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THE EFFECT OF ORGANIC AMENDMENTS ON SOIL CHEMICAL CHARACTERISTICS

V. R. ANGELOVA[1*], V. I. AKOVA[1], N. S. ARTINOVA[2] and K. I. IVANOV[1]
[1] *University of Agriculture, Department of Chemistry, BG - 4000 Plovdiv, Bulgaria*
[2] *University of Agriculture, Department of Soil Science, BG - 4000 Plovdiv, Bugaria*

Abstract

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Enrichment of soils with organic matter could reduce the content of bioavailable metal species as a result of complexation of free ions of heavy metals. In this experiment, the influences of different rate of organic amendments on the soil chemical characteristics, as well as organic content and humus fractions and their ability to reduce heavy metal availability in contaminated soils were investigated. The field experiment was conducted on soils contaminated by the Non-Ferrous-Metal Works near Plovdiv, Bulgaria. Different amounts of vermicompost and compost (5 and 10 g.kg^{-1}) were incorporated into the top 20 cm of soil. To determine the effect of the organic amendments, the soil samples were collected 1 month after addition of organic amendments from depth of 20 cm. The soil characteristics (pH, EC, organic content and humus fractions) were determined. The total content of micro, macroelements, heavy metals and concentration of their available species in the soil were determined using aqua regia and DTPA extraction, respectively. The results showed that compost and vermicompost treatments had significant effect on soil physical and chemical properties like EC, pH, organic matter, macro and micronutrients content. Compared with the unamended soil, soil treated with organic amendments showed apparent increases of organic matter, total N, pH, EC and available macroelements (P, K, Ca and Mg). Organic amendments influenced considerably the humus fractions by changing the ratio C_{HA}/C_{FA} in favour of the humic acids, in which changes of the type of humus from filvic type in control to humic-fulvic type in addition of 5 g.kg^{-1} and 10 g.kg^{-1} vermicompost and humic type in addition of 10 g.kg^{-1} compost. Application of the compost and vermicompost reduces the amount of aggressive fulvic acids, organic matter components that pose a potential environmental risk. The results of the present study indicated that soil application of compost and vermicompost decreased DTPA-extractable levels of heavy metals in the soil. This is indicative of heavy metals immobilisation by humic substances from compost and vermicompost application. Results appear that verify the function of humic acid in improving phytoremediation efficiency of soils contaminated with heavy metals and potential environmental availability of metals may be controlled by soil organic amendments.

Key words: organic amendments, organic carbon, micro and macroelements, heavy metals, availability, contaminated soils

Introduction

Contamination of soil by heavy metals is of major concern because of their toxicity. Elevated levels of heavy metals in soils may lead to their uptake by plants, which depends not only on heavy metal contents in soils but is also determined by soil pH value, organic matter and clay contents, and influenced by the fertilization (Fytianos et al., 2001). The fluctuation of mentioned parameters cannot change the total amount of heavy metals in soil but can significantly affect their bioavailability (Ge et al., 2000). Soil amendment is a major requirement for the successful establishment of vegetation in metal-contaminated soils. The addition of amendments into soils precipitates or increases metal sorption, therefore decreasing the proportion of the total element in soil solution (Brown et al., 2005; de Varennes and Queda, 2005; de Varennes et al., 2006). The addition of amendments such as fly ash, pig manure, sewage sludge, is effective in lowering the

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E-mail: vileriz@yahoo.com

metal toxicity of soil and provides a slow release of nutrient sources such as N, P, K to support plant growth (Wong, 2003; Chiu et al., 2006).

Addition of organic matter amendments, such as compost, fertilizers and wastes, is a common practice for immobilization of heavy metals and soil amelioration of contaminated soils. Some researches showed that amendment of contaminated soils with organic matter reduced bioavailability of heavy metals (Khan et al., 2000). Soil organic matter has been of particular interest in studies of heavy metal sorption by soils, because of tendency of transition metal cations to form stable complexes with organic ligands (Elliott et al., 1986). Organic matter is known to form strong complexes with heavy metals (Krogstad, 1983). The content of organic matter affects speciation of heavy metals in soil (Lo et al., 1992). High organic matter content was reported to decrease concentrations of Cd and Ni in soil solution (Arnesen and Singh, 1999). This is very important because a high content of organic matter in contaminated soil is one of the ways to exclude heavy metals from the trophic chain. Cow manure, poultry manure and pig manure were found to be effective in reducing lead availability to plants, leading to lower uptake of lead (Scialdone et al., 1980; Wong and Lau, 1985; Ye et al., 1999). They are commonly used as tailings amendments because the addition of organic matter can significantly improve the physical characteristics and the nutrient status of mine soil (Ye et al., 1999). In addition, fertilizers are an essential ingredient for successful restoration of mine wastes (Bradshaw and Chadwick, 1980)

The use of composts has been recognized generally as an effective means for improving soil aggregation, structure and fertility, increasing microbial diversity and populations, improving the moisture-holding capacity of soils, increasing the soil cation exchange capacity (CEC) and increasing crop yields (Zink and Allen, 1998).

Vermicompost contains most nutrients in plant-available forms such as nitrates, phosphates, and exchangeable calcium and soluble potassium (Orozco et al., 1996). There is accumulating scientific evidence that vermicomposts can influence the growth and productivity of plants significantly (Edwards, 1998). Various greenhouse and field studies have examined the effects of a varity of vermicomposts on a wide range of crops including cereals and legumes (Chan and Griffiths, 1988), vegetable, ornamental and flowering plants (Atiyeh et al., 2000), and field crops (Arancon et al., 2004).

The aim of this study was to evaluate the impact of different rate of organic amendments on (i) some chemical characteristics in contaminated soils, (ii) the soil organic carbon and humus fractions, (iii) DTPA-extractable micro and macronutrients and (iv) ability to reduce heavy metals availability in heavy metal contaminated soils.

