The influence of moisture on the residual effects of natural zinc chelates applied to two different soils

P. Almendros*, D. Gonzalez and J.M. Alvarez

Departamento de Química y Análisis Agrícola, Escuela Técnica Superior de Ingenieros Agrónomos (ETSIA), Universidad Politécnica de Madrid (UPM), El Greco, Ciudad Universitaria s/n 28040 Madrid, Spain. *Corresponding author: p.almendros@upm.es

Abstract

The aim of this study was to compare the behavior of residual Zn from three natural chelates (Zn-aminolignosulfonate [Zn-AML],Zn-polyhydroxyphenylcarboxylate [Zn-PHP] and Zn-ethylenediamine disuccinate [Zn-EDDS]) applied at different rates (0, 5 and 10 mg Zn/kg soil) to a flax crop grown during the previous year. This incubation experiment was carried out over 75 days under two different moisture conditions (60% field capacity and waterlogged) in two different soils (acidic [Soil_{acid}]and calcareous [Soil_{calc}]). The potential available Zn concentration and short-term available Zn were estimated under both moisture conditions using the diethylenetriaminepentaacetic acid – triethanolamine (DTPA-TEA) and low-molecular-weight organic acids (LMWOAs) methods, respectively. Watersoluble Zn was estimated in both soils under 60% field capacity conditions. Immediately available Zn (the Zn concentration) was estimated under waterlogged conditions. The pH and Eh were also determined in both soils. Incubation of both soils under both moisture conditions led to decreases in the potential available Zn and short-term available Zn concentrations. Water-soluble Zn concentrations in soils under 60% field conditions, immediately available Zn in soils and the Eh parameter under waterlogged conditions also decreased with time. The residual effect of Zn-AML applied at a rate of 10 mg Zn/kg in Soil_{acid} produced the highest available and short-term Zn concentration in Soil_{cate}.

Keywords: Zn chelates, residual effect, Zn availability, short-term available Zn, immediately available Zn, watersoluble Zn

1. Introduction

When Zn is applied to the soil, water-soluble Zn is distributed between the solid phases and the soil solution. As this metal ages in the soil, changes in Zn availability, reductions in the activity and extractability of Zn forms and alterations in the more stable forms of Zn over intermediate and longer periods can be observed (Barrow, 1986; Shuman,

1991; Ma and Uren, 2006). This process is influenced by various parameters, such as moisture conditions, soil characteristics and time.

The soil moisture content has an effect on nutrient bioavailability, as it influences the mineralization of nutrients from soil organic matter. In soils, redox conditions affect aging processes and the availability of metals. Redox conditions generally exert control over pH and influence the availability of metal ions as well as the chemical forms of ions and molecules dissolved in the soil solution (McBride, 1994; Violante *et al.*, 2010). Different soil pH levels also influence Zn availability over time. In general, the concentration of water-soluble Zn decreases as the pH increases. According to Liang *et al.* (1990), under acidic soil conditions, the concentration of exchangeable Zn is high. Payne *et al.* (1988) reported that when $ZnSO_4$ was applied to high pH soil, most of the Zn was not available to plants.

According to Martinez and McBride (2000), increasing the aging time causes a decrease in Zn solubility. Various studies have reported differences in available Zn concentrations over 8-day periods of ZnSO₄ fertilizer application (Armour *et al.*, 1989), decreases in soluble Zn concentrations over 30-dayperiods of Zn(NO₃)₂ application (Barrow, 1986) and decreases in the amount of Zn in soil solutions over 3 days of carrier-free ⁶⁵Zn application (Tiller *et al.*, 1972).

Different Zn fertilizers applied to soils demonstrate different levels of reactivity, solubility and availability. In recent years, the use of Zn chelates has been proposed as a means of providing this micronutrient to obtain high concentrations of water-soluble Zn and available Zn in soils, especially in calcareous soils (Obrador et al., 2002). The effectiveness of these sources depends on the stability of the chelate in question. Several studies have demonstrated the evolution of available Zn and Zn in soil solutions (immediately available Zn) in soils planted with maize crops (Lopez-Valdivia et al., 2002, Alvarez and Rico, 2003). However, studies examining the residual effect of applying Zn in the form of natural chelates, the influence of moisture conditions on the availability of Zn and the short-term evolution of residual Zn are still very limited (Almendros et al., 2011).

