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Research Article

VERMISTABILIZATION OF FLY ASH AMENDED WITH PRESSMUD BY EMPLOYING *EISENIA FOETIDA*

Rahat Niyazi[1*] and Sadhana Chaurasia[2]

[1]Department of Environmental Science, Institute of Biosciences and Biotechnology, Chhatrapati Shahu Ji Maharaj University, Kanpur, Uttar Pradesh-208 024, India.
[2]Department of Environmental Science, Mahatma Gandhi Chitrakoot Gramodaya University, Satna, Madhya Pradesh-485 331, India.

ABSTRACT
Vermicomposting is commonly used for the management of organic wastes. A study was conducted to evaluate the efficiency of an exotic and epigeic earthworm species (*Eisenia foetida*) for stabilization of fly ash amended with pressmud. The growth and reproduction of *E. fetida* was also monitored in a range of different feed mixtures for 60 days in laboratory under controlled experimental conditions. In all the feed mixtures, a decrease in temperature, pH, TOC, C:N ratio and faecal coliforms, but increase in EC, TKN, TAP and TK was recorded in final vermicompost. The heavy metals content in the vermicomposts was lower than initial feed mixtures. Vermicomposted samples showed 35-50% reduction of heavy metals in 40-60% FA. The growth rate and cocoon production was maximum in 100% PM. Metal analysis of *E. foetida* revealed considerable bioaccumulation of heavy metals in their body. Performance of feed mixture with 40-60% FA was better than the other feed mixtures. The results indicated that fly ash could be converted into good quality manure by vermicomposting if mixed in an appropriate ratio (40-60%) with pressmud.

Keywords: Vermistabilization, Fly ash, Pressmud, *Eisenia foetida*.

INTRODUCTION
Everyday tons of waste is generated by industrial and domestic activities in all over world, most lies on the land or enter rivers, streams through runoff and further contaminating the environment. Effective waste management with sustainable and cost effective solutions is the need of today. Open dumps and poorly designed dumping grounds can pollute surface and ground water causing public health hazards. Meanwhile, the unavailability and rising cost of land near urban areas have made dumps and landfills increasingly expensive and impractical. As communities search for safe, ecofriendly, cost-effective and sustainable ways to manage solid waste, vermicomposting is becoming a more attractive management option, which leads to a reduction of the amount of wastes that require disposal.
Fly ash (FA), a by-product of coal-fired electricity generation plants, is presenting acute waste disposal problems in different parts of the world with the large-scale generation from the consistently increasing numbers of coal-fired plants (Jamil et al., 2009; Wong and Selvam, 2009). The present outlets of fly ash disposal are as a concrete additive and in municipal land filling operation. Some possible agronomic uses of fly ash as, a fertilizer (Rautary et al., 2003; Roy & Joy, 2011), a liming material (Lee et al., 2006) and as a physical amendment (Campbell et al., 1983) have been indicated. Soil application of fly ash waste has been associated with both the favorable (Lin et al., 1983) as well as adverse (Pandey et al., 2009) effects on crop yields. The latter type effect is common at high rates of fly ash due to increased salinity and accumulation of toxic levels of elements (Gupta and Sinha, 2009). To overcome these adverse effect fly ash, a burnt material and therefore contains little organic matter, can be supplemented and vermicomposted by mixing with an additional source of organic matter like pressmud (PM). Pressmud, a by-product of

sugar industry, generates intense heat (65^{o}C), foul odor and takes long time for natural decomposition (Kumar et al., 2010; Raj and Antil, 2011). Vermicomposting is a biological technique of composting wide ranges of organic wastes with the help of the gut micro-organisms of surface-living earthworms (Lazcano et al., 2008; kumar et al., 2012). Vermicompost, the final product of this unique process, is an excellent organic fertilizer since it is homogenous, has desirable aesthetics, has reduced levels of contaminations and tends to hold more nutrients over a longer period, without adversely impacting the environment (Garg and Gupta, 2011; Molina et al., 2013). Thus, the aim of this work was to give an overall view of research idea regarding the stabilization of fly ash amended with pressmud through vermicomposting.

