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Original Article

The Effect of EDTA on the Ability to Absorb Different Concentrations of Nickel, Cadmium, and Lead in Soil by Corn Plants

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Abstract

Background & Aims: The use of resistant and widely used species such as corn in agriculture and industry can be an effective solution for the bioremediation of soil pollutants, including heavy elements. The current research aimed to investigate the effect of EDTA on the ability to absorb different concentrations of heavy metals in the soil of corn plants in 2019.

Materials and Methods: This research was conducted as a three-factor pilot design at concentrations of 0, 50, and 100 mg/kg in the greenhouse. The samples were analyzed using an atomic absorption device. The laboratory pilot design was based on Taguchi's algorithm. Finally, transfer factor (TF), bioconcentration factor (BCF), and bioaccumulation coefficient (BAC) were calculated.

Results: The results showed that different organs (root, stem, and leaf) show different levels of bioaccumulation under the influence of variable factors in different concentrations of heavy elements (nickel, cadmium, and lead). Also, there is a significant difference between the measured amounts of heavy elements in different organs of the corn plant ($P \leq 0.05$). The average TF levels for elements at concentrations of 50 and 100 mg/kg were 0.79 and 1.66 for nickel, 0.82 and 0.78 for cadmium and 1.361, 1.378, and 1.387 for lead. Based on the results, with the increase in nickel concentration, the absorption level increased, and with the increase in cadmium concentration, the absorption level decreased.

Conclusion: The results of this research showed that it is possible to use EDTA to increase the efficiency of corn plants in absorbing heavy elements of nickel, cadmium, and lead.

Keywords: Bioaccumulation, Corn, Transfer factor, Heavy metals, EDTA

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1. Introduction

Today, there is a need to develop biological methods of soil amendment which are economical in terms of cost and eliminate pollution without reducing soil fertility [1]. Much research has been conducted in the field of biostimulation and bioavailability of heavy metals [2,3]. Recently, EDTA has been widely used to reduce the toxicity of heavy metals [4,5]. EDTA is a polyamine carboxylic acid with the chemical formula CH2N(CH2CO2H)2, which is mostly used as its disodium salt. This compound forms stable complexes with a wide range of metals, so it has a high ability to release metal ions [6]. Heavy elements are among the most important environmental pollutants, which have been highly considered in the last few decades [7]. The accumulation of elements in the soil, especially in agricultural lands, is a gradual thing, and the concentration of heavy elements can reach a level that threatens human food security [8].

Every year, thousands of tons of these elements, which are caused by urban, industrial, and agricultural activities, enter the soil [9]. Heavy metals have a very destructive effect on the environment and living organisms, including humans [9]. Their greatest effect on humans is related to neurological disorders [10,11]. These metals also replace other minerals needed by the body [12]. Also, the accumulation of heavy metals in the soil and water has received much attention due to their toxicity and the dangers they pose to humans and the environment [13]. Excessive accumulation of heavy metals in the soil destroys the ecosystem and affects soil properties such as pH, electrical conductivity, cation exchange capacity, and microbial and biological activities [14,15]. Many methods have been used to reduce soil pollution from heavy metals, which are classified into physical, chemical, and biological groups or a combination of these [16,17]. One of the effective compounds in the absorption of heavy metals is the use of EDTA [18]. In various research, the effect of EDTA in increasing the absorption of heavy metals has been investigated.

Dong et al stated that biological stimulation with EDTA and electrokinetic results in increasing the absorption of heavy elements by plants [19]. Cheng et al also stated that soil washing using organic EDTA has led to the removal of significant amounts of nickel, copper, and zinc

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in industrial soils [20]. Zhang et al showed the effect of EDTA on increasing the absorption of heavy elements by bamboo by more than 38% [21]. Hamidpour et al stated that growth-promoting bacteria, along with EDTA, led to an increase in the absorption of heavy elements by biomass [22].

But the level of effectiveness of EDTA in absorbing heavy elements is different based on the type of plant and different elements [23-25]. The methods used to estimate the absorption level of heavy elements by plants are transfer factor (TF), bioconcentration factor (BCF), and bioaccumulation coefficient (BAC) indices [26]. In these indices, the ratio of the amount of heavy elements in the soil to the amount of elements absorbed in the plant organs is calculated [27]. Corn is one of the most widely used crops in the southern regions of Iran [28]. Maize, with the scientific name Zea mays, from the cereal category and the large wheat family, is a monocotyledonous plant, and its edible part is the female inflorescence (spike) that produces seeds or the same fruit [29].

