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Phytoextraction of Chromium from contaminated soil by *Zea mays* as influenced by chelating agents

Ramprakash[1*], Sachin Kumari [2], Sushil Kumari[3], A. Sangwan[1] and Anoop Singh[1]

1. Department of Soil Science, CCS Haryana Agricultural University, Hisar-125004, India
2. Department of Agriculture Chemistry, CCS Haryana Agricultural University, Hisar-125004
3. Department of Agroforestry, CCS Haryana Agricultural University, Hisar-125004, India

*Corresponding author e-mail: ramsansanwal@gmail.com

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Abstract

A pot experiment was conducted to evaluate the effect of amendments on phytoextraction of Cr by *Zea mays* using CDTA, Citric acid, DTPA, NTA (at 10 mmol kg^{-1} soil) and FYM (3%). Dry matter yield of roots and shoots of *Zea mays* increased due to application of FYM and CDTA whereas reverse trend was observed in NTA, Citric acid and DTPA treated soils. Addition of sewage sludge (3% on dry weight basis) was found beneficial in improving the plant growth. Chelating agents enhanced the Cr uptake by both roots and shoots. Significantly higher values of Cr uptake by roots and shoots were observed from amended as compared to sewage sludge unamended soil. Application of CDTA was found more effective in enhancing the Cr uptake by roots and shoots of *Zea mays* than any other chelating agents at both the growth stages. Hence, marginally Cr contaminated soil may be remediated by adding chelating agents.

Key words: Chelating agents, Chromium, Phytoextraction, Zea *mays*

Abbreviations: CA: Citric acid, **CDTA:** Cyclohexane diamino tetra acetic acid, **DTPA:** Diethylene triamine penta acetic acid, **DAS:** Days After sowing, **FYM:** Farm yard manure, **NTA:** Nitrilotriacetic acid and **SS:** Sewage sludge

Introduction

Contamination of agricultural land with heavy metals is increasing due to industrilization, urbanization and various anthropogenic activities, continued use of treated and untreated waste water, sewage sludge, (SS) manures and excessive use of fertilizer and pesticide. Phytoremediation of heavy metal contaminated soils is an approach with a motive to extract excessive metals from marginally heavy metal contaminated soils through plant uptake. *Zea mays* have been reported to produce high biomass and accumulate significant amount of heavy metals in their tissues when induced through the addition of chelating agents (Blaylock *et al*., 1997). Use of sewage sludge and FYM as source of organic matter is a common practice. Hence, higher extractability of Cr due to chelating agents may lead to their excessive accumulation in plants. Phytoremediation is emerging as a new technol-ogy that uses plants for cleaning or decreasing the toxicity of heavy metal contamination soils and waste waters (McGrath and Zhao, 2003). The development of Phytoremediation is being driven primarily by the high cost of many other soil re-mediation methods, as well as a desire to use a 'green', sustainable process (Pulfore and Watson, 2003). However, the reme-diation efficiency depends on two sticking points: the high concentration of metal in plant, especially in aerial parts, and a relatively large aboveground biomass. Chelate-induced phytoextraction is based on the fact that the application of chelating agents to the soil markedly increases metal accumulated by plants (Garbisu and Alkorta, 2001, Alkorta *et al.,* 2004).

Materials and Methods

The experiment area is located at $29^0$10' North latitude and $75^0$46'east longitude at an elevation of 215.2m above mean sea level. The region has a semi-arid climate with severe cold winter and hot dry summer with annual rainfall of about 400mm. Surface (0-15 cm) sandy loam soil (*Typic Ustochrept*) sample were collected, air–dried, homogenized and sieved using 2mm sieve. The bulk sewage sludge sample collected from Sewer treatment plant from industrial area and well decomposed farmyard manure (FYM) from the manure pit of dairy farm and transferred to 72 polyethylene lined earthen pots (25 inch diameter). Physio-chemical characteristics of the soil, FYM and Sewage sludge are given in Table 1.