In this study, we performed an incubation experiment to compare changes in the availability of residual Zn

from Zn natural chelates applied to two different soils according to moisture conditions.

2. Materials and Methods

The two original soil surface horizons used in this study were obtained from two different regions of Spain. Soil_{acid} was from Madrid (40°17' N, 4°03' W) and Soil_{cole} was from Guadalajara (40°39' N, 3°20' W). Soilacid was classified as a Typic Haploxeralf, and its main characteristics were as follows: sand, 840 g/kg; silt, 60 g/kg; clay, 100 g/kg; bulk density, 1.42 g cm⁻ ³; water-holding capacity (33 kPa), 6.60 g H₂O/100 g soil; pH, 6.13; redox potential, 458 mV; electrical conductivity, 0.037 dS/m; extractable P, 19.9 mg/kg; oxidizable OM, 5.00 g/kg; total N, 1.00 g/kg; cation exchange capacity, 4.72 cmol/kg; Fe (active Fe₂O₂), 141 mg/kg; total Zn, 9.97 mg/kg. Soil_{cale} was classified as a Typic Calcixerept, and its main characteristics were as follows: sand, 560 g/kg; silt, 260 g/kg; clay, 180 g/kg; bulk density, 1.06 g/cm³; water-holding capacity (33 kPa), 20.5 g H₂O/100 g soil; pH, 8.13; redox potential, 380 mV; electrical conductivity, 0.178 dS/m; extractable P, 12.6 mg/kg; oxidizable OM, 12.9 g/kg; total N, 1.10 g/kg; cation exchange capacity, 23.5 cmol/kg; Fe (active Fe₂O₂), 56 mg/kg; total Zn, 44.25 mg/kg (Sparks et al., 1996).

The soils were obtained from an experiment performed during the previous year, which involved growing a flax crop in a greenhouse from March to June. Before growing the flax crop, these soils were treated with aqueous suspensions of three natural chelates: Zn-aminolignosulfonate (Zn-AML), Znpolyhydroxyphenylcarboxylate (Zn-PHP) and Znethylenediamine disuccinate (Zn-EDDS). These chelates were applied at different rates: 0, 5 and 10 mg Zn/kg (Alvarez 2010). The soil from the pots was homogenized, and the pots were then placed in a greenhouse for six months. Soil (500 g) from each pot was then placed in a polyethylene container with a capacity of 600 mL. We used 84 containers in total: 21 for each soil and moisture condition with two rates of three natural chelates and a control soil (Nil-Zn). Incubation was performed under two different moisture conditions: 60% field capacity and waterlogged conditions. Waterlogged conditions were determined when the water level was 1 cm above ground level; Soil_{acid} was at five times the field capacity, and Soil_{cale} was at three times the field capacity. The containers were then sealed with Parafilm (PM-996) to prevent evaporation. The soil was mixed twice a week, and the moisture level was controlled by weight. We collected samples after 1, 15, 45 and 75 days. We then recalculated the amount of additional water needed to maintain these moisture conditions.

In the 60% field capacity samples, the available Zn concentration and short-term available Zn concentration were determined with diethylenetriaminepentaacetic acid – triethanolamine (DTPA-TEA) (Lindsay and Norwell, 1978) and low-molecular-weight organic acids (LMWOAs) (Feng *et al.*,2005), respectively. The water-soluble Zn was determined with 2.5 g of dry soil and 25 mL of deionized water; the solution was shaken for 30 min and then centrifuged (4000 rpm; 10 and 20 min for Soil_{acid} and Soil_{cale}, respectively). The weight of dry soil used was corrected based on the previous moisture calculation. Soil pH and redox potential (E_h) were measured using a Hamilton pH electrode (LP238285, 3 M KCl plus glycol electrolyte) and redox electrode (LP238145, Pt, 3 M KCl electrolyte).

In the waterlogged samples, a homogeneous 60 g sample of the supernatant was filtered using cellulose acetate membrane filters ("ALBET" AC 045 47 BL) to determine the Zn content in the soil solution (immediately available Zn). We measured pH and E_h in incubation containers. The available Zn concentration and short-term available Zn concentration were then determined using DTPA-TEA and LMWOAs, respectively. The weight of the dry soil used was corrected with the previous moisture calculation.

