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Heavy Metals Remediation from Maize (*Zea mays*) Crop by the Use of Vermicomposts Through Vermicomposting by *Eisenia fetida*

Deepak Kumar Bhartiya and Keshav Singh

Department of Zoology, D.D.U. Gorakhpur University, Gorakhpur-273009, U.P. India

Abstract: The aim of the present study to investigate the heavy metals (Co, Cr, Pb, Ni, Cd and As) concentration were observed in different animal dung, earthworm (*Eisenia fetida*) body before and after vermicomposting, soil as well as maize grain (*Zea mays*). There was significantly increase in the level of heavy metals observed in all the animal dung as well as soil. The accumulation of heavy metals was observed during the vermicomposting by earthworm *E. fetida*. The concentrations of heavy metals were observed in soil, soil with vermicompost during inoculation of earthworm in each cultured maize crop to experimented field soil. The Cr and Cd were observed maximum decreased after harvesting of maize crop in the combination of soil + goat dung (8.186±0.003 mg/kg) and soil + sheep dung (1.187±0.005 mg/kg). The heavy metals concentrations were observed in maize grains after harvesting of crop. The Co and Ni were observed in the maximum decreased the level of (BDL and 1.145±0.003 mg/kg) in the combinations of soil + cow dung and soil + sheep dung. The concentrations of heavy metals were observed in earthworm body in soil and soil with vermicompost of different animal dung during before and after harvesting of crop. The maximum concentration of Cr and Cd were observed in the combination of soil + cow dung and soil + buffalo dung the level of (104.134±0.005 and 41.323±0.007 mg/kg). These metals exhibit toxic effects on maize (*Zea mays*) plant contain due to highest amounts of heavy metals concentrations from polluted soil. There was observed remediate the concentration of heavy metals (from the toxic level) to maize seeds by the use of vermicompost and *E. fetida* from experimented field soil.

Key words: Animal dung · Accumulation · *Eisenia fetida* · Flame atomic absorption · Heavy metals · vermicomposting · Maize (*Zea mays*) and human health problems

INTRODUCTION

Abundant use of chemical fertilizer and pesticides have made of our soil sick and problematic [1-2] and also increased heavy metals in the soil [3]. Physico-chemical properties of soil are the main factors affecting by the mobility, speciation and hence availability of toxicity heavy metals [4]. Nannipieri and Badalucco [5] reported that soil is a best complicated heterogeneous system, constituted of soil organic matter, minerals, plants, microbes, faunas and toxic metals. Better growth and productivity of crop by the used of phosphate fertilizer are responsible for Cd increased in soil [6]. Bohn *et al.* [7] reported that toxic metals are present in soil to various physical and chemical forms it's easily communications to mobility and biological availability. Improvement of the soil fertility and crop production by the use of animal manure is a common practice of developing countries [8-9]. The more industrialization, advanced human activity, rapid growing population to need the food the former used agricultural amendments, chemical fertilizer and lime product to better growth and productivity therefore its soils are contaminated much heavy metals [10].

A group of elements that have density higher than about 5gm/cm^3 it term refers the heavy metals [11-12]. Heavy metals entered in the human bodies by the ingestion of foodstuffs specially grain, cereals and leafy vegetable one of the major environmental and health problems of our modern society [13-14]. It can cause several respiratory irritation lung disease, cancers, kidney problem, brain damage, heart attack, anemia, chronic

Corresponding Author: Keshav Singh, Department of Zoology, D.D.U. Gorakhpur University, Gorakhpur-273009, U.P. India. Mob: +09450433313,

nephritis, encephalopathy and disease in digestive system [15]. Heavy metals present in environment to naturally by erosion of rocks, volcanic activity, forest fire, artificially by industries, paper mills, vehicles, human activities and it can release in large quantities directly affect the flora, fauna as well as human health [16].