Table 1. Physico-chemical characteristics of the experimental soil, Sewage sludge and FYM

Properties		Content		
		Soil	Sewage sludge	FYM
Mechanical composition				
	(a) Sand (%)	69.70	-	-
	(b) Silt (%)	16.50	-	-
	(c) Clay (%)	13.80	-	-
	Sandy loam		-	-
Textural class		8.10	7.20	-
pH (1:2)		0.50	2.10	-
$EC_{1:2}$ (dS m^{-1})		0.32	12.20	27.80
Organic carbon (%)		11.80	-	-
CEC [Cmol(P^+) kg^{-1}]		0.40	0.25	-
$CaCO_3$ (%)				
Total Nutrients (%)				
(a) Nitrogen		0.09	1.29	1.18
(b) Phosphorus		0.01	0.41	0.70
(c) Potassium		0.10	0.73	2.50
Total metals (mg kg^{-1})				
Cr		0.12	7.2	
Pb		0.98	64.2	

The processed soil sample was used for laboratory and screen house studies. Two soil treatments, *viz.*, Cr at 20 mg kg^{-1} soil and Cr spiked (at 667 mg Cr kg^{-1} sludge) sewage sludge added at 3% on dry weight basis, One sixth portion of above treated soil sample was treated with FYM @ 3% on dry weight basis. Six chelating agents, *viz.* Control (Cr_{20}), Nitrilo triacetic acid (NTA), Diethylene triamine-penta acetic acid (DTPA), Cyclohexane diamino tetra acetic acid (CDTA), Citric acid (CA) and FYM were used. The treated soil was filled separately in 72 polyethylene lined earthen pots (25 inch diameter) each at 5 kg soil per pot. A basal dose of N, P, K, Mn, Fe, Zn and Cu at 50, 50, 60, 10, 10, 5 and 5 mg kg^{-1} soil respectively was applied in solution form in each pot through their analytical grade salts. After addition of nutrient solutions, pots were wetted with deionised water to field capacity moisture content, and kept for equilibration for one week and dried to workable moisture content. The contents of each pot were then taken out, mixed thoroughly, refilled and incubated for ten days at near field capacity moisture content and process of mixing was repeated again. Soil samples were drawn from each pot before sowing.

Ten healthy seeds of *Zea mays* were sown in each pot. After emergence of seedlings, only four plants per pot were allowed to grow. Second dose of nitrogen was applied at 25 mg N kg^{-1} soil in solution form 30 days after sowing (DAS). The pots were irrigated with deionised water as and when required.

Chelating agents were applied at 10 mmol kg^{-1} soil (1 mmol daily for 10 days in 10 split doses) at 40 DAS. Ten days after application of chelating agents half of the Maize crop (*i.e.* 3 replications out of 6) were harvested and remaining 3 replications of each treatment where harvested after 80 DAS. In order to determine Cr in root and shoot sample, wet digestion was carried out in a diacid mixture of nitric acid and perchloric acid in 4:1 ratio. Chromium in the digest was determined by using atomic absorption spectrophotometer (GBC-932 plus).

All treatments were replicated six times (2x6x6=72) with two Soils (Sewage Sludge unamended and amended) and six amendments (control, four chelating agents and FYM). Statistical analysis were done using factorial completely randomized design.

Results and Discussion

Dry matter yield of roots and shoots: Dry matter yield of roots and shoots of *Zea mays* was influenced differentially by chelating agents (Table 2). The mean dry matter yield of roots varied from 1.76 to 4.90 g pot^{-1}. Maximum mean dry matter yield of roots was obtained in Cr_{20}+ FYM and the least in Cr_{20}+NTA treated soil. Comparatively better growth of *Zea mays* plants was observed in soil in which sewage sludge was applied. Dry matter yield of roots increased due to the addition of FYM both in sewage amended and unamended soils at both the growth stages. Root development was drastically reduced in pots to which NTA was applied. Application of

CDTA, Citric acid, DTPA and NTA reduced the root growth but addition of FYM caused significant increase at both the growth stages. Beneficial effect of FYM on root growth might have been due to the complexation of Cr avoiding direct injury of Cr to emerging roots. It also clear that among the chelating agents, NTA was most effective in decreasing the dry matter yield of roots probably due to improved availability of Cr in the rhizosphere (Kulli *et al.*, 1999; Robinson *et al.*, 2000; Mishra, 2004). Mean dry matter yield of shoots ranged from 13.40 to 34.70 g pot^{-1} (Table 2), with maximum in Cr_{20}+ CDTA and least in Cr_{20}+NTA treated pots. Dry matter yield of shoots decreased in citric acid, DTPA, and NTA treated pots as compared to control. The mean dry matter yield of shoots obtained at first stage of growth was 15.84 and 18.03 g pot^{-1} in sewage sludge unamended and amended soils, respectively.

