doi.org/10.3389/fpls.2022.1033092>
- Sánchez-Rodríguez AR, Rey MD, Nechate-Drif H, Castillejo MÁ, Jorrín-Novo JV, Torrent J, del Campillo MC, Sacristán D (2021). Combining P and Zn fertilization to enhance yield and grain quality in maize grown on Mediterranean soils. *Scientific Reports* 11(1):7427. <https://doi.org/10.1038/s41598-021-86766-2>
- Sharma B, Arora H, Kumar R, Nayyar V (2004). Relationships between soil characteristics and total and DTPA-extractable micronutrients in Inceptisols of Punjab. *Communications in Soil Science and Plant Analysis* 35(5-6):799-818.
- Singh K, Banerjee N (1987). Critical levels of zinc in corn (*Zea mays* L.) at different stages of growth. *Indian Journal of Plant Physiology (India)* 30(1).
- Soto-Gómez D, Pérez-Rodríguez P (2022). Sustainable agriculture through perennial grains: Wheat, rice, maize, and other species. A review. *Agriculture, Ecosystems & Environment* 325:107747. <https://doi.org/https://doi.org/10.1016/j.agee.2021.107747>
- Srivastava P, Gupta UC (1996). Trace elements in crop production. Science Publishers, Inc.
- Suganya A, Saravanan A, Manivannan N (2020). Role of zinc nutrition for increasing zinc availability, uptake, yield, and quality of maize (*Zea mays* L.) grains: An overview. *Communications in Soil Science and Plant Analysis* 51(15):2001-2021. <https://doi.org/https://doi.org/10.1080/00103624.2020.1820030>
- Szécsey O, Uzinger N, Villányi I, Szili-Kovács T, Anton A (2011). Correlations between the dissolution fractions of chromium, lead and zinc and soil microbiological and biochemical parameters on a sandy soil from the Nyírség region of Hungary treated with lignite. *Agrokémia és Talajtan* 60(2):383-396. <https://doi.org/https://doi.org/10.1556/agrokem.60.2011.2.7>
- Tavallali V (2017). Interactive effects of zinc and boron on growth, photosynthesis, and water relations in pistachio. *Journal of Plant Nutrition* 40(11):1588-1603. <https://doi.org/10.1080/01904167.2016.1270308>
- Várallyay G, Szabóné-Kele G, Berényi-Üveges J, Marth P, Karkalik A, Thury I (2010). Soil conditions in Hungary: based on the data of the Soil Conservation Information and Monitoring System (SIMS). Ministry of Agriculture and Rural Development.
- Wang H, Liu R, Jin J (2009). Effects of zinc and soil moisture on photosynthetic rate and chlorophyll fluorescence parameters of maize. *Biologia Plantarum* 53:191-194. <https://doi.org/https://doi.org/10.1007/s10535-009-0033-z>
- Yadav AK, Seth A, Kumar V, Datta, A (2023). Agronomic biofortification of wheat through proper fertilizer management to alleviate zinc malnutrition: A review. *Communications in Soil Science and Plant Analysis* 54(2):154-177. <https://doi.org/https://doi.org/10.1080/00103624.2022.2110892>
- Zare M, Khoshgoftarmansh AH, Norouzi M, Schulin R (2009). Critical soil zinc deficiency concentration and tissue iron: Zinc ratio as a diagnostic tool for prediction of zinc deficiency in corn. *Journal of Plant Nutrition* 32(12):1983-1993. <https://doi.org/https://doi.org/10.1080/01904160903308101>
- Zhao A, Lu X, Chen Z, Tian X, Yang X (2011). Zinc fertilization methods on zinc absorption and translocation in wheat. *Journal of Agricultural Science* 3(1):28. <https://doi.org/https://doi.org/10.1002/jsfa.7245>
- Zhao AQ, Tian XH, Chen YL, Li S (2016). Application of ZnSO₄ or Zn-EDTA fertilizer to a calcareous soil: Zn diffusion in soil and its uptake by wheat plants. *Journal of the Science of Food and Agriculture* 96(5):1484-1491.



The journal offers free, immediate, and unrestricted access to peer-reviewed research and scholarly work. Users are allowed to read, download, copy, distribute, print, search, or link to the full texts of the articles, or use them for any other lawful purpose, without asking prior permission from the publisher or the author.



License - Articles published in *Notulae Botanicae Horti Agrobotanici Cluj-Napoca* are Open-Access, distributed under the terms and conditions of the Creative Commons Attribution (CC BY 4.0) License.

© Articles by the authors; Licensee UASVM and SHST, Cluj-Napoca, Romania. The journal allows the author(s) to hold the copyright/to retain publishing rights without restriction.

Notes:

- **Material disclaimer:** The authors are fully responsible for their work and they hold sole responsibility for the articles published in the journal.
- **Maps and affiliations:** The publisher stay neutral with regard to jurisdictional claims in published maps and institutional affiliations.
- **Responsibilities:** The editors, editorial board and publisher do not assume any responsibility for the article's contents and for the authors' views expressed in their contributions. The statements and opinions published represent the views of the authors or persons to whom they are credited. Publication of research information does not constitute a recommendation or endorsement of products involved.