Brown [17] reported that cobalt (Co) can be responsible for the beer hearth syndrome in the human and high level of Pb can caused damage to brain and kidney which ultimately leads to death, pregnant woman cause miscarriage, in the men can damage the organ responsible for sperm production. Chromium (Cr) spreads the disease in human being breathing problem such as asthma, cough wheezing and skin contact can causes skin ulcer and arsenic (As) can cause several disease like skin, lung and liver cancer and lower level of exposure causes nausea and vomiting, decreased production of RBCs, WBCs and damage to blood vessels [16]. The human and animal bodies particularly accumulate heavy metals in kidney and liver it can causes serious problems to the health [15].

Earthworms *Eisenia fetida* are important link in the food chain and they can accumulate the hazardous elements from the soil [18-19]. Heavy metals accumulate by the *E. fetida* in their body tissue from soil and different biological wastes during vermicomposting [20-24]. *E. fetida* and *Eudrilus eugeniae* are reducing of the metal toxicity from municipal solid wastes during vermicomposting [25]. The use of redworm *E. fetida* converted more rich nutrients for plants from sewage sludge and biological wastes vermicomposting [26].

The aim of the present study investigated that the use of vermicompost of different animal dung and accumulation of heavy metals from soil by earthworm *E. fetida* was significantly decreased heavy metals concentration in maize seed.

MATERIALS AND METHODS

Collection of Live Stock Excreta: Different animal wastes (buffalo, cow, goat, horse and sheep dung) were collected from different farm houses of Gorakhpur city and exposed to sun light for 5 to 10 days for removal of various harmful organism and noxious gases.

Collection of Earthworm: Red earthworm *E. fetida* an epigeic species cultured in laboratory condition. Collect the *E. fetida* from Vermiculture Research Center, Department of Zoology, D. D. U. Gorakhpur University, Gorakhpur.

Experimental Setup for Vermicomposting: Gupta [27] reported that vermicompost obtain through the vermic activity by the use of (*E. foetida*). It is conducted on cemented earth surface. The sizes of each vermibeds were 3m × 1m × 9cm. *E. fetida* inoculated 2kg cultured earthworms in each bed. The beds were covered by the jute pocket and moisture maintain up to 40-50 days by the use of tap water for the proper growth and development of earthworm become room temperature (20-30°C) were maintained aeration and moisture (40-60%). Each bed manually turned up to 3 weeks after one week interval granular tea like vermicompost appear on the surface on the each bed after 55-60 days.

Experimental Setup for Crop Production: The seed of maize were sown in the cultured experimented field of 1 m^2 area. Vermicomposts obtain from different animal dung along with earthworm (*E. fetida*) were mixed in each squires at the rate of 2 kg/m^2. Each control group of squire area was not mixed vermicompost.

Analysis of Heavy Metals

In Vermicompost, Maize Grains and Soil: The concentration heavy metal in the initial and final vermicompost, maize grains and soil were measured by the method described by Maboeta [28]. About 1 gm of initial feed mixture and final vermicompost, soil and maize seed were subjected to digestion by adding excess of nitric acid (1:1) and were placed on hot plate and heated for 4 hours at 90 to 100°C. Attentions were taken to ensure that sample did not dry out during digestion. After digestion sample will be poured into 100 ml flask through Whatman No 41 filter paper and injected into flame atomic absorption for determination concentration of the heavy metals.

In Earthworm Body: The heavy metals in the earthworm body tissue were digested using by the method of Katz and Jenneis [29]. Earthworm were individually dried, ground and burned to ash at high temperature. The ash will be placed in a test tube about 10 to 15 ml of 55% nitric acid have added in it. Solution will be left for 12 hrs at room temperature and heated again a temperature of 40 to 60°C for 2 hrs and then at a temperature of 120 to 130°C for one hrs. Reheated the sample at 120 to 130°C and 1 ml of 70% perchloric acid will be added. The sample have allowed to cooling before adding 5 ml of distilled water. Samples have again reheated up to 130°C until white fumes emitted and allowed to cool finally before being micro filtered. Filtered the solution through Whatman

No 41 filter paper into 100 ml flasks. This solution was taken for measurement of the heavy metals content in earthworm body by flame atomic absorption.