The Zn concentrations in the different extracts were determined by flame atomic absorption spectrometry

with a Perkin-Elmer precisely AAS AAnalyst 700 (Perkin-Elmer, Waltham, MA, USA), which involved direct aspiration of the aqueous solution by an airacetylene flame. AA spectroscopy with a graphite furnace was used for low absorbance samples.

Statistical analyses were performed using Statgraphics Plus-5.1 software (Manugistic Inc., Rockville, MD, USA). Multiple comparisons of variables were performed using the LSD separations of means procedure. A probability level of $p \le 0.05$ was selected to establish statistical significance.

3. Results and Discussion

3.1. Incubation under 60% field capacity moisture conditions

The mean Zn concentrations during soil incubation under 60% field capacity moisture conditions are shown in Table 1. In Soil_{acid}, there were significant differences between the mean Zn concentrations over time (p<0.05) (Figure 1).

In this soil, we observed significant differences (p <0.0001) between the mean available Zn concentrations in soils receiving the different treatments. The residual effect of the natural chelate Zn-AML applied at a rate of 10 mg Zn/kg produced the highest available and short-term available Zn concentrations. The available Zn concentrations obtained for all of the Zn treatments were higher than those reported as critical for plants grown in acidic soils by Lindsay and Norwell (1978); the observed values were between 6.10 and 13.87 times greater than the critical level (Zn-PHP applied at a rate of 5 mg Zn/kg and Zn-AML applied at a rate of 10 mg Zn/kg, respectively). This finding was in agreement with the short-term available Zn concentration values, which ranged between 2.34 and 4.87 times the Zn concentration with Nil-Zn treatment (Zn-PHP applied at a rate of 5 mg Zn/kg and Zn-AML applied at a rate of 10 mg Zn/kg, respectively).

Source of variation	Soil _{acid}			Soil _{calc}		
	Available Zn (mg kg ⁻¹) ^(b)	$\begin{array}{l} Short-term\\ available \ Zn\\ (mg\ kg^{-1})\ ^{(c)} \end{array}$	Water-soluble Zn $(\mu g L^{-1})^{(d)}$	Available Zn (mg kg ⁻¹)	Short-term available Zn (µg kg ⁻¹)	Water-soluble Zn (µg L ⁻¹)
Incubation peri-	od (days)					
1	5.41 b	4.7 с	57.82 c	2.52 b	109.1 d	18.29 c
15	5.31 ab	4.53 bc	55.42 bc	2.13 a	96.68 c	15.43 b
45	5.3 ab	4.32 ab	52.86 b	2.06 a	72.74 b	14.14 a
75	5.22 a	4.22 a	49.43 a	1.99 a	60.42 a	13.71 a
Treatment						
Nil-Zn	1.81 a	1.38 a	31.0 a	0.81 a	44.0 a	9.8 a
Zn-AML-5	4.08 b	3.94 с	50.0 b	1.89 c	55.5 b	13.5 b
Zn-AML-10	8.32 d	6.72 e	62.3 cd	3.33 f	81.5 e	15.5 c
Zn-PHP-5	3.66 b	3.23 b	59.0 c	1.64 b	57.3 с	13.3 b
Zn-PHP-10	7.63 с	6.08 d	63.5 d	2.45 d	68.2 d	19.8 e
Zn-EDDS-5	4.33 b	3.84 с	52.3 b	2.00 с	134.8 f	17.5 d
Zn-EDDS- 10	7.34 с	5.91 d	59.2 c	3.11 e	152.0 g	18.5 de

Table 1. The mean Zn concentrations under 60% field capacity moisture conditions in both soil types were influenced by the incubation period (days) and the source of Zn fertilizer^(a)

^(a) Values were compared using the LSD multiple range test at the 0.05 level of probability. Homogeneous groups are denoted with the same letter. ^(b) Estimated by the DTPA-TEA method. ^(c) Estimated by the low-molecular-weight organic acids (LMWOAs) method. ^(d)A mixture containing 2.5 g of dry soil and 25 mL of deionized water was shaken for 30 min and centrifuged.