Chromium uptake by roots and shoots

Application of chelating agents significantly increased Cr uptake by roots and shoot at both the growth stages (Table 3). The mean Cr uptake by roots ranged from 407.89 to 563.38 µg pot^{-1}, being significantly higher in plants grown on Cr_{20}+FYM treated soil and least in control. Application of CDTA increased mean Cr uptake by 33.08% as compared to control. The mean uptake of Cr by roots was 288.04 and 290.87 µg pot^{-1} in sewage sludge unamended and amended soils at first stage of growth while the corresponding values were 524.89 and 628.31 µg pot^{-1} at second stage of growth respectively.

Application of CDTA increased the Cr uptake by *Zea mays* roots at first stage by 37.32 and 44.40% in sewage sludge unamended and amended soils, respectively as compared to control in which chelating agents were not applied. Significantly higher uptake of Cr was observed in roots of *Zea mays* from amended soil as compared to sewage sludge unamended soil. FYM was found more effective in enhancing Cr uptake by roots than other chelating agents at both the growth stages. In case of shoots, Chromium uptake ranged from 943.77 to 2385.09 µg pot^{-1} (Table 3), being maximum in plants grown on Cr_{20}+CDTA treated soil and the least in Cr_{20}+CA. The mean Cr uptake by shoots, was 817.16 and 1012.10 µg pot^{-1} in sewage sludge unamended and amended soil at first stage of growth, respectively. Application of CDTA increased uptake by 176.87 and 158.98% in sewage sludge unamended and amended soils at first stage but it was relatively higher (129.30 and 122.17%) at second stage of growth, respectively as compared to control (Cr_{20}). The results further reveal that addition of sewage sludge increased Cr uptake at both the growth stages. It is clear from the data in table 3 that uptake of Cr increased due to CDTA than any other chelating agents. This is most probably due to higher dry matter yield yield and Cr content in shoots of CDTA treated pots. Similar results were also observed by Meers *et al.,* (2005) who reported that EDDS was more effective in increasing the metal uptake followed by EDTA, NTA and citric acid. Tandy *et al.* (2006) reported that uptake of metals during chelate assisted phytoextraction is related to the solublized metal concentration in the growth medium.

Table 2. Effect of chelating agents and sewage sludge on dry matter yield (g pot^{-1}) of roots and shoots of *Zea mays* crop in Cr contaminated soil.

	50 DAS		80 DAS		
Treatment	**Without SS**	**With SS**	**Without SS**	**With SS**	**Mean**
Root					
Cr_{20}	3.91	4.13	4.32	4.65	4.25
Cr_{20}+CDTA	2.83	3.16	3.65	4.00	3.41
Cr_{20}+CA	2.35	2.41	2.96	3.41	2.78
Cr_{20}+DTPA	2.61	2.75	3.23	3.69	3.07
Cr_{20}+NTA	1.23	1.46	1.91	2.43	1.76
Cr_{20}+FYM	4.00	4.35	5.31	5.96	4.90
Mean	2.82	3.04	3.56	4.02	
CD (0.05)	Soil=0.12, Time=0.12, Chelating Agent=NS, SxT=0.17, SxCA=0.30, TxCA=NS, SxTxCA=NS				
Shoot					
Cr_{20}	15.91	17.63	26.14	29.16	22.21
Cr_{20}+CDTA	26.23	29.15	40.31	43.10	34.70
Cr_{20}+CA	10.25	12.32	17.86	20.17	15.15
Cr_{20}+DTPA	13.16	15.61	24.16	27.39	20.08
Cr_{20}+NTA	9.85	11.10	15.31	17.36	13.40
Cr_{20}+FYM	19.64	22.36	34.61	37.11	28.43
Mean	15.84	18.03	26.40	29.05	
CD(0.05)	Soil=0.99, Time=0.99, Chelating Agent=NS, SxT=1.40, SxCA=2.43, TxCA=NS, SxTxCA=NS				

Table 3. Effect of chelating agents and sewage sludge on Cr uptake (ìg pot^{-1}) by roots and shoots of *Zea mays* crop in Cr contaminated soil.