MATERIALS AND METHODS

Collection of materials

Fly ash was procured from the dumping ground of Panki Thermal Power Station near Panki, Kanpur, India. Pressmud was procured from Kisan Sahkari Sugar Mill, Kayamganj, Farrukhabad, India. Earthworm species (*Eisenia foetida*) was procured from Kanpur Gaushala Society, Bhauti, Near Panki Highway, Kanpur, India and cow dung was procured from a cow farm near the university campus as a culturing material for earthworms.

Earthworms culture

The culture of earthworms (*Eisenia foetida*) was maintained under laboratory conditions by using cow dung as a culturing material. The worm's culture was needed for time to time use of earthworms for research work. Generally, *Eisenia foetida* survive on temperature range 16^{o}C – 28^{o}C and are most active on upper ends of its temperature range. In summer season worms enhance their foraging activities and are sexually more active. So the worm's culture was produced in the summer season.

Experimental design

Six feed mixtures having different proportions of fly ash and pressmud were established including one feed of 100% pressmud as control (Table 1). All the fly ash and pressmud quantities were used on dry weight basis that were obtained by drying known quantities of material at 110^{o}C to constant mass in a hot air oven. One kg of feed mixture was put in circular plastic containers (diameter 20 cm and depth 20 cm). All containers were kept in darkness at room temperature (22-26^{o}C). The moisture content of the feed mixture in each container was maintained at 60-80% throughout the study period by sprinkling adequate quantity of distilled water. These feed mixtures were turned manually every day for 21 days in order to eliminate volatile gases potentially toxic to earthworms. After 21 days, fifty nonclitellated hatchlings of *Eisenia foetida* of our own culture were introduced in each container. There were three replicates for each feed mixture and no additional food was added at any stage during the study period. After 60 days granular tea like vermicompost appear on the upper surface of each feed mixture excepting feed mixture no.1. The prepare vermicomposts and inoculated earthworms were used for analysis. Another set of feed mixtures without earthworms was established as control to compare the results.

Physico-chemical and microbiological analysis

The samples were used for chemical analysis on a dry weight basis obtained by oven drying the known quantities of material at 110^{o}C. The temperature was measured at inside the feed mixtures with the help of mercury thermometer (Hoskin G-692), the moisture content was measured using the method of Wu and Ma (2001) and the water holding capacity was measured as described by FCQAO (1994) in terms of moisture content on draining under gravity. The pH and electrical conductivity was determined using a double-distilled water suspension of 1:10 (w/v) that had been agitated mechanically for 30 minutes and filtered through Whatman filter paper No.1 and determination was done by a digital pH meter (ELICO-LI 162) and conductivity meter (ELICO-180) respectively. The total organic carbon was measured by using the Walkley and Black rapid titration method (1934) and the total kjeldhal nitrogen was estimated by microkjeldhal method (Singh and Pradhan, 1981). The total available phosphorous was analyzed by using the colorimetric method of Bray and Krutz (1945) and the total potassium was determined by Flame emission technique using flame photometer (ELICO- CL 361). Total Cr, Cu, Ni, Pb, Zn, Fe, Cd and Mn were determined by using the method of Berman (1980) by means of Atomic Absorption Spectrophotometer (AAS) (Model: 220 FS, Varian, Australia) after digestion of the sample with concentrated Nitric acid (HNO_3) and concentrated Perchloric acid ($HClO_4$) (4:1, v/v). The faecal coliforms were analyzed by the Most Probable Number (MPN) method as prescribed by USEPA (1989) procedure.

Growth study of earthworms

Earthworm growth parameters i.e. individual weight, earthworm weight gain, individual growth rate, cocoon production and juveniles production etc. were analyzed for growth study of *Eisenia foetia*. At the end of the vermicomposting period, the feed in the plastic bins were turned out. Earthworms, cocoons and juveniles were separated from the feed by hand sorting, after which they were counted and weighed after washing them with water and drying them by paper towels. Growth rate of earthworms was determined by using the method of Suthar (2006). For the analysis of heavy metals concentration in earthworms, the earthworms were rinsed free of sample particles and starved on moistened filter paper for 5 days to eliminate the organic and inorganic content of the alimentary canals. They were then oven dried at 65^oC for 4 days and crushed.