The Zn-EDDS treatments produced values similar to Zn-AML applied at a rate of 5 mg Zn/kg and to Zn-PHP applied at 10 mg Zn/kg. When this chelate was applied to the previous crop at the higher rate (10 mg Zn/kg), it produced an excessive concentration of available Zn and caused phytotoxicity in flax plants grown in the acidic soil of the previous crop. The residual effect of Zn-PHP applied at a rate of 10 mg Zn/kg produced the highest water-soluble Zn concentration. These Zn concentrations were between 1.61 and 2.05 times those associated with Nil-Zn treatment (Zn-AML applied at a rate of 5 mg Zn/kg and Zn-PHP applied at 10 mg Zn/kg, respectively). In Soil cale, there were significant differences between the mean available Zn concentrations over time (p < 0.05) (Figure 1). The mean short-term available Zn concentrations also differed over time (p < 0.0001), as

did the water-soluble Zn concentrations (p<0.001). In this soil, we observed significant differences between the mean available Zn concentrations in soil (p<0.0001) with the application of different treatments. The residual effect of the natural chelate Zn-AML, applied at the rate of 10 mg Zn/kg, produced the highest available concentration. The available Zn concentrations obtained for all of the Zn treatments were higher than the critical level for most plants grown in calcareous soils, with values reaching between 1.89 and 3.33 times the critical level (Zn-AML applied at a rate of 5 mg Zn/kg and Zn-AML applied at a rate of 10 mg Zn/kg, respectively). On the other hand, Zn-EDDS applied at both rates produced the highest short-term available Zn concentrations.

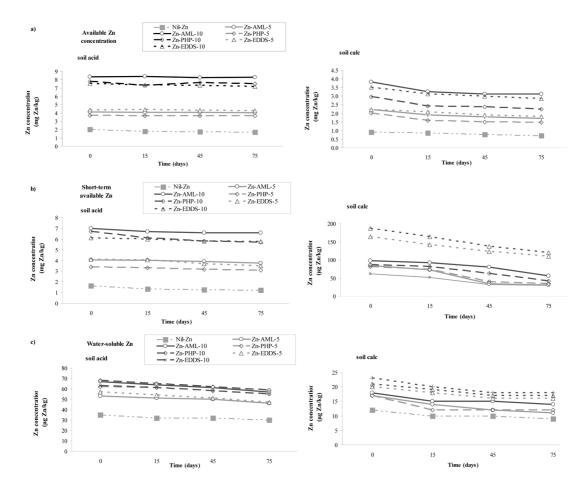


Figure 1. Time course of changes in the available (a), short-term available (b) and water-soluble (c) Zn concentrations in both soil types showing the residual effects of each treatment under 60% field capacity conditions.

These Zn concentrations were between 1.26 and 3.45 times greater than the Zn concentration with Nil-Zn treatment (Zn-AML applied at a rate of 5 mg Zn/kg and Zn-EDDS applied at a rate of 10 mg Zn/kg, respectively). The residual effect of Zn-PHP applied at a rate of 10 mg Zn/kg produced the highest water-soluble Zn concentration. The application of Zn-EDDS at both rates also produced high water-soluble Zn concentrations. Aminopolycarboxylate chelating agents (e.g., EDDS)

generally form complexes with high stability constants (log $K_{Zn-EDDS} = 13.4$), which causes high short-term availability of the micronutrient over time (Martell *et al.*, 2001; Tandy *et al.*, 2004; Nörtemann, 2005; Nowack *et al.*, 2006).

In Soil_{acid}, the mean Zn concentrations were 2.44 (available Zn), 52.43 (short-term available Zn) and 3.50 (water-soluble Zn) times greater than those in Soil_{calc}.

The characteristics of $Soil_{cale^3}$ which include its alkaline pH, high CaCO₃ concentration and high clay content, caused Zn immobilization due to the formation of hydroxides and carbonates in addition to the adsorption of Zn to the clay.

The differences between the Zn concentrations extracted by the different methods were due to the different extraction capacities of the reagents. The available Zn concentration method (DTPA-TEA) extracts water-soluble metal that is exchangeable, sorbed and organically associated, some of which is occluded in oxides and secondary clay minerals (Viets, 1962; Ure, 1995). However, the short-term available Zn (LMWOAs) method extracts the amount of metal responsible for short-term availability (Cieśliński *et al.*, 1998) and for most of the metal available to the plant. Water-soluble Zn is the most labile micronutrient fraction.