	50 DAS		80 DAS		
Treatment	**Without SS**	**With SS**	**Without SS**	**With SS**	**Mean**
Root					
Cr_{20}	249.48	285.80	514.58	581.71	407.89
Cr_{20}+CDTA	343.32	412.93	657.78	757.94	542.99
Cr_{20}+CA	259.69	294.11	400.93	492.53	361.81
Cr_{20}+DTPA	306.32	69.290	476.33	567.07	354.75
Cr_{20}+NTA	176.49	225.29	461.78	605.77	367.33
Cr_{20}+FYM	392.92	457.79	637.97	764.86	563.38
Mean	288.04	290.87	524.89	628.31	
CD (0.05) Soil=20.34, Time=20.34, Chelating Agent=35.24, SxT=28.77, SxCA=49.83, TxCA=49.83, SxTxCA=70.48					
Shoot					
Cr_{20}	567.23	726.44	1232.98	1468.04	998.67
Cr_{20}+CDTA	1570.42	1881.28	2827.23	3261.46	2385.09
Cr_{20}+CA	524.55	688.04	1168.01	1394.47	943.77
Cr_{20}+DTPA	765.39	984.75	1670.47	2026.42	1361.76
Cr_{20}+NTA	665.60	777.35	1151.67	1383.44	994.51
Cr_{20}+FYM	809.75	1014.72	1914.51	2220.38	1489.84
Mean	817.16	1012.10	1660.81	1959.04	
CD(0.05) Soil=69.55, Time=69.55, Chelating Agent=NS, SxT=98.36, SxCA=170.37, TxCA=NS, SxTxCA=NS					

References

Alkorta, I., J. Hernandez-Allica, J. M. Becerril, I. Amezaga, I. Albizu, M. Onaindia, C. Garbisu. 2004. Chelate-enhanced phy-toremediation of soils polluted with heavy metals. *Reviews in Environmental Science and BioTechnology* 3: 55–70.

Blaylock, M. J., D. E. Salt, S. Dushenkov, O. Zakharova, C. Gussman, Y. Kapulnik, B. D. Ensley and I. Raskin. 1997. Enhanced accumulation of Pb in Indian mustard by soil-applied chelating agents. *Environmental Science and Technology*. 31**:** 860-865.

Garbisu, C. and I. Alkorta. 2001. Phytoextraction: a cost-effective plant-based technology for the removal of metals from the environment. *Bioresource Technology* 77: 229–236.

Kulli, B., M. Balmer, R. Krebs, B. Lothenbach, G. Geiger and R. Schulin. 1999. The influence of nitrilotriacetate on heavy metal uptake of lettuce and ryegrass. *J. Environ. Qual*. 28: 1699-1705.

McGrath, S. P. and F. J. Zhao. 2003. Phytoextraction of metals and metalloids from contaminated soils. *Current Opinion in Bio-technology 14*: 277–282.

Meers, E., A. Ruttens, M. J. Hopgood, D. Samson and F. M. Tack. 2005. Comparison of EDTA and EDDS as potential soil amendments for enhanced phytoextraction of heavy metals. *Chemosphere*. 58 (8): 1011-1022.

Mishra, S. 2004. *Effect of chelating agents, FYM and herbicides on the phytoextractability of Indian mustard from Pb enriched soil*. M.Sc Thesis. CCS Haryana Agricultural University, Hisar, Haryana.

Pulfore, I. D. and C. Watson. 2003. Phytoremediation of heavy metal-contaminated land by trees – a review. *Environment Interna-tional* 29: 529–540.

Robinson, B. H., T. M. Mills, D. Petit, L. E. Fung, S. R. Green and B. E. Clothier. 2000. Natural and induced cadmium - accumulation in poplar and willow**:** Implications for phytoremediation. *Pl. Soil* 227: 301-306.

Tandy, S., R. Schulin and B. Nowack. 2006. Uptake of metals during chelant-assisted phytoextraction with EDDS related to the solubilized metal concentration. *Environ. Sci. Technol.*40:2753-2758.