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Waste management and bioremediation using earthworms, *Eisenia fetida* and *Eudrilus eugeniae*

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Abstract : *A study was conducted to find out the efficiency of two earthworm species Eisenia fetida and Eudrilus eugeniae in waste management and bioremediation. Earthworms were cultured in cow dung for three months and then transferred to two different kinds of wastes. Samples of these wastes were tested at 0,45 and 90 days to compare the change in N, P, K content, pH, and percent change in concentration of Cu, Mn, Zn, Fe and Pb. The N, P, K content got enhanced in the experimental set-up with worms, whereas, the concentration of all metals decreased except for Zn. The test for bioaccumulation showed enhanced level of metals in the body tissue of worms and decreased level in the substrate, which indicates that earthworms accumulate metals in their body tissues and bioremediate the wastes. Growth of chilli plants was observed and growth was seen best in the compost prepared by Eisenia fetida.*

Key Words : *Earthworms, waste management, bioremedication.*

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Introduction :

Millions of tons of solid waste comprising a great proportion of household and municipal wastes generated from different sources are creating problems worldwide. The disposal of wastes is of great concern as it poses serious management threats particularly in developing countries like, India where its management is mostly unsystematic and unscientific (Chattopadhyay et al., 2009; Pattnaik and Reddy, 2010). It causes pollution of land, water and air which affects human health and environment.

Earthworms convert organic fraction of wastes into available nutrients (Patnaik and Reddy, 2010). Studies have also shown that the earthworms accumulate heavy metals in their body tissue from contaminated substrates (Edwards and Baxter,1992; Gupta et al, 2005; Suthar et al, 2008).

Bioremediation can be defined as use of naturally occurring species to clean up the contaminated land, water and soil. Due to their biological, chemical and physical action, earthworms can be directly employed within bioremediation strategies to promote biodegradation of organic contaminants.

The present study therefore attempted to assess the remediation of certain heavy metals e.g. Zn, Cu, Mn, Fe and Pb from household and automobile wastes and also bioaccumulation of toxic wastes in the body tissues of two earthworm species i.e. *Eisenia fetida* and *Eudrilus eugeniae* which are commonly used for vermicomposting. A comparison of the nutrient (N,P,K) composition of the two types of wastes was also made before and after composting with the help of the two worm species.

Materials and Methods :

Two types of wastes, household and automobile garage waste were chosen for the present study. Household waste was collected from the kitchen and garden. It included vegetable and fruit peels and dry leaves. Peels of citrus fruits were avoided.

Automobile garage waste was collected from a motor garage at Boring Road, Patna. It mainly consisted of soil heavily stained with petrol, diesel, coolant, mobil, grease and other chemicals required for lubricating, painting and washing the vehicles.

The wastes were mixed with cow dung in 3:1 ratio. A soil sample was also prepared without any kind of waste and was also mixed with cow dung in 3:1 ratio (control). In our study cow dung was used as innoculant to accelerate the vermicomposting process as suggested by Karthikeyan et al (2007), Pramanik et al (2007) and Gupta and Garg (2009). Earthworm species used were *Eisenia fetida* and *Eudrilus eugeniae*.

Earthen pots were used for composting of the wastes. A total of three sets of earthen pots were taken, each set had three pots. The experiment was set up as follows :

- Soil + cow dung (3:1), without worms
- Soil + cow dung (3:1), with *Eisenia fetida*
- Soil + cow dung (3:1), with *Eudrilus eugeniae*
- Soil + Kitchen waste (3:1), without worms
- Soil + Kitchen waste (3:1), with *Eisenia fetida*
- Soil + Kitchen waste (3:1), with *Eudrilus eugeniae*
- Automobile waste + cow dung (3:1), without worms
- Automobile waste + cow dung (3:1), with *Eisenia fetida*
- Automobile waste + cow dung (3:1), with *Eudrilus eugeniae*

The pots were left undisturbed for a period of 90 days. Samples of compost and vermicompost from each replicate were collected at intervals of 0, 45, 90 days for nutrient and heavy metal analysis.

At the end of 90 days, equal number (45) of chilli seeds were sown in each earthen pot and their growth was monitored for a month to document the plant growth in each substrate. Earthworm tissues were also analyzed at 0 and 90 days to test for the bioaccumulation of Cu, Zn, Mn, Fe and Pb in their tissues. For this purpose earthworms' tissues were digested using a standard protocol and the

samples thus obtained were analyzed by using Atomic absorption Spectrophotometer (AAS). Bioaccumulation factor was calculated with the formula, BAF=Concentration of metal in earthworm tissue/Concentration of metal in the substrate.