The most significant correlation between the different Zn concentrations obtained from the two soils revealed a significant positive correlation between short-term available Zn and water-soluble Zn (r = 0.96; p < 0.0001). We obtained the following regression equation:

Zn-(water-soluble) = 16.7 + 7.92 Zn-(short-term available) (R² = 91.81%)

Under 60% field capacity conditions, significant differences were noted between the experimental times for redox potential values (p < 0.001 in Soil_{acid}, p < 0.0001 in Soil_{acid}). In Soil_{acid}, we observed an increase in this parameter, with mean Eh values ranging between 488 mV at 1d and 598 mV at 75d. On the other hand, in Soil_{cale}, the mean Eh values ranged between 673 mV at 1d and 477 mV at 75d. Under these moisture conditions, we also observed significant differences in soil pH between experimental times (p < 0.05 in Soil_{acid}, p < 0.001 in Soil_{cale}). In Soil_{acid}, the mean soil pH values ranged between 6.33 at 1d and 6.07 at 75d. In Soil_{cale}, the mean pH values ranged between 7.84 at 1d and 8.30 at 75d. The parameter pH + pe [pe = E_h (mV) / 59.2], where pe is the negative logarithm of free electron

activity, provides a convenient single-term expression for defining the redox status of soil systems. Significant differences in this parameter were noted under 60% field capacity conditions (p < 0.001 and p < 0.0001 for Soil_{acid} and Soil_{cale}, respectively). In Soil_{acid}, pH + pe increased with time, whereas in Soil_{cale}, it decreased from 1 to 75d. According to Sparks (1996), the pH and pe values that were obtained for the two soils under 60% field capacity conditions would correspond to "oxic" or "normal" soils (pH + pe > 14).

3.2. Incubation under waterlogged moisture conditions

The mean Zn concentrations during soil incubation under waterlogged moisture conditions are shown in Table 2. In Soil_{acid}, there were significant differences between the mean Zn concentrations over time (p< 0.05 for available and short-term available Zn; p< 0.0001 for immediately available Zn) (Figure 2).

In Soil_{acid} we observed significant differences between the mean Zn concentrations (p < 0.0001) with the different treatments. The residual effect of the natural chelate Zn-AML when applied at the highest rate (10 mg Zn/kg) produced the greatest available, short-term available and immediately available Zn concentrations. The available Zn concentrations obtained for all of the Zn treatments were higher than those reported as critical for plants grown in acidic soils. These concentrations reached values that were between 3.73 and 11.05 times greater than the critical concentration (Zn-EDDS applied at a rate of 5 mg Zn/kg and Zn-AML applied at a rate of 10 mg Zn/kg, respectively). Short-term available and immediately available Zn concentrations were also high under treatments involving residual Zn. The short-term available Zn concentrations were between 2.12 and 5.44 times greater than the Nil-Zn concentration, and the immediately available Zn concentrations were between 2.58 and 6.00 times greater than the Nil-Zn concentration (Zn-PHP applied at s rate of 5 mg Zn/kg and Zn-AML applied at a rate of 10 mg Zn/kg in both cases).

Source of variation	Soil _{acid}			Soil _{calc}			
	Available Zn (mg kg ⁻¹) ^(b)	Short-term available Zn (mg kg ⁻¹) ^(c)	Immediately available Zn (mg L ⁻¹) ^(d)	Available Zn (mg kg ⁻¹)	Short-term available Zn (µg kg ⁻¹)	Immediately available Zn (µg L ⁻¹)	
Incubation perio	od (days)						
1	3.95 b	3.79 с	2.00 d	1.96 c	107 d	29.7 d	
15	3.74 ab	3.61 bc	1.77 с	1.86 bc	91.7 с	26.7 с	
45	3.59 a	3.54 ab	1.40 b	1.76 ab	58.7 b	20.7 b	
75	3.47 a	3.38 a	1.16 a	1.72 a	42.6 a	15.3 a	
Treatment							
Nil-Zn	1.36 a	1.09 a	0.43 a	0.64 a	25.8 a	17.5 a	
Zn-AML-5	3.21 c	2.96 с	1.37 с	1.45 b	46.8 b	19.8 b	
Zn-AML-10	6.63 f	5.93 e	2.58 e	2.73 е	67.5 c	28.8 d	
Zn-PHP-5	2.30 b	2.31 b	1.11 b	1.45 b	44.5 b	20.8 b	
Zn-PHP-10	4.84 d	4.62 d	1.99 d	2.08 d	60.3 c	21.3 b	
Zn-EDDS-5	2.24 b	3.25 с	1.50 c	1.60 c	130 d	25.5 с	
Zn-EDDS-10	5.26 e	4.90 d	2.12 d	2.85 f	150 e	28.3 d	