All the chemicals used were analytical reagent (AR) grade supplied by Merck Limited, Mumbai. Alkali resistant borosilicate glass apparatus supplied by Borosil Glass Works Limited, Mumbai and double distilled water was used throughout the study for analytical work. All the samples were analyzed in triplicate and results were averaged. Homogenized samples of final vermicompost were stored in airtight plastic vials for further chemical analysis.

RESULTS AND DISCUSSION

Quality assessment of final vermicompost

Eisenia foetida could not tolerate the 100% fly ash. Addition of some other organic waste was essential for the survival of the earthworms in the fly ash. The vermicompost was much darker in color than originally in all the vermireactors and had been processed into homogeneous manure after 60 days of earthworm's activity. Physico-chemical characteristics of the initial feed mixtures (after mixing different composition of fly ash and pressmud) and vermicompost obtained at the offset of the experiment have been encapsulated in Table 2 and Table 3 respectively. The physical parameters like temperature, moisture content and water holding capacity are easily controllable and indicate progress of the vermicomposting process. The temperature below 16^oC and above 28^oC while moisture content below 25% and above 75% is not fit for the survival and growth of *E. foetida* (Devi et al., 2009; Fernandez-Gomez et al., 2010)._Water holding capacity was ranged from 31.35% - 68.38% in the initial feed mixtures. With the exception of extreme heat or cold, nothing will kill worms faster than a lack of adequate moisture. The water holding capacity less than 40% is dangerous to the worms. Water holding capacity in final product was ranged from 43.60% - 78.10%, it increased due to the turning and aeration of the feed material by the worms. There were little changes in the pH of vermicompost as compared to initial values (Table 2 and Table 3). The pH decreased from alkaline (7.30 – 7.80) to acidic (6.62 – 6.56) in all feed mixtures. The pH shift towards acidic conditions has been attributed to mineralization of the nitrogen and phosphorus into nitrites/nitrates and orthophosphate; bioconversion of the organic material into intermediate species of organic acids (Ndegwa et al., 2000). They have also reported that different substrates result in the production of different intermediate species and hence different wastes show a different behavior in pH shift. Haimi and Hutha (1986) postulated that lower pH in the final vermicomposts might have been due to the production of CO_2 and organic acids by microbial activity during the process of bioconversion of different substrates in the feed given to worms. The electrical conductivity (EC) was increased from 27.18% - 33.15% for different feed mixtures after vermicomposting, the variation was significant among all the feed mixtures. This increase in EC might have been due to loss of organic matter and release of different mineral salts in available form such as phosphate, ammonium, potassium etc. (Kaviraj and Sharma, 2003). Gunadi and Edwards (2003) have reported that EC and pH of feed could be the limiting factor for survival and growth of *E. foetida*. Mitchell (1997) reported that *E. foetida* was unable to survive in any material with pH of 9.5 and EC of 5.0 dsm^{-1}. Total organic carbon (TOC) of the final vermicompost was remarkably reduced as compared to the initial feed mixtures. There was a loss of 25.40% - 53.41% TOC in different feed mixtures by the end of vermicomposting period. Data revealed that TOC loss was higher (52.31%) in feed mixture no. 4. Further, TOC reduction was inversely related to the PM content in the feed mixtures, i.e. the reduction was maximum for feed mixture no. 6 (53.41%) and minimum for feed mixture no. 1 (25.40%). This finding was supported by other workers (Kaviraj and Sharma, 2003), who reported 45% loss of carbon during vermicomposting of municipality and industrial wastes. Suthar (2006) reported that earthworms promoted such microclimatic conditions in the vermireactors that increased the loss of TOC from substrates through microbial respiration. Whereas Elvira et al., (1996) have attributed this loss to the presence of earthworms in the feed mixtures. A significant increase in the total kjeldhal nitrogen