Results and Discussion :

No significant change in N%, P%, K% and pH were found in the control set-up without worms (Table 1). However N% increased significantly after 90 days in the experimental set with kitchen and automobile wastes and with *Eudrilus eugeniae* (Table 1). K% increased significantly in the experimental set up after 90 days in the automobile waste with *Eudrilus eugeniae* (Table 1). pH reduced significantly from alkaline to neutral in all the experimental set ups except the control set up without worms (Table 1). According to Ndegwa et al (2000), shifting of pH to lower level can be attributed to mineralization of nitrogen and organophosphates and bioconversion of organic materials into intermediate species of organic acids.

There was no significant change in the concentration of metals (Cu, Mn, Fe and Zn) in the control set ups without worms except that the concentration of Cu decreased significantly after 90 days in both control and experimental set ups (Table 2). Further, there was significant increase in the concentration of Zn after 90 days in the set up with kitchen waste and *Eisenia fetida* and in automobile waste with *Eudrilus eugeniae* (Table 2). Mn concentration decreased significantly in the automobile waste with both the species of earthworm (Table 2). Fe concentration was found to decrease significantly in the kitchen and automobile wastes with *Eisenia fetida* as worm species. According to Edwards and Bohlen (1992) the waste ingested by earthworms undergo chemical and microbial changes while passing through the gut and a great proportion of the organic fraction is converted into soluble form which may be the cause of reduction of metal in the substrate.

Table 1. Changes in nutrient composition and pH due to vermicomposting.

	N%			P%			K%			pH		
	0 DAY	45 DAY	90 DAY	0 DAY	45 DAY	90 DAY	0 DAY	45 DAY	90 DAY	0 DAY	45 DAY	90 DAY
S+CD, NO WORM	2.3±.32	2.36±0.31	2.72±0.11	2.42±0.09	2.55±0.06	2.39±0.19	3.18±0.5	3.28±0.53	3.28±0.44	7.68±0.07	6.85±0.17	6.19±0.01
S+CD +EF	1.48±0.02	1.54±0.03	1.61±0.01*	0.95±0.2	1.02±0.2	1.14±0.1	2.6±0.36	2.5±0.21	2.5±0.2	8.37±0.2	8.01±0.13	7.74±0.02*
S+CD +EE	1.42±0.01	1.46±0.07	1.55±0.04	1.26±0.17	1.33±0.15	1.53±0.05	3.17±0.22	3.26±0.21	3.35±0.16	8.2±0.11	7.75±0.13	7.72±0.003*
S+KW, NO WORM	1.49±0.03	1.48±0.005	1.45±0.01	0.69±0.03	0.78±00	0.66±0.02	3.18±0.5	3.28±0.53	3.28±0.44	8.64±0.12	8.36±0.01	8.01±0.2
S+KW+EF	1.49±0.02	1.56±0.03	1.64±0.05	0.91±0.13	1±0.1	1.12±0.08	2.8±0.21	2.9±0.26	3±0.26	7.57±0.18	7.47±0.1	6.96±0.15
S+KW+EE	1.41±0.02	1.49±0.01	1.54±0.03*	1.16±0.06	1.24±0.05	1.29±0.05	3.14±0.29	3.31±0.27	3.49±0.25	8.2±0.11	7.75±0.14	7.72±0.003*
AW+CD, NO WORM	0.61±0.02	0.61±0.02	0.53±0.01	0.17±0.04	0.18±0.06	0.14±0.04	3.18±0.5	3.28±0.53	3.28±0.44	8.64±0.22	8.56±0.14	8.67±0.05
AW+CD+EF	0.59±0.04	0.66±0.05	0.71±0.06	0.23±0.14	0.25±0.15	0.4±0.11	0.73±0.22	0.45±0.21	0.89±0.25	8.2±0.04	7.95±0.02	7.72±0.11*
AW+CD+EE	0.79±0.21	0.85±0.19	0.92±0.23*	0.47±0.1	0.5±0.08	0.62±0.09	0.79±0.07	0.88±0.07	1.12±0.04*	8.13±0.02	7.82±0.04	7.69±0.07*

*Significant at P<0.05 as compared to 0 days

Values are mean ± S.E; S=Soil; CD= cow dung; KW= kitchen waste; AW=automobile waste; EF= Eisenia fetida ; EE= Eudrilus eugeniae