Table 2. The mean Zn concentrations under waterlogged conditions in both soil types, were influenced by the incubation period (days) and the source of Zn fertilizer^(a)

^(a) Values were compared using LSD multiple range test at the 0.05 level of probability. Homogeneous groups are denoted with the same letter. ^(b) Estimated by the DTPA-TEA method. ^(c) Estimated by the low-molecular-weight organic acids (LMWOAs) method. ^(d) Zn in the soil solution.

In Soil_{cale}, there were significant differences between the mean Zn concentrations over time (p<0.05, available Zn concentration; p<0.0001, short-term and immediately available Zn concentrations).

In Soil_{cale}, there were significant differences between the Zn concentrations associated with the different treatments (p< 0.0001). The residual effect of Zn-EDDS applied at the highest rate (10 mg Zn/kg) produced the highest available, short-term available and immediately available Zn concentrations. The residual effect of Zn-EDDS applied at a rate of 5 mg Zn/kg also produced high Zn concentrations that, in some cases, were even greater than those obtained with other natural chelates, such as Zn-PHP and Zn-AML applied at a rate of 10 mg Zn/kg. This characteristic of Zn-EDDS could be explained by

the high stability of the complex and, more specifically, by the relatively strong complexing ability of Zn, which allows the maintenance of high Zn concentrations even under waterlogged conditions. Zn concentrations, especially the concentrations of available and shortterm available Zn, showed major decreases when Zn-EDDS was applied at both doses (Figure 2). This result was in agreement with those reported by Tandy et al. (2006), who observed decreases in metal concentrations over time that were clearly coupled to the degradation of EDDS. Tandy et al. showed that the application of EDDS under temporarily anoxic conditions (24 hours) in three different saturated soils resulted in initially high soluble Zn concentrations that gradually decreased to only trace levels by day 21; after day 35, the concentration of the Zn-EDDS complex was extremely low.

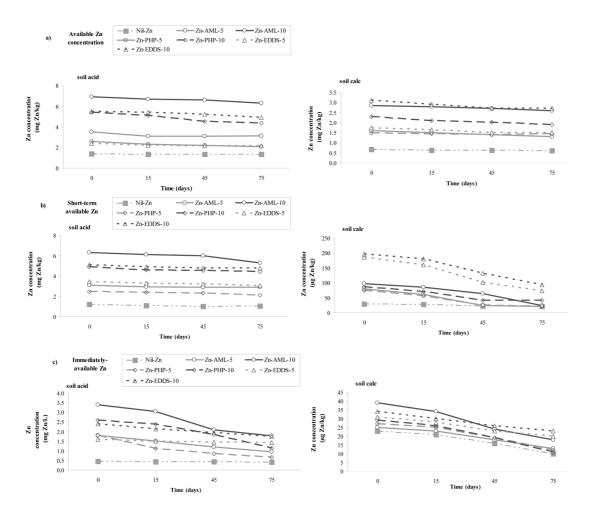


Figure 2. Time course of changes in available (a), short-term available (b) and immediately available (c) Zn concentrations in both soil types showing the residual effects of each treatment under waterlogged conditions.

In Soil_{acid}, the available, short-term and immediately available mean Zn concentrations were 1.96, 21.33 and 82.8 times higher, respectively, than those in Soil_{calc}. The alkaline pH most likely caused the Zn to be less soluble.

Immediately available Zn, or the Zn concentration in the soil solution, is the water-soluble form of Zn (ion, small

molecule or dissolved gas) that can rapidly move through protein transporter channels in plant and microbial cell membranes (Hedley, 2008). The correlation between the immediately available Zn and short-term available Zn concentrations in the two soils showed a significant negative correlation (r = -0.79; p < 0.001). The following regression equation was obtained:

Zn-(immediately available) = 20.19 - 4.29 Zn-(short-term available) (R² = 63.13%)

In both soils, the redox potential values measured under waterlogged conditions significantly decreased (p < 0.0001 and p < 0.05 for Soil_{acid} and Soil_{cale}, respectively) between 1 and 75d. In Soil_{acid}, these values ranged between 379 and 327 mV at 1and 75d, respectively. In Soil_{cale}, the mean values at 1 and 75d were 266 and 245 mV, respectively. The pH values did not significantly differ between 1 and 75d, with mean values ranging between 5.10 and 5.29 in Soil_{acid} and between 7.19 and 7.27 in Soil_{cale}. Under waterlogged conditions, the pH + pe parameter did not show any significant differences with time. According to Sparks (1996), the pH and pe values obtained for the two soils would correspond to "wet" or "seasonally saturated" soils.