Statistical Analysis: All the data are mean ±SD of 6 replicates. Students't' test was applied to determine the significant ($P<0.05$) difference between initial and final vermicompost, earthworm (*E. fetida*) body during inoculation and after vermicompost, before and after harvesting of maize crop and earthworm body by the method of Sokal and Rohlf [30].

RESULTS AND DISCUSSIONS

The maximum decrease the concentration of heavy metals Co, Cr and As were observed in sheep dung to final vermicomposting by earthworm (*E. fetida*) the level of (BDL, 0.043 ± 0.002 mg/kg and BDL). Pb and Ni were observed the level of (BDL and 0.027±0.003 mg/kg) in goat dung, whereas Cd was observed in horse dung in the level of (0.023±0.002 mg/kg) (Tables 1 and 2).

The heavy metals concentration was observed in earthworm body after vermicomposting of different animal dung. The maximum amount of heavy metals such as (Co, Cr, Pb, Ni and As) were accumulated from buffalo dung during vermicomposting (4.358±0.003, 103.015±0.004, 11.369±0.004, 5.249±0.005 and 7.662±0.003 mg/kg), whereas Cd was observed from the goat dung vermicompost (35.994±0.003 mg/kg) (Table 3). Bhartiya and Singh [31] reported that Cr was significantly increased in the earthworm body obtain from the combination of buffalo dung control during vermicomposting (155.187±0.002 mg/kg). The heavy metals accumulation by the *E. fetida* in their body tissue from soil and different biological wastes during vermicomposting [20-24]. Sellanduria *et al.* [25] reported that the *Eisenia fetida* and *Eudrilus eugeniae* are reducing of the metal toxicity from municipal solid wastes during vermicomposting.

The concentrations of heavy metals were observed in soil, soil with vermicompost and during inoculation of earthworms in crop field. The heavy metals concentration

Table 1: Concentration of heavy metals (mg/kg) of different animal dung in initial feed mixture and final vermicompost

	Heavy metals (mg/kg)					
	Co		Cr		Pb	
Vermicompost	Initial	Final	Initial	Final	Initial	Final
Buffalo Dung	0.266±0.003	0.078±0.003*	0.818±0.002	0.121±0.003*	0.376±0.004	0.103±0.004*
Cow Dung	0.152±0.004	0.045±0.002*	0.611±0.003	0.064±0.003*	0.128±0.002	0.039±0.002*
Goat Dung	0.122±0.002	0.032±0.003 *	0.508±0.003	0.048±0.004*	0.102±0.003	BDL*
Horse Dung	0.234±0.005	0.092±0.004*	0.686±0.005	0.081±0.002*	0.255±0.002	0.054±0.003*
Sheep Dung	0.057±0.002	BDL*	0.527±0.003	0.043±0.002*	0.172±0.003	0.039±0.003*

BDL= below detectable limit

Each value is the Mean ± SD of six replicates.

Significant P<0.05 "t" test between initial feed mixture and final vermicompost

Table 2: Concentration of heavy metals (mg/kg) of different animal dung in initial feed mixture and final vermicompost

	Heavy metals (mg/kg)					
	Ni		Cd		As	
Vermicompost	Initial	Final	Initial	Final	Initial	Final
Buffalo Dung	1.208±0.003	0.197±0.004*	0.282±0.006	0.043±0.003*	0.321±0.003	0.042±0.005*
Cow Dung	0.852±0.002	0.102±0.005*	0.734±0.005	0.188±0.002*	0.183±0.003	0.055±0.003*
Goat Dung	0.122±0.003	0.027±0.003*	0.509±0.001	0.102±0.004*	0.246±0.002	0.068±0.003*
Horse Dung	0.634±0.005	0.069±0.004*	0.104±0.003	0.023±0.002*	0.302±0.005	0.073±0.006*
Sheep Dung	0.210±0.005	0.035±0.003*	0.182±0.004	0.031±0.003*	0.109±0.003	BDL *