Nickel, cadmium, and lead are heavy elements that are mainly rooted in industrial activities [30]. The results of various studies have shown that the level of soil contamination with heavy metals in Khuzestan province is more than in other regions of Iran. The results of Sahipour and Sabzalipour's study showed that the intensity of soil contamination with heavy elements lead, zinc, nickel, and chromium is at the "contaminated" level [31]. Shahidi Kaviani and Paykanpour Fard also found that the contamination of the soils of the Ahvaz oil field, especially in the vicinity of oil and gas wells, with heavy metals cadmium and copper is higher than the world average [32]. Considering the existence of many industries such as oil, gas, and steel in the southwest of Iran, as well as the areas under corn cultivation, in this research (in 2019), the effect of EDTA on the ability to absorb different concentrations of heavy metals in the soil by corn plants has been studied.

2. Material and Methods

This research is based on the practical purpose and descriptive data collection, which was conducted to investigate the ability of grain corn species to absorb heavy metals (nickel, cadmium, and lead) from soil using EDTA in 2019. The research is a three-factor factorial experiment in the form of a completely randomized design with three replications in the greenhouse. Soil samples were collected from the surrounding areas of Ahvaz city at a depth of 0-60 cm. After collection, the samples were dried and passed through a 2 mm sieve before use [33]. The measurement of nickel and cadmium elements was also done by an Agilent 240 AA furnace atomic absorption device. In the study of Guo et al, EDTA at the level of 12 mmol was determined as the optimal limit of bioavailability of heavy metals from the soil, so the amount of EDTA at the level of 12 mmol/kg was used. Also, to investigate the effect of pollution level on the absorption of heavy metals, nickel concentration (zero, 50, 100 mg nickel/kg soil as nickel sulfate) and cadmium concentration in three levels (zero, 50, 100 mg cadmium/kg soil as cadmium nitrate) and lead concentration in three levels (zero, 50, 100 mg lead/kg soil as lead nitrate) were used [23]. For preparation, 3 kg of prepared dry soil was transferred to the pots and uniformly contaminated with heavy metal salt treatments of cadmium, nickel, and lead. After about a month, EDTA treatments were added to the pots. The pots were kept for 45 days to create balance in the soil [34]. Then corn seeds were planted in each pot in such a way that three corn plants were harvested from each pot. For 12 weeks, daily, pots containing corn plants were repeatedly visited and irrigated with distilled water, low-quality plants were thinned in terms of growth, and the edges of the pots were also removed for the uniformity of the absorption conditions of the plants. Also, to achieve uniform environmental growth conditions, the pots were changed every week. After about 100 days, when the plant achieved proper growth, the plants were harvested, and after washing the roots, three tissues of the root, stem, and leaf were separated until the absorption of heavy metals was done by the atomic absorption device. The method (Jackson 1958) was used to digest plant samples [35].

After determining the amount of extractable heavy metals in corn plant and soil samples, we measured the TF index (transfer factor: ratio of metal concentration in aerial parts of plants to metal concentration in roots), BCF index (Bioconcentration: ratio of metal concentration in plant roots to metal concentration in soil), and BAC index (biological accumulation coefficient: ratio of metal concentration in aerial parts of plants to metal concentration in soil).

2.1. Transfer factor

This factor is used to evaluate the plant's ability to transfer metal from the root to the stem and is calculated by dividing the concentration of the element in the aerial part by the concentration of the element in the root [36].
Equation 1: Transfer factor calculation

$$TF = C(Cd,Ni),sh / C(Cd,Ni),r \qquad \text{Eq. (1)}$$

2.2. Bioconcentration factor index

One of the important factors used to measure the concentration of heavy elements in plant samples is the bioconcentration factor, and it is calculated by dividing the concentration of the element in the aerial part by the concentration of the element in the soil [36].

Equation 2: Calculation of biological inhibition index

$$BCF = C(Cd,Ni),r / C(Cd,Ni),s \qquad \text{Eq. (2)}$$

2.3. Bioaccumulation coefficient

This index shows the ratio of metal accumulation in plant roots to metal accumulation in soil. (accumulation in roots to accumulation in soil) [37].

Equation 3: Calculation of bioaccumulation coefficient

$$BAC = C(Cd,Ni),Sh/C(C\ ,Ni),s \qquad \text{Eq. (3)}$$

3. Results

The results of measuring the heavy elements of nickel-cadmium in soil, root, stem, and leaf samples of corn plants at concentrations of 0, 50, and 100 mg/kg and their standard deviation are presented in Table 1.