We compared the available Zn concentrations in soil under different moisture conditions. The available Zn concentration under moisture conditions of 60% field capacity was compared with the sum of the available Zn and immediately available Zn concentrations under waterlogged conditions (considering bulk density). The percentages of recovered Zn under 60% field capacity conditions with respect to waterlogged conditions were 8.81% for Soil_{acid} and 19.51% for Soil_{cale}. We also compared the short-term available Zn under 60% field capacity conditions and the sum of the shortterm available Zn and the immediately available Zn concentrations under waterlogged conditions. The percentage of recovered Zn under waterlogged conditions with respect to 60% field capacity conditions reached 11.54% for Soil_{acid} and 11.87% for Soil_{cale}. These results could be explained by the fact that under waterlogged conditions, the redox potential decreases and Fe (III) and Mn (III, IV) oxides are dissolved because of the reduction of Fe and Mn (Millaleo et al.,2010). This effect may have been caused by the Zn redistribution and the increase in the Zn concentration in the most labile fractions (in the form of immediately available and short-term available Zn).

4. Conclusions

Moisture conditions influence the concentrations of available, short-term available, immediately available and water-soluble Zn in soils. These concentrations also depend on the length of the experiment, the soil type and the Zn-chelate used. Over time, changes were observed in the concentrations of residual Zn from natural chelates in both soils and under both moisture conditions. Available, short-term available and immediately available Zn concentrations decreased from 1 to 75 d. Under waterlogged conditions, shortterm available Zn and immediately available Zn concentrations were increased compared with their concentrations under 60% field capacity conditions. In general, the residual effects of Zn-AML applied at a rate of 10 mg Zn/kg in Soilacid and Zn-EDDS applied in Soil produced the highest Zn concentrations under both moisture conditions. The Zn concentrations in soils that received Zn treatments were sufficient to meet the needs of most crops, particularly in the acidic soil, where Zn concentrations reached their highest values.

Acknowledgements

Financial support for this study was provided by Spain DGI (MEC; project: AGL2009-12741).

References

- Almendros, P., Gonzalez, D., Alvarez, J.M. 2011. Residual effect of natural and synthetic zinc chelates on zinc in a soil solution of a waterlogged acidic soil. Evolution of the pH and redox potential. EGU General Assembly, Viena.
- Alvarez, J.M. 2010. Influence of soil type and natural Zn chelates on flax response, tensile properties and soil Zn availability. Plant and Soil. 328, 217–233.

- Alvarez, J.M., Rico, M.I. 2003. Effect of zinc complexes on the distribution of zinc in calcareous soil and zinc uptake by maize. Journal of Agricultural and Food Chemistry. 51, 5760–5767.
- Armour, J.D., Ritchie, G.S.P., Robson, A.D. 1989. Changes with time in the availability of soil applied zinc to navy beans and in the chemical extraction of zinc from soils. Australian Journal of Soil Research. 27, 699–710.
- Barrow, N.J. 1986. Test a mechanistic model. II. The effects of time and temperature on the reaction of zinc with a soil. Soil Science. 37, 277–286.
- Cieśliński, G., Van Rees, K.C.J., Szmigielska, A.M., Krishnamurti, G.S.R., Huang, P.M. 1998. Lowmolecular-weight organic acids in rhizosphere soils of durum wheat and their effect on cadmium bioaccumulation Plant and Soil. 203, 109–117.
- Feng, M.H., Shan, X.Q., Zhang, S.Z., Wen, B. 2005. Comparison of a rhizosphere-based method with other one-step extraction methods for assessing the bioavailability of soil metals to wheat. Chemosphere. 59, 939–949.
- Hedley, M.J. 2008. Techniques for assessing nutrient bioavailability in soils: current and future issues. Developments in Soil Science. 32, 283-327.
- Liang, J., Steward, J.W.B., Karamanos, R.E. 1990. Distribution of zinc fractions in preirie soils. Canadian Journal of Soil Science. 70, 335–342.
- Lindsay, W.L., Norvell, W.A. 1978. Development of a DTPA soil test for zinc, iron, manganese, and copper. Soil Science Society of America Journal. 42, 421–428.
- Lopez-Valdivia, L.M., Fernández, M.D., Obrador, A., Alvarez, J.M. 2002. Zinc transformations in acidic soil and zinc efficiency on maize by adding six organic zinc complexes. Journal of Agricultural and Food Chemistry. 50, 1455–1460.