BDL= below detectable limit

Each value is the Mean ± SD of six replicates

Significant P<0.05 "t" test between initial feed mixture and final vermicompost

Table 3: Concentration of heavy metals (mg/kg) in earthworm (*E. fetida*) body after vermicomposting of different animal dung

	Heavy metals (mg/kg) in earthworm body					
Vermicompost	Co	Cr	Pb	Ni	Cd	As
During inoculation						
Earthworm body (Control)	4.265±0.003	102.408± 0.005	11.142±0.003	4.346±0.003	35.645±0.002	7.450±0.004
After vermicomposting						
Buffalo dung	4.358±0.003*	103.015±0.004*	11.369±0.004*	5.249±0.005*	35.805±0.003*	7.662±0.003*
Cow dung	4.325±0.002*	102.809±0.002*	11.211±0.003*	5.125±0.003*	36.053±0.002*	7.581±0.002*
Goat dung	4.312±0.004*	102.765±0.003*	11.203±0.005*	4.432±0.005*	35.994±0.003*	7.603±0.005*
Horse dung	4.357±0.003*	102.784±0.003*	11.342±0.003*	4.735±0.003*	35.701±0.005*	7.658±0.003*
Sheep dung	4.310±0.005*	102.771±0.004*	11.238±0.005*	4.408±0.002*	35.739±0.005*	7.546±0.003*

Each value is the Mean ± SD of six replicates

* Significant P<0.05 “t” test between earthworm body before inoculation in vermibeds and after vermicomposting

Table 4: Concentration of different heavy metals (mg/kg) of soil and soil with vermicompost and inoculation of earthworm (*E. fetida*) in crop field

	Heavy metals (mg/kg) of experimental soil					
	Co		Cr		Pb	
Vermicompost	Before sowing	After harvesting	Before sowing	After harvesting	Before sowing	After harvesting
Soil control	4.365±0.005	3.127±0.003*	13.116±0.005	11.003±0.002*	7.790±0.005	8.852±0.003*
Soil with vermicompost and inoculation of earthworm						
Soil + Buffalo Dung	4.369± 0.004	2.292±0.005*	13.144±0.003	8.275±0.003*	7.825±0.002	3.569±0.005*
Soil + Cow Dung	4.352± 0.002	2.254±0.006*	13.123±0.002	8.269±0.005*	7.798±0.005	3.392±0.007*
Soil + Goat Dung	4.358± 0.003	2.266±0.003*	13.120±0.003	8.186±0.003*	7.790±0.003	3.385±0.003*
Soil + Horse Dung	4.380± 0.005	2.335±0.003*	13.126±0.004	8.272±0.006*	7.801±0.004	3.416±0.002*
Soil + Sheep Dung	4.365± 0.003	2.319±0.002*	13.122±0.002	8.268±0.004*	7.791±0.003	3.305±0.006*

Each value is the Mean ± SD of six replicates

Significant P<0.05 “t” test between before sowing and after harvesting in soil of crop

Table 5: Concentration of different heavy metals (mg/kg) of soil and soil with vermicompost and inoculation of earthworm (*E. fetida*) in crop field