Materials and Methods

The experiment was performed on an agricultural field contaminated by the Non-Ferrous-Metal Works near Plovdiv, Bulgaria. The field experimental was a randomized complete block design containing five treatments and four replications (20 plots). The treatments consisted of a control (no organic amendments), compost amendments (added at 5 and 10 g.kg^{-1}), and vemicompost amendments (added at 5 and 10 g.kg^{-1}). Plot size was 24 m^2 (3 m x 8 m). The soil from each plot were combined and mixed with amendments. Characteristics of soils and organic amendments are shown in Table 1. The soils used in this experiment were slightly acidic, moderate content of organic matter and essential nutrients (N, P and K) (Table 1). Pb, Zn, Cd and Cu total concentrations were high and exceeded the limit value in agriculture soils. The psedo-total content of Zn, Pb and Cd is high (1430.7 mg.kg^{-1} Zn, 876.5

Table 1
Characterization of the soil and the organic amendments used in the experiment

Parameter	Soil	Compost	Vermo-compost
pH	6.5	6.9	7.5
EC, dS.m^{-1}	0.2	0.2	2.2
Organic content, g.kg^{-1}	2.22	72.1	38.58
N Kjeldal, g.kg^{-1}	0.24	2.22	1.57
C/N	9.4	32.4	24.6
Pseudo-total P, mg.kg^{-1}	642	12654	10211
Pseudo-total K, mg.kg^{-1}	5518	6082	10495
Pseudo-total Ca, mg.kg^{-1}	10608	32159	31848
Pseudo-total Mg, mg.kg^{-1}	9578	2087	7755
Pseudo-total Pb, mg.kg^{-1}	876.5	12	32.3
Pseudo-total Cu, mg.kg^{-1}	124.8	43.23	53.3
Pseudo-total Zn, mg.kg^{-1}	1430.7	170.8	270.3
Pseudo-total Cd, mg.kg^{-1}	31.4	0.19	0.69
Pseudo-total Fe, mg.kg^{-1}	41651	3177.3	11814
Pseudo-total Mn, mg.kg^{-1}	1244.5	360.5	423.3

mg.kg^{-1} Pb and 31.4 mg.kg^{-1} Cd, respectively) and exceeds the maximum permissible concentrations (200 mg.kg^{-1} Zn, 70 mg.kg^{-1} Pb, 1.5 mg.kg^{-1} Cd).

To determine the effect of the organic amendments, the soil samples were collected 1 month after addition of organic amendments. A soil subsample was air-dried, passed through a 2-mm sieve and characterized for soil pH (H_2O) and electrical conductivity in deionised water suspension of 1:5 (w/v); total nitrogen by the Kjeldahl method (N Kjeldahl); total oxidizable organic carbon according to Tjurin's method in modification of Nickitin and Fishman (with titration) (1969); organic fractions – accelerated method of Kononova – Belchikova (Kononova, 1966), following one time extraction with sodium pyrophosphate (0.1 M $Na_4P_2O_7.10H_2O$ mixed with 0.1 N NaOH). The humic acids (HA) and the fulvic acids (FA) were separated by precipitation of the HA, bringing the alkaline extract to pH ≈ 1 with concentrated H_2SO_4. The fraction of the combined with R_2O_3 and movable humic acids was obtained by additional treatment of the soil sample with a solution of 0.1 N NaOH, and the so-called 'brown' humic acids were separated by precipitation with concentrated H_2SO_4 to pH ≈ 1. Optical characteristics of humic acid were measured on spectrophotometer BOECO S-22 (absorption at λ 465 nm and 665 nm). The same procedures were applied to oganic amendments.

The pseudo-total and DTPA-extractable concentration of heavy metals, micro and macroelements in the soils, after four weeks' equilibration were determined. Pseudo-total content of metals in soils was determined in accordance with ISO 11466. The available nutrients and mobile heavy metals contents were extracted by a solution of DTPA (1 M NH_4HCO_3 and 0.005 M DTPA, pH 7.8) (Soltanpour and Schwab, 1977). The same procedures were applied to oganic amendments.

To determine the heavy metal content in the plant and soil samples, inductively coupled emission spectrometer (Jobin Yvon Horiba "ULTIMA 2", France) was used. Three independent replicates were performed for each sample and blanks were measured in parallel.

Statistical analysis

Statistical analyses were conducted with Statistica v. 7.0. Pearson's linear correlations were used to assess the relationships among pH, the soil organic content, various organic carbon fractions and available Pb, Cu, Zn and Cd in soil.

Results and Discussion

The effects of compost and vermicompost on pH, soil organic content and organic fractions, and total N are summarized in Figure 1. The results obtained by us showed that the soil organic properties depended on the type and rate of the soil amendments and treatment.

pH

Soil pH varied with amendment treatment (Figure 1). Application of compost reduced the soil pH significantly as

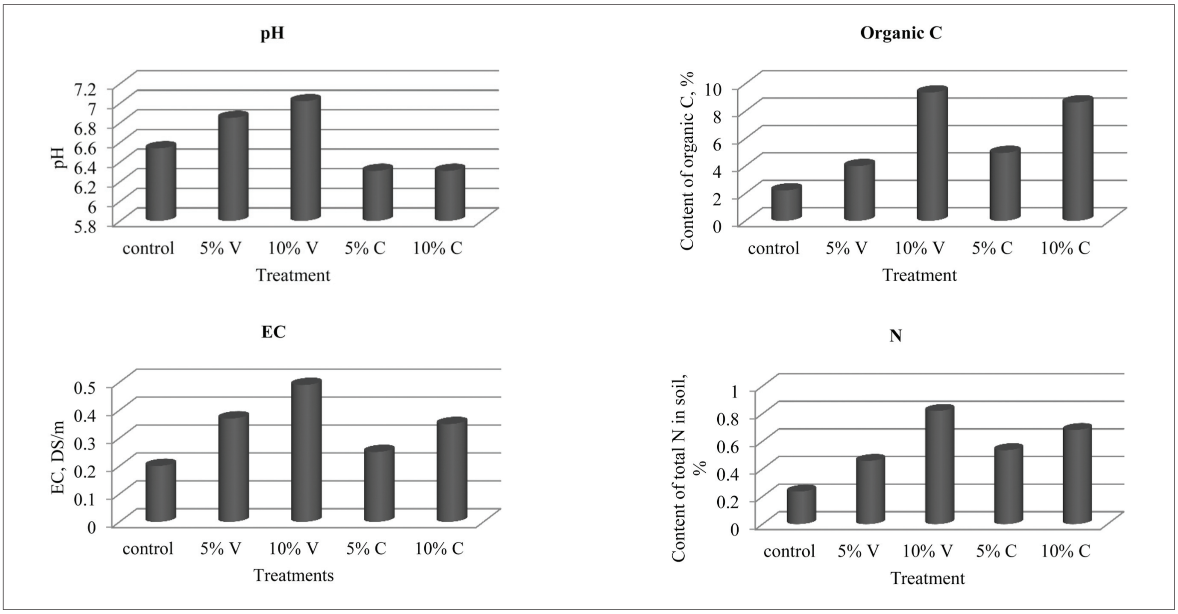

Fig. 1. Influence on compost (C) and vermicompost (V) treatments on pH, EC, soil organic C and total N in soil