(TKN) content occurred following the vermiconversion of FA and PM into vermicompost in different vermireactors. The initial TKN content of the different mixtures was in the range of 0.13% - 1.28% (Table 2). Total nitrogen (TKN) content increased in the range of 0.16% - 1.81% in different feed mixtures (Table 3) after vermicomposting. There was a gain of 23.08% - 47.36% TKN in different feed mixtures by the end of vermicomposting period. Data revealed that TKN addition was higher (46.91%) in feed mixture no. 4. This confirms that if FA is mixed in appropriate quantities (upto 50% on dry weight basis) with PM, would not have antagonistic impact on the final TKN content of the vermicompost. Other workers have also reported similar observation (Bansal and Kapoor, 2000; Atiyeh et al., 2000; Elvira et al., 1996). According to Viel et al., (1987) losses in organic carbon might be responsible for nitrogen addition. However, there are contradictory reports on nitrogen content and its variation in vermicomposting. Ndegwa et al., (2000) and Mitchell (1997) found no significant difference between total nitrogen concentration in the original substrate and the resulting vermicompost. Where as Parvaresh et al., (2004) have reported a great variation in nitrogen concentrations over the whole vermicomposting period. The reason for discrepancies observed in total nitrogen variations in vermicomposting of different wastes lies in the fact that the quality of substrate in feeding the earthworms together with their physical structure and chemical composition affects mineralization of nitrogenous organic compounds and the amount of nitrogen from the compounds (Bohlen et al., 1999). A significance increase in the total available phosphorus (TAP) occurred following the vermiconversion of FA and PM into vermicompost in different vermireactors. The initial TAP of the different feed mixtures was in the range of 0.28% - 1.60% (Table 2). TAP increased in the range of 0.34% - 2.37% (Table 3) in different feed mixtures after vermicomposting. There was a gain of 21.34% - 49.31% TAP in different feed mixtures by the end of vermicomposting period. Data revealed that TAP addition was higher (42.20%) in feed mixture no. 4. According to Lee (1992), if the organic materials pass through the gut of earthworms, then some of phosphorus being converted to such forms that are available to plants. Moreover, he concluded that availability of phosphorus to plants is mediated by *phosphatase* produced within the earthworms and further release of phosphorus may be introduced by microorganisms in their casts, after their excretion. Similarly, Ghosh et al., (1999) have reported that vermicomposting can be an efficient technology for the transformation of unavailable forms of phosphorus to easily available forms for plants. The initial total potassium (TK) content was in the range of 0.06% - 0.23% in different feed mixtures (Table 2). Final TK content was increased in the range of 0.05% - 0.29% (Table 3) in different feed mixtures after vermicomposting. There was a gain of 14.28% - 31.25% TK in different feed mixtures by the end of vermicomposting period. Data revealed that TK addition was higher (31.25%) in feed mixture no. 4. Suthar (2007) suggested that earthworm processed waste material contains higher concentration of exchangeable potassium due to enhanced microbial activity during the vermicomposting process, which consequently enhances the rate of mineralization. The C: N ratio is used as an index for maturity of organic wastes. As evident from the Table 2, 3 and 4 that C: N ratios decreased with time in the entire worm worked feed mixtures. Initial C: N ratio was in the range of 9.7 – 26.0 at zero day. Final C: N ratios of vermicompost were in the range of 5.9 – 9.5, depicting the overall decrease of 39.2% - 67.3% after 60 days of worms' activity. Decline of C: N ratio to less than 20 indicates an advanced degree of organic matter stabilization and reflects a satisfactory degree of maturity of organic wastes (Senesi, 1989). So, in the present study, a high degree of organic matter stabilization was achieved in all the feed mixtures. It was found that there was a rapid decrease in C: N ratio after vermicomposting (Table 3) as compared to the value of C: N ratio in different feed mixtures of without earthworms (Table 4). This demonstrates the role of earthworms in much more rapid decomposition and rate of mineralization of organic matter. The fly ash (FA) was not expected to contain pathogens, however the pressmud (PM) may contain pathogens and therefore microbial analyses was considered essential to assess the safety of the product. The initial samples were found to have a high numbers of faecal coliforms upto 1.7 MPN/g (Table 2). Table 3 showed that faecal coliform levels were low after 21 days precomposting (thermocomposting) and vermicomposting. The samples that were only composted, retained high level of pathogens even after 81 days. Thermocomposting (precomposting) prior to vermicomposting was effective in inactivating the pathogens (Nair et al., 2006).