	Heavy metals (mg/kg) of experimental soil					
	Ni		Cd		As	
Vermicompost	Before sowing	After harvesting	Before sowing	After harvesting	Before sowing	After harvesting
Soil	3.193±0.003	2.680±0.005*	5.073±0.002	3.103±0.003*	4.171±0.005	3.058±0.003*
Soil with vermicompost and inoculation of earthworm						
Soil + Buffalo Dung	3.195±0.005	0.837±0.002*	5.176±0.003	2.114±0.004*	4.169±0.005	1.599±0.004*
Soil + Cow Dung	3.207±0.004	0.978±0.003*	5.102±0.003	1.953±0.007*	4.172±0.002	1.554±0.002*
Soil + Goat Dung	3.191±0.004	0.801±0.003*	5.073±0.004	1.879±0.003*	4.176±0.006	1.559±0.004*
Soil + Horse Dung	3.201±0.003	0.988±0.006*	5.085±0.002	1.892±0.003*	4.181±0.003	1.568±0.006*
Soil + Sheep Dung	3.196±0.002	0.835±0.003*	5.075±0.003	1.867±0.005*	4.170±0.005	1.562±0.003*

Each value is the Mean±SD of six replicates

Significant P<0.05 “t” test between before sowing and after harvesting in soil of crop

of Co and As were observed significantly decrease after harvesting of maize crop to the combination of soil + cow dung the level of (2.254±0.006 and 2.554±0.002 mg/kg). Cr and Pb were significantly decreased in the combination of soil + goat dung and soil + buffalo dung at the level of (8.186±0.003 and 3.569± 0.005 mg/kg).Whereas, Ni and Cd were decreased in the level of (0.801 ± 0.003 and 1.867 ±0.005 mg/kg) of the combination of soil with vermicomposting after harvesting of maize crop (Table 4, 5). Epastein, [32] and Garcia *et al.* [33] reported that heavy metals accumulated in clay soil of clay fraction of soil because clay sized particle have contain more number of ionic bonding site due to the high amount of heavy metals present on the surface area of the soil. Cd, Pb, Ni and Hg are not essential for plant growth but Cd is more than 5mg/kg responsible for the phytotoxic to plant leaves [34]. Earthworms are one of the most important groups of soil invertebrates and ubiquitous animals living in soils to improve soil fertility by physical, chemical and biological characteristics of the soil [35-38].

Table 6: Concentration of different heavy metals (mg/kg) during maize (*Zea mays*) seed growing before sowing and after harvesting in soil and soil with vermicompost during inoculated earthworm (*E. fetida*)

	Heavy metals (mg/kg) in maize seed					
Vermicompost	Co	Cr	Pb	Ni	Cd	As
Before sowing	0.068±0.003	5.531±0.002	2.497±0.005	3.093±0.003	0.140±0.003	BDL
After harvesting of crop						
Soil + Buffalo dung	0.072±0.002*	3.013±0.003*	1.258±0.003*	1.474±0.003*	0.135±0.005*	BDL
Soil + Cow dung	BDL*	2.751±0.004*	1.192±0.006*	1.924±0.004*	0.152±0.007*	BDL
Soil + Goat dung	0.054±0.005*	2.835±0.003*	1.831±0.003*	1.215±0.005*	0.116±0.002*	BDL
Soil + Horse dung	0.091±0.003*	2.729±0.004*	1.593±0.005*	1.429±0.003*	0.045±0.003*	BDL
Soil + Sheep dung	0.059±0.003*	2.793±0.006*	1.237±0.002*	1.145±0.003*	0.019±0.003*	BDL

BDL= below detectable limit

Each value is the Mean ± SD of six replicates.

Significant P<0.05 "t" test between before sowing and after harvesting of maize crop

Table 7: Concentration of different heavy metals (mg/kg) in earthworm body in soil with vermicompost of different animal dung during cultivation of crop Maize (*Zea mays*)