compared to control, whereas application of vermicompost in increased soil pH. The direction of the change in soil pH as a result of treatment application reflected the initial pH of the amendment material. The increase in pH could be due to the higher pH value of the vermicompost (pH of 7.5) in relation to soil (pH of 6.5). However, this increase is not considered dangerous to soil quality because the values remained close to neutrality. The sensitivity of soil pH to the organic amendments was likely due in part to the low buffering capacity (Neilsen et al.,1998). The addition of vermicompost increased soil pH and pH increased with the higher dose of vermicompost. Contrary, Atiyeh et al. (2001) reported that the increase of vermicompost rate in the soil resulted in the decrease in soil pH. The production of NH_4^+, CO_2 and organic acids during microbial metabolism in vermicompost may be contributed to the decrease in soil pH (Albanell et al., 1988).

The addition of compost decreased soil pH. The obtained results confirm finding from Walker et al. (2003) that addition of compost to soil let to decrease soil pH. Smiciklas et al. (2002), Pattanayak et al. (2001) and Yaduvanshi (2001) also observed a decrease in soil pH after the use of organic materials. The production of organic acids (amino acid, glycine, cystein and humic acid) during mineralization (amminization and ammonification) of organic materials by heterotrophs and nitrification by autotrophs would have caused this decrease in soil pH.

Electrical conductivity (EC)

Electrical conductivity is a soil parameter that indicates indirectly the total concentration of soluble salts and is a direct measurement of salinity. Electrical conductivity showed an increasing trend with the application of vermicompost and compost to the soil (Figure 1). Incorporation of composts into soil increases the salt content as well as soil electrical conductivity, especially if high doses of compost are applied, because of the high salinity of composts (Gallardo-Lara and Nogales, 1987). The addition of compost and vermicompost to the soils led to slightly increase in EC values compared with control soil. The soils amended with vermicompost had higher EC than the untreated soils. The soil EC increased with increasing an application rate of vermicompost in soil as reported by Atiyeh et al. (2001) with pig manure. The EC of vermicompost depends on the raw materials used for vermicomposting and their ion concentration (Atiyeh et al., 2002).

Although EC of the soil increased in different treatments but the actual values did not cross the critical limit of 4.0 $dS.m^{-1}$. Such similar results have been reported in the literature (Sarwar et al., 2003; Niklasch and Joergensen, 2001; Selvakumari et al., 2000), which indicated that EC increased in acidic as well as alkaline soils when organic materials of different nature were applied to the soil**.** Similar results were found by Gonzalez et al. (2010).

Organic matter

Organic matter plays an important role in soil, because of its higher CEC and water holding capacities as well as its chelation ability and influence on soil stability. So, it is considered as a good resource of available elements. It improves soil structure, aeration and aggregation (Sparks, 1995).

Total organic carbon (TOC)

The increase of soil organic carbon with addition of organic amendments to soil is caused by high organic matter content of compost and vermicompost (Table l). The application of organic amendments led to a significant increase in organic carbon content, compared to its initial level (Table 2). The soils treated with compost and vermicompost exhibited a higher organic content than the control. Therefore, the addition of compost was able to affect the soil organic matter content, however, in contrast to previous data (Bohn et al., 1985; Giusquiani at al., 1995), our data demonstrates that the organic content increase was proportional to the compost dose used.

The organic content reached 3.99 and 9.36 % in result of adding of vermicompost at rate 5 $g.kg^{-1}$ and 10 $g.kg^{-1}$ respectively. Compost application led to increase organic content to 4.95 and 8.64 $g.kg^{-1}$ (Table 2). Both tested organic amendments had capacity to raise soil organic content (Figure 1), and there is no significant difference between these amendments, in spite of higher large organic matter content in compost (72.9 $g.kg^{-1}$). The use of organic amendments increases the soil organic carbon and improves soil structure. Fortuna et al. (2003) argued that the vermicompost amendment could increase the carbon contents up to 45 $g.kg^{-1}$ of the original levels, and thus contribute to increase the soil structural stability, particularly that of the macroaggregates.

Organic matter characteristics

Total extractable C, Humic and Fulvic acids, extracted with 0.1M $Na_4P_2O_7$+0.1N NaOH

Humus components were measured using the method proposed by Kononova and Belcikova (Kononova, 1966), by dividing the soil sample into several subsamples and by performing extractions with sodium pyrophosphate solution (0.1 M $Na_4P_2O_7 \times 10H_2O$, 0.1 N NaOH, pH $\approx$ 13). The solution based on sodium pyrophosphate is considered the best extractant for humic fractions. The resulting extract is used for measuring the total extractible carbon (TEC) and the carbon from huminic acids (HA), after removing the fulvic acids (FA). The total content of fulvic acids results from calculations.

The values for total extractable carbon extractable by 0.1M $Na_4P_2O_7$+0.1N NaOH (TEC) in control soil reached to 27.65 g.kg^{-1}. Application of organic amendments led to decrease TEC content to 19.28 and 19.08 g.kg^{-1} (Table 2). There is a correlation between the TEC in the samples and TOC (r=0.78) (Table 3).

Changes occurred in the content of humic acids and fulvic acids. Application of compost and vermicompost increases the content of humic acids and reducing fulvic acid compare to control.

