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CASE STUDY

Effect of compost and humic acid in mobility and concentration of cadmium and chromium in soil and plant

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ABSTRACT: The effect of compost and humic acid in mobility and concentration of cadmium and chromium in contaminated soil were investigated. Experiment was carried out with three levels of soil cadmium and chromium and two organic matters (compost and humic acid). The study was performed in a randomized complete block design with 3 replicates. Results indicated that application of organic substances enhanced movement of cadmium and chromium in soil column. Humic acid is more effective than compost on the mobility of cadmium and chromium in soil. Mobility of cadmium and chromium in the lower depths of soil column were increased. Cadmium and chromium concentration in shoots and roots enhanced due to increasing those concentration in soil and application of organic substances. Increase in cadmium in shoots can be attributed to the high mobility of this element in maize plant. Maize root chromium concentration was greater than shoot chromium concentration. Humic acid was more effective than compost as cadmium and chromium concentration in root and shoot was concerned. Low mobility of chromium in plant and accumulation of chromium in roots can be reasons of decreasing of chromium concentration in shoot of plant and its bioaccumulation.

KEYWORDS: *Cadmium; Chromium; Compost; Humic acid; Mobility*

INTRODUCTION

Heavy metal pollution in soils is a common problem in several countries (John *et al.,* 2009). Today soil pollution with heavy metals is a major environmental problem in the world. This pollution has harmful effects on the soil fauna, which reduces the yield and quality of agricultural products (Raymond and Okieimen, 2011). Physiological and biochemical process in plants were affected by high concentration of heavy metals such as cadmium and chromium (Kim *et al*., 2004; Strobel *et al*., 2005). Cadmium is one of the most toxic metals that effects on all biological processes of humans, animals and plants; such as leaf chlorosis, photosynthesis, reduce root growth, decrease absorption and transport of nutrient elements (Gabrijel *et al.,* 2009; Orrono; Lavado, 2009; Karbassi *et al.,* 2015). The contents of Cd in soils range from 0.06 to 4.3 mg/kg and in plants range from 0.005-0.252 mg/kg (Kabata and Mukherjee, 2007; Wojcik *et al*., 2005). Chromium in high concentration also affected human health and plant growth. The world soil average content of Cr in soils was 60 mg/kg and it rang in plants from 0.01-0.08 mg/kg (Kalantari *et al*., 2014; Sanita *et al*., 2010). Besides soil pH, the content of organic matter affect availability of heavy metals. Organic matter (OM) can bind with heavy metals and controlling their behavior in soils (Park *et al.,* 2011; Salati *et al*., 2010; Tingqiang *et al.,* 2013; Wyszkowski *et al.,* 2013; Karbassi and Pazoki, 2015). Soil organic matter is one of the important soil components that affects soil reactions such as form of ion uptake by plants (Ouni *et al.,* 2014). OM also effect plant growth by increasing in improvement of photosynthesis system, micro and macro element

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uptake and changing in growth of root cells (Pizzeghello *et al.,* 2013). Some researchers believe that application of OM, such as compost, humic acid and sewage sludge, is a recommend way for mobilization of heavy metals in soils. In many of that experiments the stimulating substances were undoubtedly largely chelated elements, chiefly iron (Khan *et al.,* 2000). They find that application of compost and humic acid in contaminated soils enhanced bioavailability and mobility of metals (Pizzeghello *et al.,* 2013; Topcuoglu, 2012) while others showed that organic substances have the ability to bind metal ions and lead to stabilization of them (Zhang *et al.,* 2013). Thus, the influence of OM such as compost and humic acid on availability and movement of Cd in soil and plant require further investigation. Therefore the research was aimed to investigate the effect of compost and humic acid on the mobility and accumulation of chromium and cadmium in soil and plant. This study has been performed in greenhouse of soil department of Shahid Chamran University of Ahvaz, Iran during April to September 2015.