	Heavy metals (mg/kg) in earthworm body					
Vermicompost	Co	Cr	Pb	Ni	Cd	As
During inoculation						
Earthworm body (Control)	2.391±0.005	101.517±0.003	07.531±0.004	3.825±0.003	38.152±0.003	5.013±0.004
After harvesting of crop						
Soil + Buffalo dung	3.548±0.002*	103.825±0.003*	10.322±0.006*	4.691±0.004*	41.323±0.007*	6.845±0.002*
Soil + Cow dung	3.211±0.004*	104.134±0.005*	09.795±0.003*	5.425±0.003*	40.763±0.005*	6.309±0.003*
Soil + Goat dung	3.275±0.003*	103.729±0.002*	09.362±0.004*	4.519±0.007*	39.642±0.003*	6.912±0.006*
Soil + Horse dung	3.693±0.003*	103.745±0.002*	10.362±0.003*	4.426±0.002*	39.522±0.006*	7.461±0.002*
Soil + Sheep dung	3.238±0.002*	103.683±0.002*	09.501±0.005*	4.282±0.003*	40.335±0.003*	6.784±0.003*

Each value is the Mean ± SD of six replicates

*Significant P<0.05 "t" test between earthworm body before inoculation in soil and after harvesting of soil

The concentrations of different heavy metal were observed in maize seed during sowing and after harvesting of maize crop. The Co and Pb were significantly decreased after harvesting of maize grains containing the (BDL and 1.192 ± 0.006 mg/kg) level of heavy metals concentration in the combination of soil + cow dung. Cr was significantly decreased in the combination of soil + horse and cow dung (2.729±0.004 mg/kg) and concentration of Ni and Cd were decreases in the maize grains the level of (1.145±0.003 and 0.019±0.003 mg/kg) in the combination of soil + sheep dung, whereas As was observed in BDL level of maize grains before and after harvesting of crop (Table 6). Higher concentration of heavy metals in soil are responsible for the gradually decrease seedling growth in rice and corn [39]. Ogri [40] stated that heavy metals present in the oil polluted soil which harmful for the maize cods and leaves potentially toxic and harmful to human by the use as food. Heavy metals accumulate by the foodstuffs in the human bodies can causes much disease such as brain damage, heart attack, cancer, anemia, gout, chronic nephritis, encephalopathy and disease in digestive system [15]. Essential micronutrients are (Co, Cu, Fe, Mn, Mo, Ni and Zn) required for normal growth of plants, essential metabolic activity and other important mechanism in plant cells [41]. The highest concentration of Fe, Pb and Hg metals loss of soil fertility and agricultural crops [42]. However, at high concentrations, these metals exhibit toxic effects on plant cells [43]. Cd and Pb are not essential for plant growth such elements accumulation in crop plants by the root from food and metals contamination in soil [44].

The concentrations of heavy metals were observed in earthworm body in soil and soil + vermicompost of different animal dung during before and after harvesting of maize crop. The Co and As were significantly increased in earthworm (*E. fetida*) body (3.693±0.003 and 7.461±0.002 mg/kg) of the combination of soil + horse dung. The combination of Pb, Ni and Cd were accumulate by earthworm (*E. fetida*) from the combination of soil + buffalo dung the level of (10.322±0.006, 4.691±0.004 and 41.323±0.007 mg/kg), whereas Cr was observed in the combination of soil + horse dung the level of (3.693±0.003 mg/kg) (Table 7). Krauss *et al.* [45] reported that the concentrations of heavy metals are closely related to metals contaminated in food. The different biological wastes converted into rich nutrient through the vermicomposting by use of *E. fetida* [46-47]. The earthworm *E. fetida* is important link in the food chain and they can accumulate the hazardous elements from the soil [19]. Earthworm is a most important invertebrate improve the soil fertility and balance is an ecosystem [36].

CONCLUSION

The earthworm *E. fetida* was a most important invertebrate to remediate the higher concentration of heavy metals by the application of a vermicompost of different animal dung (buffalo, cow, goat, horse and sheep dung) in soil. The *E. fetida* was accumulate the heavy metals concentrations during vermicomposting and soil field of maize crop therefore the good productivity of maize seeds free from heavy metals which safe for environment, human health and eco-friendly.

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