Ratio of carbon of humic acids to carbon of fulvic acids. One of the major quality indicators of soil organic matter and its humified part is the ratio of carbon of humic acids to carbon of fulvic acids (C_{HA}:C_{FA}). The C_{HA}:C_{FA} ratio was calculated to determine the types of humus: humic (C_{HA}:C_{FA} >1.5), fulvic-humic (C_{HA}:C_{FA} 1.0-1.5), and humic-fulvic (C_{HA}:C_{FA} 0.5-1.0) (Grishina, 1986). The lowest values of the ratio C_{HA}/C_{FA} were registered in control soils (C_{HA}/C_{FA}=0.34). The C_{HA}:C_{FA} < 1 ratio showed that the bigger part of carbon was present in fulvic acids' composition compared with humic acids. In organic soils, this ratio is indicative of the maturity and stability of soil organic matter. A C_{HA}:C_{FA} ratio about or below 1.0 indicates a significant supply of plant debris, as fulvic acids are the first product of the humification process. Generally, a C_{HA}:C_{FA} ratio in excess of 1.0 is beneficial, indicating that organic matter is stable and mature, and that permanent bonds with the mineral soil phase dominate over mobile formations, which are easily transported inside the soil profile (Kononova, 1966).

Organic supplements influenced considerably the humus fractions by changing the ratio C_{HA}/C_{FA} in favour of the humic acids, in which changes of the type of humus from filvic type in control to humic-fulvic type in addition of 5 g.kg^{-1} and 10 g.kg^{-1} vermicompost and humic type in addition of 10 g.kg^{-1} compost. The changes in C_{HA}/C_{FA} ratio because of compost and vermicompost addition to soils are presented in Table 2. As expected, addition of compost to soils significantly increased the C_{HA}/C_{FA} ratio This is indicative of an increase in the carbon associated with the HA fraction. However, the increase was small in comparison with the large amount of

Table 2
Organic matter fractions according to Konova-Belichikova for organic amendments and treated soils

Object	Total C (TOC)	Extracted with 0.1N H_2SO_4	Extracted with a mixture of 0.1 N NaOH and 0.1M $Na_4P_2O_7$			Insoluble residue humin	C_{HA}/C_{FA}	Extracted with 0.1 N NaOH		D4/D6	Total N	C/N
		Aggressive fulvic acids	Total (TEC)	HA	FA			HA combined with R_2O_3	HA combined with Ca			
Compost C	72.9	1.017 1.08	4.91 21.92	3.85 5.29	1.05 1.44	1.606 72.349	3.67	2.689 61.997	38	10.4	1.49	48.9
Vermicompost (V)	38.58	0.829 2.15	3.77 9.77	2.28 5.91	1.49 3.86	34.81 90.23	1.53	0.945 4.54	95.46	8.3	1.67	23.1
Control soil	2.22	0.416 18.75	0.61 27.65	0.15 6.91	0.45 20.34	1.606 72.349	0.34	0.079 51.81	48.19	4.06	0.24	9.4
Soil + 5g.kg^{-1} V	3.99	0.168 4.22	0.94 23.73	0.39 9.89	0.55 13.85	3.04 76.26	0.71	0.259 65.77	34.23	4.65	0.46	8.7
Soil + 10 g.kg^{-1} V	9.36	0.269 2.88	1.8 19.28	1.02 10.89	0.785 8.39	9.36 80.72	1.3	0.66 64.9	35.1	5.83	0.82	11.4
Soil + 5g.kg^{-1} C	4.95	0.185 3.74	1.47 29.76	0.83 16.83	0.64 12.93	3.476 70.23	1.3	0.54 65.22	34.78	5.77	0.54	9.2
Soil + 10g/kg^{-1} C	8.64	0.271 3.13	1.649 19.08	1.096 12.689	0.55 6.39	6.99 80.92	1.98	0.82 75.18	24.82	6.19	0.68	12.7

TOC – total organic carbon (% from soil mass); TEC – total extractible carbon (% from soil mass / % from TOC);
HA – carbon from huminic acids (% from soil mass / % from TOC / % from TEC);
FA – carbon from fulvic acids (% from soil mass / % from TOC / % from TEC);
Humines (% from soil mass / % from TOC); Aggressive FA (% from soil mass / % from FA)

Table 3

Linear correlation coefficients among pH, the soil organic content, humus fractions and available Pb, Cu, Zn and Cd

	pH	TOC	FA aggr,	TEC (pyr)	HA (pyr)	FA (pyr)	Humin	C_{HA}/C_{FA}	HA R_2O_3	HA Ca	D4/D6	N	C/N	Pb	Cu	Zn	Cd	Fe	Mn	K	Ca	Mg	P
pH	1																						
TOC	ns	1																					
FA aggr,	ns	0.98	1																				
TEC(pyr)	0.54	0.78	0.65	1																			
HA (pyr)	0.87	ns	ns	0.71	1																		
FA (pyr)	ns	0.94	0.9	0.8	ns	1																	
Humin	0.54	0.78	0.65	0.99	0.71	0.8	1																
C_{HA}/C_{FA}	0.52	0.7	0.72	0.4	0.35	0.85	0.4	1															
HA R_2O_3	0.42	0.42	0.34	0.5	ns	0.7	0.5	0.81	1														
HA Ca	0.85	ns	ns	ns	0.51	0.36	ns	0.74	0.83	1													
D4/D6	0.46	0.76	0.83	0.27	0.48	0.79	ns	0.92	0.52	0.52	1												
N	0.31	0.97	0.98	0.69	ns	0.83	0.69	0.55	ns	ns	0.69	1											
C/N	ns	0.92	0.88	0.78	ns	0.99	0.78	0.87	0.73	0.41	0.8	0.8	1										
Pb	0.74	0.47	0.5	0.33	0.36	ns	ns	ns	0.58	0.83	ns	0.67	ns	1									
Cu	ns	0.52	0.67	ns	0.42	ns	ns	ns	0.34	0.26	0.62	0.67	0.26	0.67	1								
Zn	0.96	ns	ns	0.37	0.71	ns	0.37	0.58	0.62	0.95	0.41	ns	ns	0.87	0.21	1							
Cd	0.91	0.47	0.44	0.54	0.65	ns	0.54	0.3	0.46	0.84	ns	0.4	ns	0.94	0.4	0.95	1						
Fe	0.21	0.62	0.53	0.7	0.16	0.85	0.7	0.82	0.96	0.67	0.57	0.22	0.87	0.35	0.22	40	0.2	1					
Mn	0.95	0.49	0.41	0.68	0.79	0.27	0.68	0.26	0.3	0.77	ns	0.21	0.22	0.84	0.21	0.93	0.97	ns	1				
K	0.76	0.63	0.63	0.52	0.45	0.34	0.52	ns	0.4	0.74	ns	0.62	0.29	0.97	0.62	0.84	0.96	ns	0.9	1			
Ca	0.9	0.59	0.53	0.68	0.7	0.35	0.68	ns	0.29	0.74	ns	0.35	0.3	0.89	0.35	0.9	0.98	ns	0.99	0.95	1		
Mg	0.93	0.54	0.47	0.68	0.75	0.31	0.68	0.21	0.3	0.76	ns	0.28	0.26	0.87	0.28	0.92	0.98	ns	0.99	0.93	0.99	1	
P	0.56	0.91	0.88	0.76	0.4	0.73	0.76	0.33	ns	0.37	0.47	0.58	0.69	0.79	0.58	0.56	0.8	0.3	0.79	0.89	0.87	0.83	1