MATERIALS AND METHODS

Experimental design

The study was car-ried out in greenhouse of the Shahid Chamran University, using soil columns. Treatments were performed with the level of Chromium (0, 100, 200 mg Cr/kg soil) using $K_2Cr_2O_7$, Cadmium (0, 25, 50 mg cd/kg soil) and using $CdCl_2$ along with two levels of organic matter; compost (0, 40 mg/kg soil) and humic acid (0, 2.5 g). Investigation of these elements and their value were performed due to reports of their contamination in the soils in Ahvaz, Iran (Jafarnghadi *et al.,* 2010; Parsafar and Marofi, 2013). Treatments were performed: 1) control: no Cd and Cr (T_0) + no organic substance (M_0); 2) compost (M_1) + T_1 (25 mg cd/kg soil+ 100 mg Cr/kg soil); 3) humic acid (M_2) + T_1 (25 mg cd/kg soil+ 100 mg Cr/kg soil); 4) compost (M_1) + T_2 (50 mg cd/kg soil+ 200 mg Cr/kg soil); 5) humic acid (M_2) + T_2 (50 mg cd/kg soil+ 200 mg Cr/kg soil). The treatments were performed in factorial randomized complete design with 3 replicates. Factorial design of treatments shows in Table.1. The surface of soil was mixed with 40 g compost/kg soil. Humic acid was purchased from the Flvka Institute, as the commercial sample (product No. 35069288) and as described by Hoop (1990). Seeds of plant (single grass 704) were sowed in plastic columns containing 14 kg of soil. Up to harvest received proper operations. Prior to column filling, soil simultaneously mixed with required amount of Cd and Cr solution then incubated for one month fallowed by application of NPK as per soil test. Based on molecular weight of used salt for each understanding element, required amount of each salt for every treatment were calculated. The required quantity of each salt for each treatment was dissolved in 500 ml distilled water and sprayed over the soil informally. The soil texture was sandy loam and the general properties was provided in Table.2. At the end of the study, plants were harvested then roots were isolated from soli and washed with deionized water. The maize plant components (shoot and roots) then dried in an oven at 65º C for 72 h and then in fallowed by powdering the samples.

Determination of heavy metals

Concentration of heavy metals in plant was analyzed by atomic absorption spectroscopy at the following wavelengths: cadmium at 228.8 nm and chromium at 357.9 nm (Mireles, 2004). Cd and Cr concentration in soil column was measured in three depth (0-15, 15-30 and 30-45 cm) as previously described by Chapman and Pratt (1961). The samples were measured against a calibration graph which was obtained with measurements on matrix-compensated standards (Boner and Pohl, 2010).

Transportation index

The transportation index (Ti) was determined by the ratio between shoot and root heavy metal concentration (Marchiol *et al.,* 2004).

Ti = Cd or Cr concentration in shoot (mg/kg)/ Cd or Cr concentration in roots (mg/kg)

Bio accumulation factor

Bio accumulation factor is the ratio of the metal concentration in the plant to its concentration in the soil which is calculated from the following equation: (Parsafar and Marofi, 2013).

BAF = Mpc/Msc

Mpc and Msc represents the concentration of element in plant and soil mg/kg respectably.

Statistical analysis

Statistical analysis employed SAS windows version 9.1. The significance of differences between variables

Table 1: Factorial design of treatments

R1	M0T2	M1T1	M2T1	M0T0	M1T2	M0T1	M1T0	M2T0	M2T1
R2	M1T0	M0T0	M2T1	M2T0	M1T1	M2T1	M0T1	M1T2	M0T2
R3	M2T1	M1T2	M2T0	M1T0	M0T2	M2T1	M1T1	M0T0	M0T1

Table 2: Characteristics of soil and organic matter

Characteristics	texture	TOC* (%)	pH	Total N (%)	Total P (mg/Kg)	Total K (mg/Kg)	Total Cd (mg/Kg)	EC (ds/m)
Soil	sandy loam	0/68	7.8	0.06	12	105	0.043	2.2
Compost	---	21.3	7.53	1.39	0.65	2.1	0.02	2.5