- Ma, Y.B., Uren, N.C. 2006. Effect of aging on the availability of zinc added to a calcareous clay soil. Nutrient Cycling in Agroecosystems. 76, 11–18.
- Martell, A.E., Smith, R.M., Motekaitis, R.J. 2001. NIST critically selected stability constants of metal complexes database.
- Martinez, C.E., McBride, M.B. 2000. Aging of coprecipitated Cu in alumina: changes in structural location, chemical form, and solubility. Geochimica et Cosmochimica Acta. 64, 1729–1736.
- McBride, M.B. 1994. 'Environmental chemistry of soil.' (Oxford University Press: NY)
- Millaleo, R., Reyes-Diaz, M., Ivanov, A.G., Mora, M.L., Alberdi, M. 2010. Manganese as essential and toxic elements for plants: transport, accumulation and resistance mechanisms. Journal of soil science and plant nutrition. 10, 470–481.
- Nörtemann, B. 2005. Biodegradation of chelating agents: EDTA, DTPA, PDTA, NTA, and EDDS. In: B. Nowack, J.M. VanBriesen (eds). Biogeochemistry of chelating agents. ACS Symposium Series 910. Journal of the American Chemistry Society: Washington DC, USA. pp: 150-170.
- Nowack, B., Shulin, R., Robinson, B.H. 2006. Critical assessment of chelant enhanced metal phytoextraction. Environmental Science and Technology. 40, 5225–5232.
- Obrador, A., Alvarez, J.M., Fernandez, M.D., Lopez-Valdivia, L.M. 2002. Changes with time of zinc forms in an acid, a neutral, and a calcareous soil amended with three organic zinc complexes. Australian Journal of Soil Research. 40, 137–148.
- Payne, G.G., Martens, D.C., Winarko, C., Perera, N.F. 1988. Form and availability of copper and zinc following long-term copper sulfate and zinc sulfate applications Journal of Environmental Quality. 17, 707–711.

- Shuman, L.M. 1991. Chemical forms of micronutrients in soils. In: J.J. Mortvedt, F.R. Cox, L.M. Shuman, R.M. Welch (eds). Micronutrients in agriculture. Soil Science Society of America: Madison, WI. pp. 113–144.
- Sparks, D.L., Bartels, J., Bigham, J. 1996. Methods of soil analysis. Part 3, chemical methods. Soil Science Society of America: Madison, WI.
- Tandy, S., Bossart, K., Mueller, R., Ritschel, J., Hauser, L., Schulin, R., Nowack, B. 2004. Extraction of heavy metals from soils using biodegradable chelating agents. Environmental Science and Technology. 38, 937–944.
- Tandy, S., Ammann, A., Schulin, R., Nowack, B. 2006. Biodegradation and speciation of residual SS-ethylenediaminedisuccinic acid (EDDS) in soil solution left after soil washing. Journal of Environment and Pollution. 142, 191–199.

- Tiller, K.G., Honeysett, J.L., De Vries, M.P.C. 1972. Soil zinc and its uptake by plants. I. Isotopic exchange equilibria and the application of tracer techniques. Australian Journal of Soil Research. 10, 151–164.
- Ure, A.M. 1995. Methods of analysis for heavy metals in soils. In B.J. Alloway (ed). Heavy metals in soils. Blackie Academic and Professional: Glasgow, UK. pp. 58–102.
- Viets, F.G. 1962. Micronutrient availability, chemistry and availability of micronutrients in soils. Journal of Agricultural and Food Chemistry. 10, 174–178.
- Violante, A., Cozzolino, V., Perelomov, L., Caporale, A.G., Pigna, M. 2010. Mobility and bioavailability of heavy metals and metalloids in soil environments. Journal of Soil Science and Plant Nutrition. 3, 268–292.