TOC – total organic carbon, FA aggr,- aggressive FA, extracted with 0,1N H2SO4, HA (pyr) – extracted with a mixture of 0,1 N NaOH and 0,1M Na4P2O7, FA (pyr) - extracted with a mixture of 0,1 N NaOH and 0,1M Na4P2O7, TEC- extracted with a mixture of 0,1 N NaOH and 0,1M Na4P2O7, HA R2O3 - combined with R2O3, HA Ca – combined with Ca

ns-not significant

organic matter applied. The small increase was in keeping with the quality of the compost used (Table 1). The compost had a high content of total carbon (72.9 g.kg^{-1}), but only (< 10 g.kg^{-1}) of which was associated with humus like substances (C_{HA} and C_{FA}) extractable using NaOH (Wu, 2001).

Knowing the quantity of humic substances in the soil is important because humic materials have a relatively high content of free radicals which play important roles in polymerisation and redox reactions. This affects the mobility of metals (both those with nutrient value and those that are of concern because they are pollutants), because cations with estimated reduction potentials of 0.5–0.7 eV are easily reduced by these radicals (Skogerboe and Wilson, 1981).

Insoluble residues – Humin

Part of soil organic matter was not extracted because heterogeneous organic compounds, called humins, are resistant to chemical solvents. Humins are an insoluble part of humus, which is tightly bound to clay minerals, or colloidal iron hydroxide and aluminum hydroxide. In the control soils most of the carbon was accumulated in insoluble humus compounds (humins and particulate organic matter). Addition of organic amendments led to an increse of the quantity of the humin, from 72.3 g.kg^{-1} in control to 80.72 - 80.92 g.kg^{-1} in soils with 10 g.kg^{-1} vermicompost and 10 g.kg^{-1} compost addition (Table 2). Humin content was positively correlated with total nitrogen (r=0.69) and TOC content (r=0.78) (Table 3).

In addition to specific humus compounds (humic acids, fulvic acids, and humins), organic matter also comprises non-specific compounds (carbohydrates, amino acids, lignin, tannins, and others) that may not be extractable and are contained in humins (Simpson et al., 2007). In contrast to specific humus compounds, some non-specific humus compounds are susceptible to rapid microbial decomposition (Stevenson, 1994).

The content of humic compounds in TOC is also defined by the degree of humification, expressed as the sum of humic and fulvic acids in TOC (Kononova, 1966). With the progression of humification in organic soils, nitrogen is accumulated in the complex humic compounds in the humin fraction (Okruszko, 1993).

HA extracted with 0.1 N NaOH

Application of the compost and vermicompost to soils led to increase content of the free humic acids and those bound to mobile R_2O_3. Application of compost and vermicompost led to increase the humic acid bound to R_2O_3 from 51.81 g.kg^{-1} in control soil to 64.90 g.kg^{-1} in soils amended with 10 g.kg^{-1} vermicompost and 75.18 g.kg^{-1} in soils amended with 10 g.kg^{-1} compost (Table 2). Application of compost and vermicompost led to decrease the organic component mobility and most of the organic matter components bound very hard to minerals.

Changes in the content of humic acids bound to Ca^{2+} were found. Application of compost and vermicompost led to decrease the humic acids bound to Ca^{2+} from 48.19 g.kg^{-1} (control soil) to 35.10 g.kg^{-1} and 24.82 g.kg^{-1} after compost and vermicompost application (Table 2).

FA extracted with 0.1 N H_2SO_4

Organic carbon extracted by 0.1 N H_2SO_4 (the most mobile and low molecular fraction of the organic matter) is presented in moderate values (18.75 g.kg^{-1}) in control soil and could be able to influence the organic components mobility. Application of the compost and vermicompost reduces the amount of aggressive fulvic acids to 2.88 and 3.13 g.kg^{-1} (Table 2).

It is well known that free and/or HA bonded to three valence cations (fraction 0.1 N NaON) and acid fraction (aggressive fulvic acids, 0.1 N H_2SO_4 extract) are organic matter components which pose a potential environmental risk. So, their lower content is a precondition for the low potential environmental risk of the soil with organic amendment studied.