Table 2. Change in the concentration of heavy metals due to vermicomposting

	Zn (ppm)			Cu (ppm)			Mn (ppm)			Fe (ppm)		
	0 DAY	45 DAY	90 DAY	0 DAY	45 DAY	90 DAY	0 DAY	45 DAY	90 DAY	0 DAY	45 DAY	90 DAY
S+CD,												
no worm	1.39±0.08	1.39±0.02	1.51±0.12	0.76±0.003	0.81±0.13	0.79±0.08	6.76±0.46	7.55±0.66	7.64±0.62	14.28±0.38	14.75±0.13	14.41±0.32
S+CD+EF	4.97±0.95	5.49±0.47	6.84±0.09	0.85±0.22	0.88±0.28	0.59±0.01	5.16±0.72	8.56±1.61	5.87±0.22	5.93±1.26	11.19±4.03	4.0±0.19
S+CD+EE	5.52±0.37	6.31±0.31	6.34±0.46	1.45±0.08	1.72±0.11*	0.76±0.1*	7.0±1.02	7.53±0.97	5.7±0.58	4.73±1.07	5.13±0.93	3.44±0.76
S+KW,												
no worm	3.85±0.56	4.25±0.14	4.89±0.09	1.31±0.03	1.54±0.12*	0.55±0.02*	3.73±1.01	3.7±1.13	2.32±0.59	4.53±0.75	5.34±0.15	5.69±0.44
S+KW+EF	0.45±0.16	0.21±0.11	7.2±0.03*	0.95±0.33	1.13±0.28	1.14±0.16	22.78±5.63	22.38±5.65	9.81±0.31	41.49±4.43	39.67±4.52	23.85±2.58*
S+KE+EE	5.52±0.36	6.31±0.31	6.34±0.45	1.45±0.08	1.72±0.11*	0.76±0.1*	7.0±1.02	7.5±0.97	5.7±0.58	4.73±1.07	5.13±0.93	3.44±0.76
AW+CD,												
no worm	7.27±0.65	7.31±0.55	7.27±0.57	3.66±0.55	3.76±0.34	4.04±0.52	18.08±0.03	17.37±0.15	17.8±0.16	53±2.1	43.47±3.35	42.75±1.31
AW+CD+EF	6.2±0.82	5.4±0.03	7.34±0.71	4.44±0.94	15.76±1.67*	4.4±0.42*	17.7±0.81	19.49±0.67*	7.5±0.93*	49.07±6.1	36.03±1.95	24.79±0.84*
AW+CD+EE	5.26±0.33	5.36±0.08*	8.02±0.09*	3.43±0.56	16.64±1.21*	3.58±0.29*	16.69±0.69	20.58±1.19*	9.87±1.05*	38.72±3.52	36.07±2.23	14.05±6.22

*Significant at P <0.05 as compared to 0 days.

Values are mean ± S.E; S=Soil; CD= cow dung; KW= kitchen waste; AW=automobile waste; EF= *Eisenia fetida* ; EE= *Eudrilus eugeniae.*

It was found that the concentration of Pb decreased significantly in the automobile waste and bioaccumulation of Pb and other metals increased significantly in the body tissues of earthworms (Table 3). The concentration of Pb reduced in the substrate with both the species of earthworms. On the other hand, the concentration of Pb increased from 0.005 to 0.67 in the body tissue of *Eisenia fetida* and from 0.005 to 1.16 in the body tissue of *Eudrilus eugeniae* after 90 days. The concentration of Pb in the substrate with *Eisenia fetida* decreased by 45.9% whereas the concentration of Pb in the substrate with *Eudrilus eugeniae* decreased by 43.05% .The concentration of metals Zn, Cu, Fe, Mn also increased significantly in the tissues of earthworms after 90 days in both the species (Table 4). The concentration of Zn, Cu, Mn, Fe in the earthworm tissue increased from 0 to 90 days by 0.89 to 2.97, 0.73 to 3.09, 1.69 to 13.08, 10.91 to 9.51 respectively in *Eisenia fetida*_ and 1.16 to 3.77, 0.82 to 3.08, 3.14 to 15.4, 12.78 to 17.21 respectively in *Eudrilus eugeniae*.The earthworm chloragosomes consist of modified epithelial cells, eleocytes of the gut containing constituents of ion exchange system capable of taking up and accumulating heavy metals (Morgan and Morgan,1988,1990,1991).