*TOC: Total organic carbon

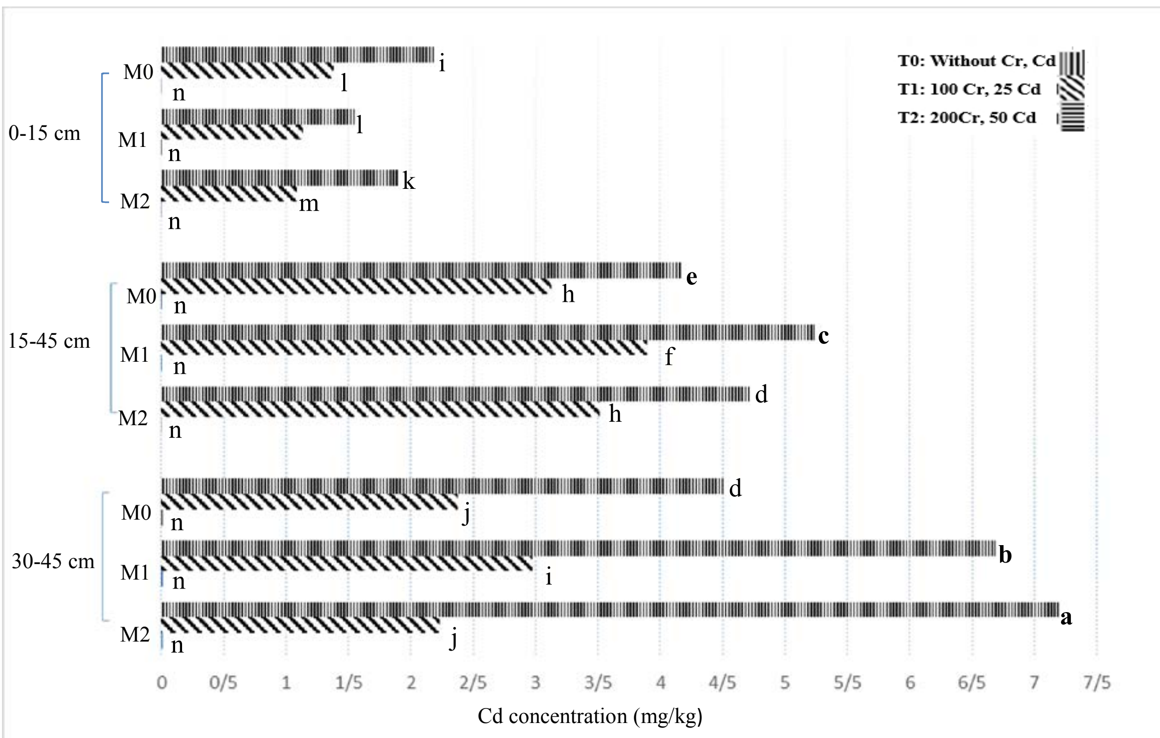

Fig. 1: Effects of compost and humic acid on Cd concentration in soil column. Values are means (n = 3) Similar letters are not significantly different at *P<0.05*, according to Duncan's test. M0: without organic substance, M1: compost, M2: Humic acid.

at P<0.05 was checked with a multiple comparison on Duncan's test.

RESULTS AND DISCUSSION

Concentration of Cd in soil column

Application of organic substances compered to control led to increase available Cd concentration in all treatments (Fig. 1). Highest concentration of Cd recorded in depth 30-45 cm for M_2T_2 treatment. It was clear that application of organic substances enhanced Cd concentration by depth. At T_1 treatment, application of organic substances with no significant difference between compost and humic acid decreased Cd concentration in 0-15cm but increased in lower depths. In M_1T_2 treatment and depth 30-45cm, Cd concentration increased about 74 and 20% compare to depth 0-15