Heavy metals concentration in final vermicompost and in earthworms

Heavy metals appear in the Fly ash and Pressmud from a variety of sources like coal, soil and dust etc. So, the vermicompost made from fly ash and pressmud may have higher heavy metal concentrations. In small amounts, many of these elements may be essential for plant growth, however, in higher concentrations they are likely to have detrimental effects upon plant growth (Whittle and Dyson, 2002). So, prior to vermicompost application to the soils, there is a need to determine the heavy metal concentrations in the final vermicomposts. In the present study, initial heavy metal content of FA, PM and their different proportions were analyzed which resulted in higher heavy metal concentrations in initial feed mixtures (Table 5). A significance decrease in results showed that heavy metals, viz, Cr, Cu, Ni, Pb, Zn, Fe, Cd and Mn concentrations in final vermicompost in all the feed mixtures were lower than in the initial feed mixtures (Table 6). Our findings are supported by Martin and Bullock (1994) who reported a decrease in heavy metal concentration in vermicompost of oak wood. Similarly, Kumar et al., (2008) have attributed the greater decrease in heavy metals in the castings, as opposed to in the municipal solid waste without earthworms, to the mineralization process that earthworms accelerate during municipal solid waste decomposition and stabilization. While considering the risks associated with heavy metal contaminations in soils, it was found that the concentrations of heavy metals studied in the final vermicompost obtained from the different feed mixtures were lesser than limits set for composts in USA and European countries (Brinton, 2000; Table 9). A significant amount of heavy metals was accumulated by earthworms *Eisenia foetida* in their body (Table 7). The data revealed that maximum accumulation of different heavy metals by earthworms was found in feed mixture no.5 excepting control (feed mixture no. 6) and minimum in feed mixture no.1 (Table 8). Other workers have also reported similar observation (Suthar and Singh, 2009; Das et al., 2012). According to Wang et al., (2013) earthworm species *Eisenia foetida* have ability to bioaccumulate heavy metals in their body tissues.

Biomass growth and cocoon production of *Eisenia fetida*

The biomass production by *E. foetida* in different mixtures has been given in Figure 1. The net weight gain by *E. foetida* was highest (859 mg earthworm^{-1}) in feed mixture no. 6 and lowest (364 mg earthworm^{-1}) in feed mixture no. 1. Increasing percentage of PM in the feed mixtures promoted the increase in biomass gain by *E. foetida.* The growth rate expressed in terms of mg weight gained day^{-1} worm^{-1} has been considered as a good index to compare the growth of earthworms in different feeds (Edwards et al., 1998). The fastest growth rate (9.58 mg worm^{-1} day^{-1}) was observed in feed mixture no. 6 (Figure 2) where as feed mixture no. 1 supported the least growth (1.18 mg worm^{-1} day^{-1}). The total number of cocoons after 60 days in different feed mixtures has been represented in Figure 3. The maximum no. of cocoons was observed in feed mixture no. 6 and minimum were in feed mixture no. 2. It is evident from the figure 3 that the cocoons production was directly related to PM concentration in the studied feed mixtures. Similarly, the total number of juveniles was highest in feed mixture no. 6 and lowest in feed mixture no. 2. The results suggest that addition of FA in PM is not suitable for earthworm production (vermiculture) as the cocoon production is lesser if FA is present in the earthworm feed. Data revealed that number of cocoons and juveniles were not significant in feed mixture no. 1 and 2 (Figure 3). The difference between biomass and cocoon production in different feed mixtures could be related to the biochemical quality of the feed, which was one of the important factors in determining onset of cocoon production (Majlessi et al., 2012). Suthar (2007) summarized that except to the chemical properties of waste, the microbial biomass and decomposition activities during vermicomposting were also important. Finally the results indicated that the addition of 40% fly ash to the pressmud is acceptable during the vermicomposting of FA in terms of fertilizer quality of the vermicompost so obtained. But if prime concern is vermiculture (production of earthworms), then addition of FA in the PM is not suggested.