The results showed that the amounts of nickel, cadmium, and lead in the factor variable and biomass in unpolluted pots were lower than the samples contaminated with concentrations of 50 and 100 mg/kg (Figures 1, 2, and 3). Also, the amounts of nickel, cadmium, and lead elements in the soil samples were lower than in the corn biomass samples (root, stem, and leaf) (Figure 2). The difference between the amounts of heavy elements nickel and cadmium in soil and plant samples was statistically proven with 95% confidence. The highest amount of nickel in the biomass of root samples at a concentration of 100 mg/kg was measured as 5.79 mg/kg, and the lowest amount was measured as 1.09 mg/kg in control pots (without heavy elements). Also, the highest amount of lead in soil samples was 1.56 mg/kg, and in corn leaf organ was 2.38 mg/kg.

The results of one-way analysis of variance are presented in Table 2. According to these results, there is a significant difference between the measured amounts of nickel, cadmium, and lead in soil and corn plant organs (root, stem, and leaf) ($P < 0.05$). Also, a significant difference was observed between the amounts of cadmium in concentrations of 0, 50, and 100 mg/kg with 99% confidence ($P < 0.05$).

Table 1. The results of measuring the samples

Samples	Statistics	Pb (mg/kg)			Cd (mg/kg)			Ni (mg/kg)		
		100	50	0	100	50	0	100	50	0
Soil Samples	Mean	1.56	1.49	1.37	0.44	0.36	0.29	1.51	1.22	1.09
	SD	0.042	0.04	0.061	0.055	0.026	0.023	0.1345	0.085	0.11
Corn root samples	Mean	1.72	1.42	0.021	0.58	0.46	0.16	5.79	5.39	2.48
	SD	0.037	0.04	0.021	0.083	0.042	0.096	0.3711	0.315	0.059
Corn stalk samples	Mean	1.24	1.03	0.94	0.46	0.39	0.1 >	4.98	4.26	2.08
	SD	0.072	0.03	0.011	0.061	0.082	0	0.1124	0.286	0.15

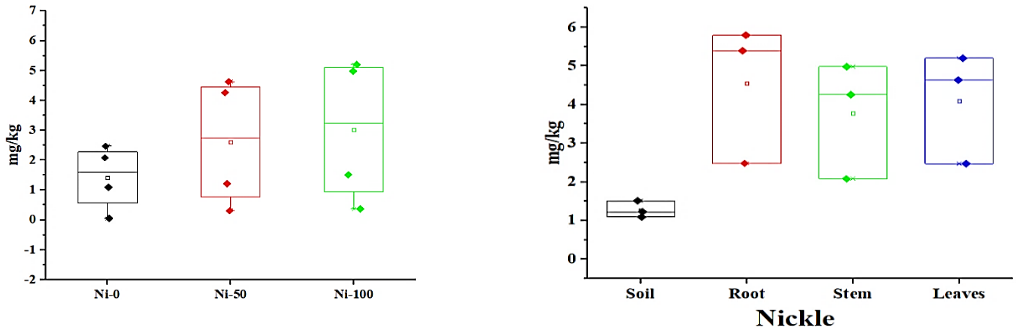

Figure 1. Comparison of measured amounts of nickel in samples (left side: soil samples - right side: corn plant samples)

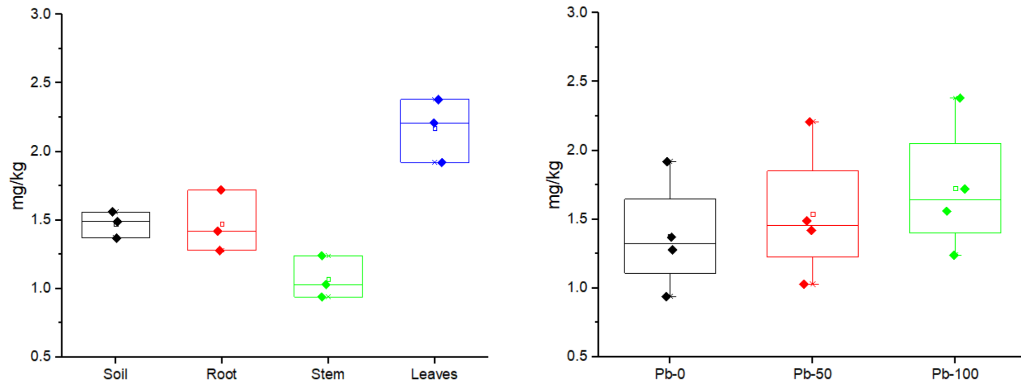

Figure 2. Comparison of measured lead values in samples (left side: soil samples - right side: corn plant samples)