D4/D6 ratio

The coefficient of colour Q, introduced by W. Shpringer, was widely used for evaluating the character of the humus substances' colour. It was equal to the ratio of the optical densities of two wave lengths with sufficiently intensive maximum of absorption - 465 and 665 nm: $Q = \lambda_{465} / \lambda_{665}$ (Tomov and Artinova, 2005). The D4/D6 ratio of the HA fraction increased significantly as a consequence of the compost and vermicompost application (Table 2). Larger values of D4/D6 ratio have been associated with the presence of smaller size organic molecules or more aliphatic structures and usually with higher content of functional groups (Stevenson, 1994). It signifies the increase of carbon linked to HA, which was confirmed through the C_{HA}/C_{FA} analysis. There was positive correlation coefficient of 0.92 between C_{HA}/C_{FA} and D4/D6 ratios. The presence of smaller sized, highly aliphatic molecules suggested that the organic matter fraction was more reactive and soluble through a wider pH range, characteristics often associated with the FA fraction of soil organic matter. Numerous researchers have shown the FA fraction of organic matter to be associated more with metal leaching than the HA fraction (Gaffney et al., 1996; Chirenje et al., 2002a,b). Humic acids tend to be more aromatic and more prone to precipitation under the acidic conditions common in many soils, making them less mobile. Therefore, it could be argued that, while the FA fraction makes organic matter more reactive, it may also lead to greater leaching of both cationic nutrients and pollutants.

The increase of the optical density was correlated with the level of hydrolisability of the humic acids and more precisely with the participation of inhydrolisable residium in the molecule. A dependence between the correlation of the most important fractions in the humic acids' molecule and their colour was revealed. The "more mature" humic acids had the highest values of optical density. The aromatic nuclei, carriers of carboxylic groups, prevailed in their structure (Tomov and Artinova, 2005).

Soil C/N ratio

The soil C/N ratio is often used to explain different turnover rates for early residue decomposition.The C/N ratios of the soils are narrow. N can be easily mineralised when the C/N ratio is less than 20:1. According to Mikkelsen and Hartz (2008) the C/N ratio of added organic materials is a good, but not an absolute, predictor of whether N immobilization is likely (C/N ratio > 25:1) or if mineralization is likely (C/N ratio < 20:1). In our study, the mean of C/N of soil was narrow (9.41 -12.67), below 20 in all soils (Table 2).

Effect of compost and vermicompost on soil macronutrients (N, P, K, Ca and Mg)

Application of compost and vermicompost increased the amounts of macro- and micro nutrients in the soil (Figures 1 and 2). The increases were significant for both amendments, especially for 10 g.kg^{-1} treatment.

Total N

The changes in the organic content in soils brought about changes in the total nitrogen content (Figure 1). Our calculations showed that there was a strong positive correlation between the total organic content (TOC) and the total nitrogen content with value of the correlation coefficient r=0.97 (Table 3).

The results showed that the total N concentration in soil was significantly affected by compost and vermicompost treatments. The soils treated with vermicompost at the rate of 10 g.kg^{-1} had more total N compared to soils without vermicompost application. Vermicompost might have produ-ced more residual N in soil than those in control plots. There have been other reports of increase of N in soil after application of vermicompost (Nethra et al., 1999). Remarkable amount of N is available for plant because of organic matter, acidic pH and proper moisture in soil (Zupanc and Zupancic, 2010).

P

The content of phosphorus in the soil is influenced by applications of organic amendments (Figure 2). The lowest content of mobile phosphorus in the control soil (21.05 mg/kg) was found.

There was a significant increase in the soil extractable phosphorus with the increase of the compost and vermicompost doses applied. Soils treated with vermicompost at the rate of 10 g.kg^{-1} had significantly more P as compared to control plots. Vermicompost amendments could help to recovering the nutrient contents.This implied that the continuous inputs of P to the soil were probably from slow release from vermicompost and release of P was due largely to the activity of soil microorganisms (Arancon et al., 2006). Marinari et al. (2000) showed similar increases in soil P after application of organic amendments. The enhancement of phosphatase activity and physical breakdown of material resulted in greater mineralization (Sharpley and Syres, 1997).

The data obtained in our experiment agree with those of numerous studies in which the vermicompost applied increases the concentration of soil P (Padmavathiamma et al., 2008). Devliegher and Verstraete (1997) found a significant increase in the P contents after the vermicompost amendment, reaching the double of the initial value for some treatments, and even the triple in others, attributing the results to increases in the enzymatic activity of phosphatases from earthworms. The increase of DTPA - extractable P with vermicompost may be attributed to the release of humic acid during organic matter decomposition, results in a convert's unavailable soil phosphate into available forms. Higher positive correlation (r = 0.91) that existed between available P and organic C was an indication of organic matter suppressing P fixation into soils (Afif *et al.,* 1993) just as an increase in soil pH further complimented the effects of organic matter. Because of oxidation and organic matter degradation in soil a lot of nutrients like P, are going to be available to plant (Gallardo-Lara and Nogales, 1987). Applying compost to soil increases soil available P and similar results has reported by other researchers (Gallardo-Lara and Nogales, 1987).

DTPA extractable P was also significantly related to DTPA-extractable Ca (r = 0.87) and Mg (r = 0.83) (Table 3).

K

The DTPA- extractable potassium (K) were increased by the application of vermicompost which may be attributed to the vermicompost and compost which have high K contents (Table 1). Significantly, higher values of available K were obtained after the introduction of vermicompost compared to compost (Figure 2). The addition of organic matter in the form of mulch, vermicompost and the addition of earthworm casts cause the increase of the biovailability of elements (Germida and Siciliano, 2000).The selective feeding of earthworm on organically rich substances which breakdown during passage through the gun, biological grinding, together with enzymatic influence on finer soil particles,