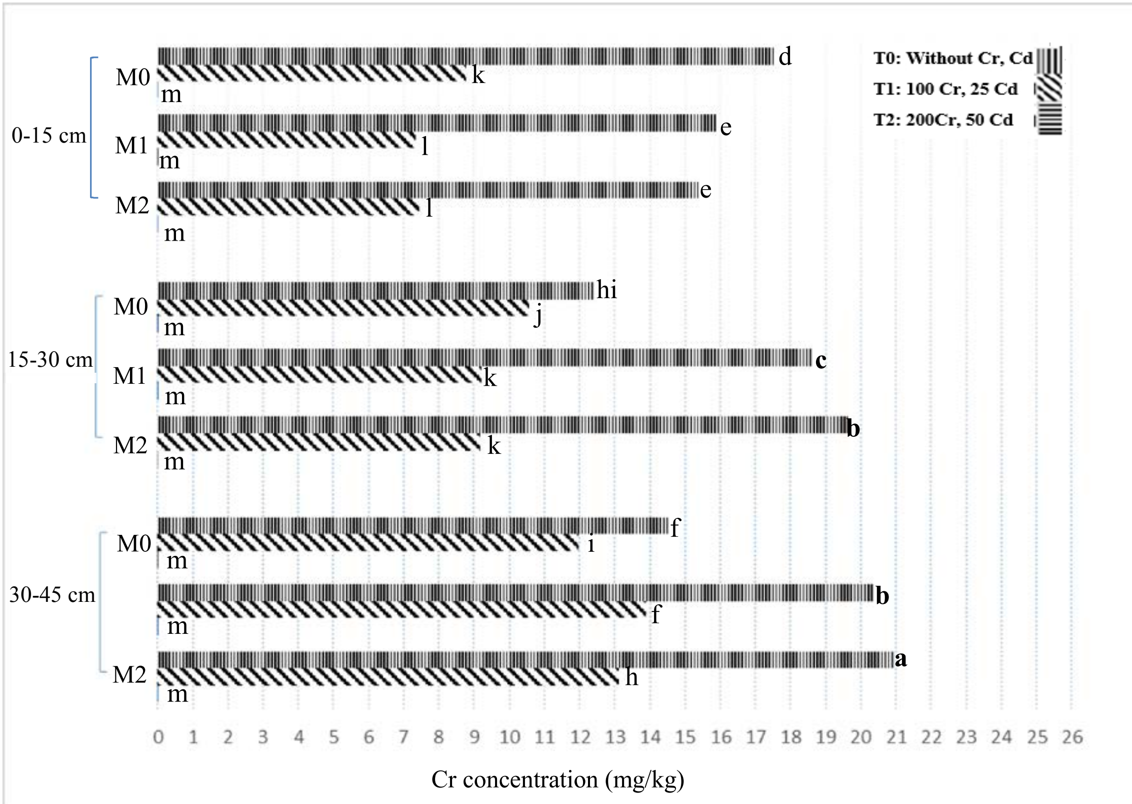

Fig. 2: Effects of compost and humic acid on Cr concentration in the soil column. Values are means (n = 3). Similar letters are not significantly different at $P<0.05$, according to Duncan's test. M0: without organic substance, M1: compost, M2: Humic acid.

and 15-30cm respectively. Cadmium movement in soil column was more pronounced by humic acid then compost. In general over the test period, the concentration of cadmium decreased in 0-15 cm and accumulation of Cd was happened in the depth 30-45 cm. Therefore application of organic substances enhanced mobility of Cd. Increased heavy metals amount such as Pb and Cd by compost have been reported (Ben Achiba *et al.,* 2009). Enhancement in Cd availability and mobility eventually was due to multiple functional groups of organic substances (Zhang *et al.,* 2013) that can interacted with metal ions (Cd). This result is alike with other researchers (Angelova *et al.,* 2013; Meers *et al.,* 2005) indicating that Cd concentration was effected by soil properties such as organic substances.

Concentration of Cr in soil column

Fig. 2 is a presentation of the concentrations of Cr in soil column treated with organic substances. Application of humic acid and compost increased the concentration of chromium in lower depths. Highest concentration of Cr observed in 30-45 cm and M_2T_2 treatment. Cr concentration in 30-45 cm and M_1T_1 was increased about 16.6 % compared to M_0T_1. There was no significant difference between types of organic substances for 0-15 cm. Cr concentration in 15-30 cm enhanced about 50% in M_2T_2 and 43% in M_1T_2 compare to M_0T_0. Humic acid was effective than compost as Cr concentration in lower depths. Concentration of Cr in soil column followed increasing trend but it was less than cadmium. Organic substances can form complex compounds with Cr (Barancikova and Makovnikova, 2003) and also maintaining Cr in an exchangeable situation. Cr competition with other elements such as Ca in the creation of complexes with organic materials (Mohammad *et al.,* 2013; Hernandez *et al.,* 2012) and low mobility in soil (Janos *et al.,* 2010) can be the reasons for those results.