Bioaccumulation of Zn and Mn was found to be more in *Eisenia fetida* (Table 4). *Eisenia fetida* accumulated 233% and 673.9% of their initial concentration Zn and Mn respectively, whereas *Eudrilus eugeniae* accumulated 225% and 39% of their initial concentration of Zn and Mn respectively.

Bioaccumulation of Cu and Fe was more in the body tissue of *Eudrilus eugeniae* (Table 4). They accumulated 225% and 345% of their initial concentration of Cu and Fe respectively, whereas in *Eisenia fetida*, the concentration of Cu increased by 332% whereas the concentration of Fe decreased by 12%.

Table 3. Bioaccumulation of Pb in earthworms from automobile waste

	Pb in substrate			Pb in earthworm tissue	
	A+CD+no worm	AW+CD+EF	AW+CD+EE	AW+CD+EF	AW+CD+EE
0 day	0.86±0.05	0.73 ±	0.72±0.01	0.005±0.002	0.005±0.0008
90 day	0.86±0.04	0.39 ±	0.41±0.02	0.67±0.06*	1.16±0.1*

*Significant at P<0.05 as compared to 0 day.

AW= Automobile waste, CD= Cow dung, EF= *Eisenia fetida,* EE=*Eudrilus eugeniae*

Table 4. Bioaccumulation of metals in earthworms from automobile waste

	Zn		Cu		Mn		Fe	
	0 day	90 day	0 day	90 day	O day	90 day	0 day	90 day
Eisenia fetida	0.89±0.03	2.97±0.28*	0.73±0.04	3.09±0.44*	1.69±0.05	13.08±1.03*	10.91±0.43	9.51±4.15
Eudrilus euginae	1.16±0.06	3.77±0.46*	0.82±0.05	3.08±0.16*	3.14±0.05	15.4±0.45*	12.78±0.22	17.21±0.31*

*Significant at P<0.05 as compared to 0 days

The compost prepared by *Eisenia fetida* appeared to be more beneficial for the growth of chilli plants.The health of chilli plants seemed to be better in the automobile wastes bioremediated with worms as compared to other set ups (Fig 1).The Bio Accumulation Factor (BAF) was more than unity for Mn,Pb, and Fe and was less than unity for Cu and Zn (Table 5). BAF for Pb and Fe showed significant variation among the two species (Table 5).The degree of BAF mainly depends upon level of contamination and characterstics of waste, and the earthworm species used (Suthar and Singh, 2009).

Table 5. Bioaccumulation factor of different metals in earthworm tissue

	Eisenia fetida	*Eudrilus eugeniae*
Cu	1.70	0.86
Mn	1.74	1.52
Zn	0.40	0.47
Fe	0.38	1.22
Pb	1.71	2.82

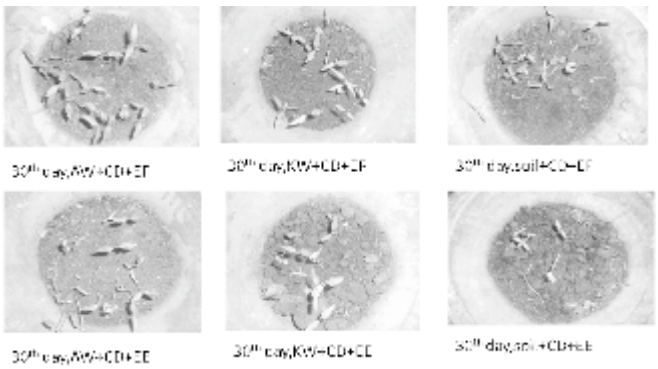

Fig 1. Growth of chilli plants in different substrates on 30th day

AW = Automobile Waste
CD = Cow Dung
KW = Kitchen Waste
EE = *Eudrilus eugeniae*
EF = *Eisenia fetida*

Conclusion :

We deduced from the present study that the earthworms, especially *Eisenia fetida* can be utilized effectively for *ex situ* remediation of Zn, Mn and Pb from urban waste as was observed in the case of automobile waste. *Eudrilus eugeniae* remediated Cu and Fe more effectively. The body tissue analysis showed increased concentration of heavy metals in the earthworm tissues.

The present findings suggest that vermicomposting could be an appropriate technology for remediation of metals from obnoxious wastes. This technique is eco-friendly and improves the nutrient composition of the soil.

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