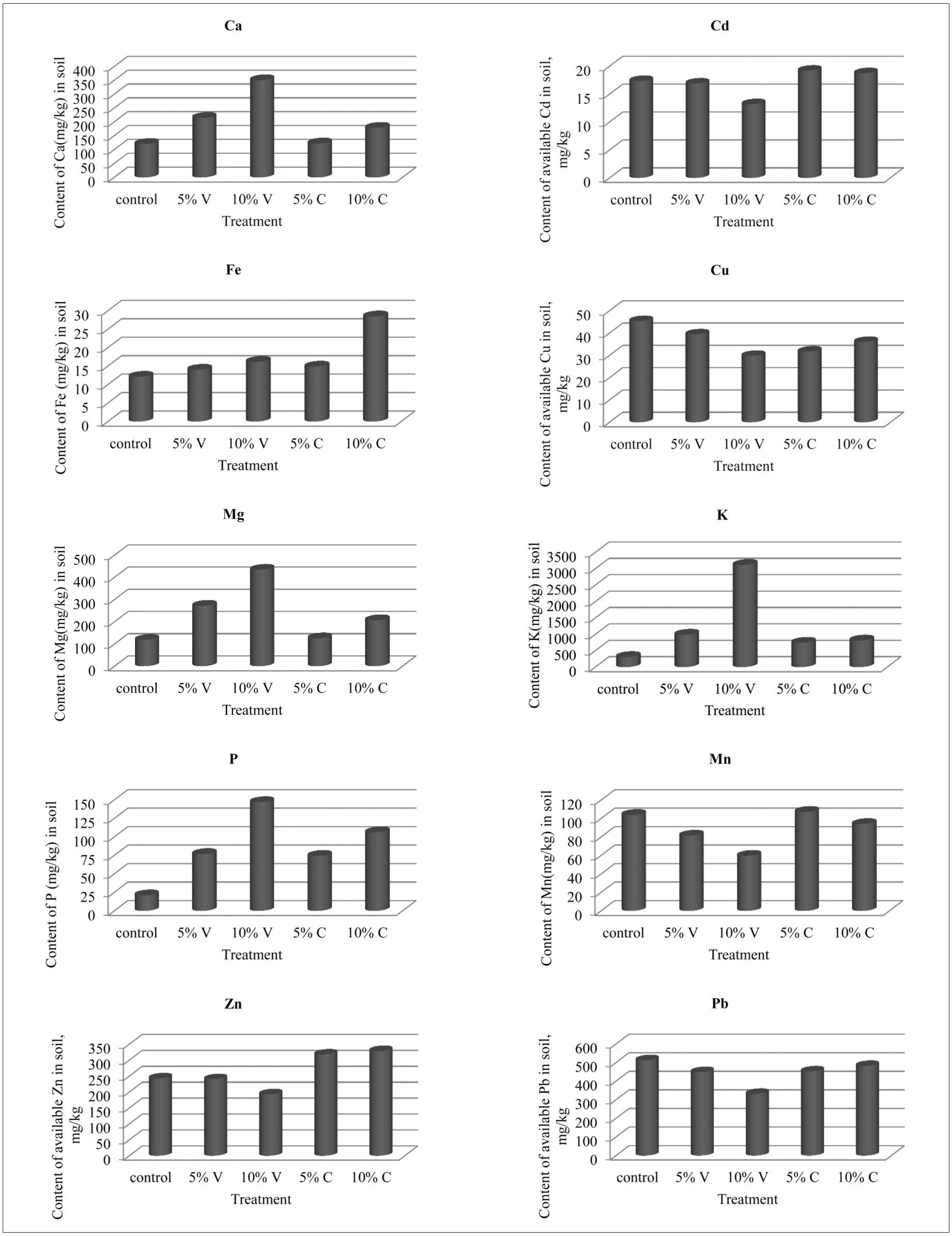

Fig. 2. Effect of different organic amendments (compost (C) and vermicompost (V)) applications to soil on availability of heavy metals, micro and macroelements

were likely responsible for increasing the different forms of K (Rao et al., 1996). The increase of soil organic matter resulted in decrease K fixation and subsequent increase K availability (Olk et al., 1993). Swarup and Yaduvanshi (2000), Singh et al. (2001), Khoshgoftarmanesh and Kalbasi (2002), Singh al. (2002) and Verma et al., (2005) reported that prolonged use of mineral fertilizers, manure, compost and other meliorants increases the potassium content in the soil. Because of high amount of K in organic amendments that increases CEC, the K amount rises in soil. Those results agree with results of other researchers (Khoshgoftarmanesh and Kalbasi, 2002).

Like P, DTPA-extractable K were correlated significantly with TOC (r=0.63) (Table 3).

Ca and Mg

From Figure 2, it can be seen that compost and vermicompost treatments significantly raised soil available Ca and Mg content. The increase of soluble calcium and magnesium content with the addition of vermicompost and compost to soil is caused by high calcium and magnesium content of vermicompost and compost (Table 1). The highest increase was observed after addition of vermicompost. Vermicompost contains most nutrients in plant available forms such as phosphates, exchangeable calcium and soluble potassium (Orozco et al., 1996)

Similar to DTPA-extractable P, DTPA-extractable Ca generally increased after compost and vermicompost application. DTPA-extractable Ca was significantly correlated with TOC (r = 0.59) (Table 3).

Trends in Mg were similar to Ca for DTPA extractions. Increasing the compost and vermicompost application rate increased DTPA-extractable Mg (Figure 2). DTPA-extractable Mg was significantly related to TOC (r = 0.54) (Table 3).

Increase in soil pH was accompanied by an increase in available Ca and Mg; hence, the high positive coefficient of correlation between exchangeable Ca and soil pH (r=0.90) and exchangeable Mg and soil pH (r=0.93). A similar study with animal manure showed significant improvement in soil pH, which increased as the values for exchangeable Ca and Mg increased (Ano and Agwu, 2005).

Sorption of dissolved organic carbon (DOC) and formation of DOC cation complexes may be pH-dependent (Temminghoff et al., 1997). As soil pH increases above neutrality, DOC sorption to soil becomes weaker and potential for formation of DOC-cation complexes increases (Romkens et al., 1996). For the high pH soil after vermicompost addition, vermicompost likely promoted the formation of DOC-cation complexes, subsequently increasing macronutrient mobility.

Effect of compost and vermicompost on soil micronutrients (Fe and Mn)

The high amount of organic matter in compost, its oxidation and degradation and neutral pH increase micronutrients availability such as Fe, Mn, Zn and Cu in soil (Gallardo-Lara and Nogales, 1987). Similar findings have been reported (Antoniadis and Alloway, 2003).

The Mn availability in the soil was significantly affected by vermicompost treatments (Figure 2). Mn decreased with the rate of amendments. The application of vermicompost decreased the available Mn concentration, as compared with the control. Contraty results were obtained from Jordao et al. (2006). According to Jordao et al. (2006) high content of extracted Mn with DTPA can be due to the dissolution of Mn precipitates (carbonates, hydroxides and phosphate) caused by microbial activity that changes soil pH and gaseous composition.