Cd concentration in shoot and root

Data of the Table. 3 showed that the concentration of Cd in shoot and root of plant (received Cd treatments) was enhanced with application of organic substances. At T_0 treatment, the Cd concentration of maize plant (shoot and root) showed no significant difference. humic acid in combination with T_1 and T_2 which was more effective than compost as increasing Cd concentration in root and shoot. Increasing in cadmium concentration in maize plant can be attributed availability and mobility of this element in soil (Fig. 1). At M_1T_2 and M_2T_2 treatments, Cd concentration increased by 26% and 132% in shoot and 4.5% and 80% in root as compared to control (M_0T_2) respectively. These results suggest that soil organic substances can effectively increase Cd concentration in maize plant in polluted soils. Furthermore, root Cd concentration exceeded that of shoot. M_2T_1 and M_2T_2 treatments increased cd concentration in root by 100 and 80% compared to control (M_0T_2), respectively. In order to preventing toxic accumulation of heavy metals in shoot, plant roots can reserve heavy metal by retention of them in vacuoles (Orrono and Lavado, 2009; Fischerova *et al.,* 2005). Cadmium accumulation in root of maize plant than shoot can be due to connecting of Cd in cation exchange sites of roots (Zeng *et al.,* 2011) and creating complexes with ligands containing –S group (Sulfhydryl) (Topcuoglu, 2012) which led to sediment and accumulation of cd in apoplast of root. Maximum Ti of Cd with no significant different between organic substances was observed in T_0 treatment (Table 2). Compared with the respective T_0 treatment increment of soil Cd concentration decreased Ti significantly. Such an increment in Cd concentration in soil (Fig. 1) or in root (Table 2) will activate mechanisms of retention of Cd in root and causes hindrance of its translocation. Minimum amount of Ti was observed in M_0T_2. Even though no significant different was observed in Ti recorded for application of organic substances but increasing level of Cd concentration from T_1 to T_2 in combination with M_1 and M_2 increased Ti. The decreasing trend in T_0 to T_1 and increasing trend from T_1 to T_2 in combination with M_1 and M_2 treatment indicated that eventually increasing Cd concentration in root reduces Cd retention capacity which in turn increase Cd up ward translocation. Such reduction in Cd retention capacity might be due to neutralization of negative charges in apoplast (Park *et al.,* 2013; Strobel *et al.,* 2005) or limited capacity of phytochelation production of plant cells (Parsafar and Marofi, 2013). As a consequence, the transmission of Cd in plant (root to shoot) specified the distribution of Cd concentration in plant (Haliru *et al.,* 2009).

Cr concentration in shoots and roots

Enhancement of Cr application increased it concentration in plant (shoot and root) (Table 4).The influence of organic substances on concentration of Cr in shoot followed decreasing trend. In M_1T_2 and M_2T_2 treatments, Cr concentration in shoot was

Table 3: Effect of application of heavy metals and type of organic substances in concentration and transportation index of Cd in maize plant (shoot and root)

Treatment	shoot Cd (mg/kg dw)			root Cd (mg/kg dw)			Transportation index (Ti)		
	M_0	M_1	M_2	M_0	M_1	M_2	M_0	M_1	M_2
T_0	0.08^e	0.07^e	0.07^e	0.09^e	0.08^e	0.08^e	0.88^a	0.87^a	0.87^a
T_1 ($Cr_{100}Cd_{25}$)	8.5^d	10.4^d	16.2^b	14.3^d	15.4^d	28.4^b	0.6^b	0.67^b	0.57^{bc}
T_2 ($Cr_{200}Cd_{50}$)	13.3^c	16.7^b	30.9^a	25.1^c	24.2^c	45.1^a	0.53^c	0.69^b	0.68^b

Treatments mean (n = 3). The same letters in each column showing no significant effect ($P<0.05$), by Duncan's test.
M_0: without organic substance, M_1: compost, M_2: Humic acid.

Table 4: Effect of application of heavy metals and type of organic substances in concentration and transportation index of Cr in maize plant (shoot and root)

Treatment	shoot Cr (mg/kg dw)			root Cr (mg/kg dw)			Transportation index (Ti)		
	M_0	M_1	M_2	M_0	M_1	M_2	M_0	M_1	M_2
T_0	0.04^f	0.03^f	0.03^f	0.09^e	0.07^e	0.08^e	0.44^d	0.42^d	0.42^d
T_1 ($Cr_{100}Cd_{25}$)	71.6^b	30.4^e	24.3^e	54.3^d	61.7^d	98.1^b	1.31^a	0.49^d	0.24^e
T_2 ($Cr_{200}Cd_{50}$)	98.8^a	59.8^c	38.9^d	91.9^c	92.6^c	144.7	1.07^b	0.64^c	0.26^e

Treatments mean (n = 3). The same letters in each column showing no significant effect ($P<0.05$), by Duncan's test.
M_0: without organic substance, M_1: compost, M_2: Humic acid.