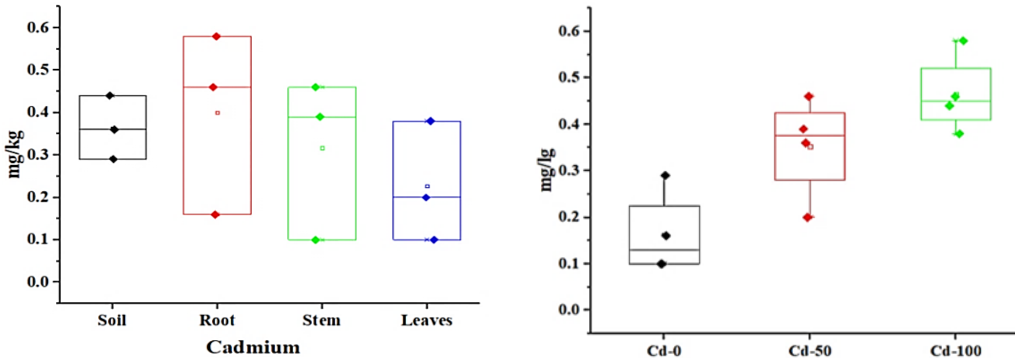

Figure 3. Comparison of measured amounts of cadmium in samples (left side: soil samples - right side: corn plant samples)

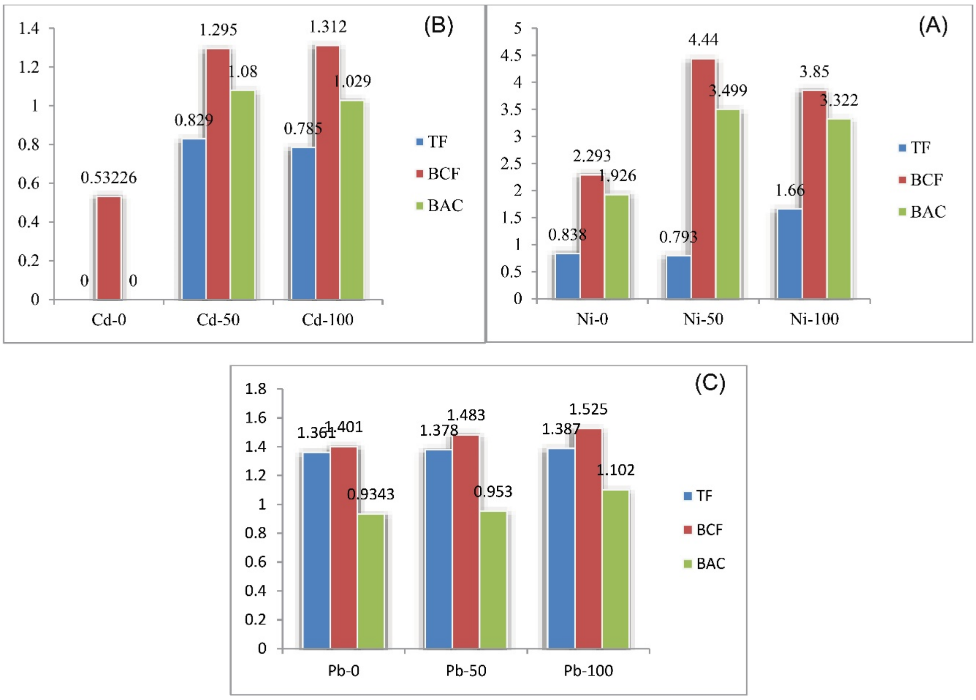

Figure 4. Comparison of absorption of heavy elements based on TF, BCF, and BAC indices in different concentrations of heavy element pollution (A: Cadmium-B: Nickel-C: Lead)

Table 2. The results of the one-way analysis of variance for the studied variables

Variable	Mean square	F	*P* value
Nickel (between concentrations)	5.406	1.776	0.173
Nickel (between soil and plant organs)	8.567	2.974	0.033*
Cadmium (between concentrations)	9.097	3.262	0.002*
Cadmium (between soil and plant organs)	6.032	2.046	0.04*
Lead (between concentrations)	5.85	1.876	0.43
Lead (between soil and plant organs)	9.21	2.31	0.01*

* Significance Relation

3.1. Calculate absorption factor

The results of the calculation of TF, BCF, and BAC indices for the analysis of the absorption of heavy elements by the organs of the corn plant in different concentrations are presented in Table 3. In studies to estimate the efficiency of plants in absorbing heavy elements, the proportion of the element in the soil to its proportion in the plant organ is important.