The Fe availability in the soil was significantly affected by treatments (Figure 2). Fe increased with the rate of amendments. The application of vermicompost increased the available Mn concentration to 28.45 mg.kg^{-1}, as compared with the control (12.1 mg.kg^{-1}).

Effect of compost and vermicompost on soil heavy metals (Pb, Cd, Zn and Cu)

In Figure 2 are presents the results for quantities of mobile forms of DTPA extracted Pb, Cd, Cu and Zn from naturally contaminated soil from the region of Plovdiv and their change after adding organic soil amendments. An important factor influencing the mobility of Pb, Cu, Cd and Zn is the quantity of organic matter in the soil (Ross, 1994; McGrath et al., 1998, 2000). Many authors have found that soils with high organic carbon content, as well as adding organic fertilizer cadmium content in soil decreases. This effect is explained by the high cation exchange capacity of organic matter and its ability to form chelate complexes with Cd. Haghiri (1974) found that reducing the cadmium content of plants by increasing the amount of imported organic meliorants due to higher cation exchange capacity of the soil.

Organic amendments affect DTPA-extractable heavy metals. The results show that organic supplements affect differently on the amount of mobile zinc. Quantities extracted with DTPA mobile forms of zinc increases with the amount rate of the compost to soils. Application of vermicompost reduces the amount of available Zn to 193.1 mg.kg^{-1}, while the addition of compost led to its increase to 328. 8 mg.kg^{-1}.

The total Zn content, pH, organic matter, adsorption sites and microbial activity of the soil affect the Zn availability (Alloway, 1995). The soil pH is the most important factor controlling Zn availability, which decreases with the increase of the pH (Shuman, 1999). In this experiment increase Zn availability after compost application was attributed to the pH reduction and the greater organic matter degradation. Shuman (1999) found that retention of Zn in the soil increased in the presence of organic fertilizers. Zn can form insoluble compounds - precipitates during the mineralization of organic meliorants (Walker et al., 2003) and insoluble compounds in the form of $ZnCO_3$ in calcareous soils (Usman et al., 2004). Shuman (1999) found that pH, clay content and organic matter and cation-exchange capacity influence the adsorption of Zn in soils. Mandal and Hazra (1997) found that the addition of organic amendments and lower soil pH leads to an increase in the amount of available Zn.

A correlation between the content of mobile zinc and soil pH and between mobile zinc and organic matter in the soil was found (Table 3).

Similar are the results obtained with respect to cadmium. Application of vermicopmost reduced the amount of DTPA-extractable mobile forms of Cd from the soil, while compost can even increase Cd to 18.8 -19.2 mg/kg. Lowering the pH caused metal ions to be more soluble and thus more available to the plant which includes both micronutrients and heavy metals that are considered pollutants and also plants and animals poisons (Khoshgoftarmanesh and Kalbasi, 2002).

Compost may increases heavy metals amount in soil as it increases essential nutrients amount in soil. Increased heavy metals amount such as Pb, Cd and Ni by compost have been reported (Ben Achiba et al., 2009). Similar results were obtained from Karaca (2004) on the application of mushroom compost and grape marc. Reducing extracted with DTPA mobile forms of Cd may be due to the high cation exchange capacity of organic matter and the ability to connect Cd from the soil. Korcak and Fanning (1985) found a positive correlation between DTPA-extractable mobile forms of Cd and quantity of ogranic matter content in the soil.

Downward trend in mobile forms by the action of soil amendmenrs is expressed more clearly in terms of Pb and Cu. In most cases, mobile forms of Pb and Cu diminished. Organic supplements lead to lower content of DTPA extractable Cu. The reduction of Cu content may be due to the transformation of organic matter in a stable form that could link more Cu.

Humic acids from organic amendments tend to form complexes that are different for each metal and depend on soil conditions such as pH, cation exchange capacity and clay mineral fraction (Barancikova and Macovnikova, 2003). Organic matter not only forms complexes with these metals but it also retains them in exchangeable forms, affecting each metal differently. Some metals are bound and rendered unavailable while others are bound and readily available (Kononova, 1966; Stevenson, 1994). The results of the present study indicated that enrichment of soil with organic matter could reduce the content of bioavailable metal species as a result of complexation of free ions of heavy metals. This is indicative of heavy metals immobilisation by humic substances from compost and vermicompost application. Results appear that verify the function of humic acid in improving phytoremediation efficiency of soils contaminated with heavy metals and potential environmental availability of metals may be controlled by soil organic amendments.

Conclusions

Soil physical properties, such as hydraulic conductivity are positively affected with the application of tested organic amendments. Soil chemical properties and nutrient availability, are substantially improved in soils receiving organic amendments. Compared with the unamended soil, soil treated with organic amendments showed apparent increases of organic matter, total N, EC, available macroelements (P, K, Ca and Mg).

Compost and vermicopmost application increases the soil organic content. It is important to determine other parameters in order to better understand the effects of compost and vermicompost on soil organic matter quality. These parameters provide information about soil organic matter component evolution after compost and vermicompost application. For example, knowing the ratios of HA to FA helps us predict the possible reactions of the organic matter fraction with heavy metals because FAs (more aliphatic and richer in carboxiylic acid, phenolic and quinone groups) are more soluble and reactive than HAs (more aromatic and insoluble when carboxylate groups are protonated at low pH).

Organic amendments influenced considerably the humus fractions by changing the ratio C_{HA}/C_{FA} in favour of the humic acids, in which changes of the type of humus from filvic type in control to humic-fulvic type in addition of 5 $g.kg^{-1}$ and 10 $g.kg^{-1}$ vermicompost and humic type in addition of 10 $g.kg^{-1}$ compost.

Application of the compost and vermicompost reduces the amount of aggressive fulvic acids, organic matter components which pose a potential environmental risk. So, their lower content is a precondition for the low potential environmental risk of the soil treated with organic amendment.

The results of the present study indicated that soil application of compost and vermicompost in most cases decreased DTPA-extractable levels of heavy metals in the soil. This is indicative of heavy metals immobilisation by humic substances from compost and vermicompost application. Results appear that verify the function of humic acid in improving phytoremediation efficiency of soils contaminated with heavy metals and potential environmental availability of metals may be controlled by soil organic amendments.

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