Table 5: Bio accumulation factor (BAF) of heavy metals

Treatment	Cd BAF			Cr BAF		
	M_0	M_1	M_2	M_0	M_1	M_2
T_0	1/09[a]	1/05[a]	0/9[b]	3/09[a]	2/06[c]	2/7[b]
$T_1(Cr_{100}Cd_{25})$	1/1[a]	0/5[c]	0/5[c]	1/4[d]	0/57[f]	0/53[f]
$T_2(Cr_{200}Cd_{50})$	1/16[a]	0/44[c]	0/38[d]	1/06[e]	0/7[f]	0/6[f]

Treatments mean (n = 3). The same letters in each column showing no significant effect ($P<0.05$), by Duncan's test. M_0: without organic substance, M_1: compost, M_2: Humic acid.

decreased by 39.4 and 60.6% compared to control (M_0T_2), respectively. Minimum amount of Cr concentration in shoot was observed in M_2T_1 treatment. In T_1 and T_2 treatments, application of organic substances increased concentration of Cr in root compared to T_0. M_0 and M_1 in T_0 and T_1 treatments showed no significant difference. In M_1T_1 and M_2T_1 treatments, Cr concentration in root was increased by 13.6 and 80.6% compared to control (M_0T_1), respectively. Highest accumulation of Cr concentration in root was observed in M_2T_2 treatment. Low mobility of Cr in plant (root to shoot) can be reasons by saturation and accumulation of Cr in apoplast of cells (Kim *et al.,* 2004; Topcuoglu, 2012). Root Cr concentration was greater than shoot Cr concentration. In general chromium is mainly accumulated in roots due to immobility of Cr in vacuoles of root cells (Park *et al.,* 2011). Enhance in Cr concentration due to application of organic substances attributed to the formation of complex organic matter with metal. Therefore the highest amount of Cr uptake remain in roots. Experiments indicated that Cr has low mobility then Cd in plants (Haliru *et al.,* 2009; Kalantari *et al.,* 2014). Researchers find that Cd and Cr can complex by different functional groups of organic substances, this compounds were regulated heavy metal mobility and concentration in soil (Angelova *et al.,* 2013; Lesage *et al.,* 2005). In general chromium by containing negative charges, was immobilized in roots of plant and its translocation was low. The transportation index (Ti) of Cr is shown in Table 3. The fewest amount of Ti was in M_2T_2. In T_1 and T_2 treatments, Transportation index of Cr followed decreasing trend with application of organic substances. The low Transportation index of Cr represents the accumulation of Cr in roots. Application of organic substances decrease Ti. The concentration of Cr between root and shoot indicated that there was a limitation root to shoot movement of Cd and Cr (Wojcik *et al.,* 2005).

Bio accumulation factor (BAF)

The Lowest values of BAF after control was observed in M_1T_2 (Table 5). This factor is directly related to the concentration of elements in plant shoot. So that increasing concentrations of cadmium and chromium in plant shoot (Tables 3, 4) increased this factor. High mobility of cadmium in the effect of organic substances in soil led to decline BAF in M_1 and M_2 treatments. In T_1 and T_2, application of organic matter was decreased BAF due to the low concentration of chromium in plant against soil. According to study bio accumulation factor, can be realized the behavior of plant in uptake of heavy metals. BAF was used for qualification of plant species for phytostabilization of contaminated soils. The BAF values in the range of 0.1-0.50 (as potential phytostabilization) and values from 0.5-1.0 (as partial phyto-stabilization) should not be neglected (Balabanova, *et al.,* 2015).

CONCLUSION

In conclusion, it seems that chromium is likely to increase in plant roots enhances root to shoot movement of cadmium. Cr was accumulated more than Cd by the roots. Root heavy metal concertation was greater than shoot. Mobility of cadmium and chromium in soil was effected by application of organic substances. Organic substances by multiple functional groups can creation metal-humic complexes and increase heavy metals mobility in soil.

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CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest regarding the publication of this manuscript.

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