The results of the analysis of nickel element changes showed that with the increase of soil pollution load to the level of 50 mg/kg, the absorption level of heavy elements by leaf and root organs increased. The average BCF index was 4.44, and BAC was 3.499 (Figure 4). But with the increase of pollution level to 100 mg/kg of nickel, the absorption level of heavy elements by these corn plant organs has decreased. The same situation has happened in the element cadmium. But in the lead element, with the increase of pollution concentration to 100 mg/kg, the absorption level of heavy metals by corn organs has also increased in a limited way. The most accumulation of lead was done by the leaf organ. To check the correlation between heavy metal concentration and indicators, Pearson's correlation test was used (Table 4). Its results showed that there is a significant correlation between the

Table 3. The results of calculating TF, BAC, and BCF indices

Index	Nickel 0	Nickel 50	Nickel 100	Cadmium 0	Cadmium 50	Cadmium 100	Lead 0	Lead 50	Lead 100
TF (index)	0.838	0.793	1.66	-	0.829	0.785	1.361	1.378	1.387
BCF (index)	2.293	4.44	3.85	0.53226	1.295	1.312	1.401	1.483	1.525
BAC (index)	1.926	3.499	3.322	-	1.08	1.029	0.9343	0.953	1.102

Table 4. The results of Pearson correlation analysis between the average concentration of heavy metals and TF, BCF, and BAC indices

		TF	BCF	BAC
The average concentration of heavy metals	The correlation coefficient	0.515	0.948 **	0.937**
	meaningful	0.156	0.000	0.000

** Significance Correlation

concentration of heavy metals in the organs of the corn plant with BCF and BAC indices at the level of 0.01.

These results show that, in general, the highest level of biological accumulation of heavy elements nickel, lead, and cadmium was related to the organ of the corn leaf. Also, considering that the average value of the indicators is more than 1, it can be concluded that corn is a favorable accumulator of nickel, cadmium, and lead elements, but the highest level of biological accumulation is related to nickel elements.

4. Discussion

The use of plants is one of the most effective biological methods to deal with pollution, such as heavy elements [38]. One of the consequences of human activities in the environment is soil pollution, which has also attracted the attention of natural science researchers [39]. The presence of heavy elements in the soil (due to their biological accumulation) is considered a serious threat to the health of humans and other animals. Therefore, it seems necessary to use methods to reduce the level of heavy elements in the soil. Research has confirmed the optimal efficiency of biological methods, such as bioremediation, to deal with heavy element pollution [40,41]. The use of resistant and widely used species in agriculture and industry can be an effective solution for absorbing soil pollutants, including heavy elements. According to the morphological characteristics, such as very long roots (about 2 m), the corn species can be a good option for removing heavy elements, which is investigated in the current research.

The results of other studies, such as Saifullah et al, Jelusic et al, and Bloem et al, have shown that EDTA compounds with metals can increase the solubility and availability of metals in soils [42-44]. When EDTA is used in soils without creating a strong acidic culture medium, most of the metals are dissolved and available for green extraction [45]. The results showed that the most lead absorption was related to the leaf organ in corn species. The results of the study by Nazir et al and Shahid et al also showed that the most lead absorption was done by the leaves of the orchid and *Gluconobacter potus* species [46,47]. In another research, Kumar et al stated that due to the terrestrial origin of cadmium and nickel elements, the amounts of these metals in the roots of trees in black spur forests were more than in other organs [48]. The results of the present research showed that there is a significant difference between the measured amounts of nickel at a concentration of 0 mg/kg with 50 and 100 mg/kg of soil samples with roots, stems, and leaves of corn plants ($P \leq 0.05$). As a result, under the influence of 2% EDTA, the bioaccumulation of nickel by the organs of the corn plant has been at a favorable level. There is a significant difference between the measured amounts of cadmium at a concentration of 0 mg/kg with a concentration of 100 mg/kg in the soil and root and stem samples of corn plants ($P \leq 0.05$). But this difference with corn plant leaves was not significant ($P \geq 0.05$). These results show that EDTA had a favorable effect on the bioaccumulation of cadmium in corn plant stems and roots at a concentration of 100 mg/kg.

5. Conclusion

The results of this research and its comparison with other research show that EDTA can be used to increase the efficiency of corn plants in absorbing heavy elements of nickel and cadmium. Also, the absorption of nickel and cadmium by the roots and lead by the leaves of corn species has been higher. Due to the existence of many industrial areas in the south and southwest of Iran, this method can be used for bioremediation.

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Competing Interests

The authors declare that there is no conflict of interest regarding the publication of this manuscript. Furthermore, the ethical issues have been completely observed by the authors including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and/or submission, and redundancy.

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