CONCLUSION

Disposal of fly ash by environmentally acceptable means is a serious problem. Our trials have demonstrated that vermicomposting can be an alternate technology for the management of fly ash mixed with pressmud. In the present study, the vermicomposting of FA amended with PM resulted in the conversion of a waste into value added product i.e. vermicompost. A high degree of FA stabilization was achieved after 60 days of worm activity. The results indicated that after the addition of fly ash in appropriate quantity (40%) to the pressmud, it can be used as a raw material in the vermicomposting. The fertilizer quality of FA based vermicomposts was almost equal to control (prepared by using the pressmud only). But addition of FA in the PM is not suggested if prime concern is vermiculture (production of earthworms) as the cocoon production is lesser if FA is present in the earthworm feed. The study also inferred that the application of FA based vermicompost in the agricultural fields as a soil conditioner or manure, would not have any adverse effect due to heavy metals.

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Table 1: Content of Fly ash and Pressmud in initial feed mixtures

FEED MIXTURE NO.	FLY ASH	PRESSMUD
1	1000[a] (100)[b]	-----
2	800[a] (80)[b]	200[a] (20)[b]
3	600[a] (60)[b]	400[a] (40)[b]
4	400[a] (40)[b]	600[a] (60)[b]
5	200[a] (20)[b]	800[a] (80)[b]
6	-----	1000[a] (100)[b]

a – The figures indicate the weight content in the initial feed mixtures (d/w).
b – The figures in parentheses indicate the percentage content in the initial feed mixtures.

Table 2: Physico-chemical and microbial characteristics of initial feed mixtures

Parameter	Feed Mixture No.					
	1	2	3	4	5	6
Temperature (°C)	18.4 ± 1.6	22.8 ± 0.8	31.2 ± 1.5	39.2 ± 2.8	44.5 ± 2.6	49.7 ± 1.7
Moisture Content (%) (d/w)	4.30 ± 0.38	15.28 ± 1.48	28.23 ± 1.83	40.14 ± 2.32	53.15 ± 2.85	63.90 ± 2.33
Water Holding Capacity (%)	31.35 ± 4.30	35.62 ± 3.19	45.40 ± 2.89	55.56 ± 3.87	62.07 ± 3.17	68.38 ± 2.51
pH	7.30 ± 0.24	7.35 ± 0.23	7.44 ± 0.24	7.59 ± 0.26	7.70 ± 0.20	7.80 ± 0.28
EC (dS/m)	0.92 ± 0.13	1.05 ± 0.03	1.21 ± 0.04	1.46 ± 0.15	1.63 ± 0.06	1.81 ± 0.16
Total Organic Carbon (%)	1.26 ± 0.14	6.34 ± 0.12	13.84 ± 0.29	20.72 ± 0.66	27.44 ± 1.04	33.23 ± 1.06
Total Kjeldhal Nitrogen (%)	0.13 ± 0.04	0.31 ± 0.06	0.54 ± 0.09	0.81 ± 0.07	1.09 ± 0.08	1.28 ± 0.11
Total Available Phosphorous (%)	0.28 ± 0.03	0.51 ± 0.04	0.79 ± 0.08	1.09 ± 0.08	1.40 ± 0.10	1.60 ± 0.15
Total Potassium (%)	0.06 ± 0.01	0.09 ± 0.02	0.12 ± 0.03	0.16 ± 0.05	0.20 ± 0.03	0.23 ± 0.05
C:N ratio	9.69 ± 0.30	20.45 ± 0.19	25.62 ± 0.62	25.58 ± 1.14	25.17 ± 0.87	25.96 ± 0.96
Fecal Coliforms (MPN/g)	< 0.2 ± 0.2	0.2 ± 0.2	0.4 ± 0.2	0.9 ± 0.2	0.9 ± 0.2	1.7 ± 0.2

Values are means of three replicates ±; Standard deviation

Table 3: Physico-chemical and microbial characteristics of final vermicompost

Parameter	Feed Mixture No.					
	1	2	3	4	5	6
Temperature (°C)	15.5 ± 0.54	16.5 ± 0.60	19.2 ± 0.64	22.8 ± 0.83	24.5 ± 0.84	26.9 ± 0.96
Moisture Content (%) (d/w)	39.20 ± 1.67	49.26 ± 1.86	62.91 ± 2.54	62.42 ± 2.57	62.28 ± 1.98	65.62 ± 2.34
Water Holding Capacity (%)	43.60 ± 1.93	48.52 ± 1.66	68.58 ± 2.24	73.84 ± 3.60	73.80 ± 2.62	78.10 ± 2.96
pH	6.62 ± 0.15	6.74 ± 0.29	6.85 ± 0.32	6.80 ± 0.23	6.69 ± 0.29	6.56 ± 0.24
EC (dS/m)	1.17 ± 0.10	1.41 ± 0.05	1.73 ± 0.13	2.08 ± 0.10	2.22 ± 0.08	2.41 ± 0.16
Total Organic Carbon (%)	0.94 ± 0.13	3.56 ± 0.30	7.04 ± 0.20	9.88 ± 0.18	14.08 ± 0.23	15.48 ± 0.29
Total Kjeldhal Nitrogen (%)	0.16 ± 0.03	0.42 ± 0.03	0.77 ± 0.04	1.19 ± 0.09	1.53 ± 0.11	1.81 ± 0.10
Total Available Phosphorous (%)	0.34 ± 0.05	0.63 ± 0.03	1.07 ± 0.09	1.56 ± 0.10	1.93 ± 0.16	2.37 ± 0.14
Total Potassium (%)	0.05 ± 0.01	0.11 ± 0.03	0.14 ± 0.02	0.21 ± 0.05	0.24 ± 0.03	0.29 ± 0.03
C:N ratio	5.87 ± 0.14	8.47 ± 0.10	9.14 ± 0.53	8.30 ± 0.28	9.20 ± 0.26	8.55 ± 0.30
Fecal Coliforms (MPN/g)	< 0.2 ± 0.2	< 0.2 ± 0.2	< 0.2 ± 0.2	< 0.2 ± 0.2	< 0.2 ± 0.2	0.2 ± 0.2

Values are means of three replicates ±; Standard deviation

Table 4: Physico-chemical and microbial characteristics of compost

Parameter	Feed Mixture No.					
	1	2	3	4	5	6
Temperature (°C)	12.8 ± 0.74	12.9 ± 0.54	17.5 ± 0.57	19.9 ± 1.02	25.0 ± 1.07	30.0 ± 1.33
Moisture Content (%) (d/w)	30.98 ± 1.71	40.53 ± 2.20	50.93 ± 2.55	64.25 ± 4.72	60.58 ± 2.53	63.50 ± 2.77
Water Holding Capacity (%)	33.34 ± 2.11	32.16 ± 5.05	58.28 ± 3.88	65.36 ± 5.78	62.24 ± 4.26	62.40 ± 3.65
pH	7.30 ± 0.23	7.25 ± 0.20	7.28 ± 0.21	7.16 ± 0.30	7.10 ± 0.26	7.07 ± 0.20
EC (dS/m)	0.89 ± 0.11	1.20 ± 0.06	1.39 ± 0.16	1.71 ± 0.15	1.73 ± 0.14	2.08 ± 0.19
Total Organic Carbon (%)	1.20 ± 0.12	4.82 ± 0.24	9.27 ± 0.48	13.58 ± 0.63	20.26 ± 0.84	25.51 ± 0.95
Total Kjeldhal Nitrogen (%)	0.10 ± 0.04	0.37 ± 0.08	0.63 ± 0.05	0.93 ± 0.08	1.20 ± 0.14	1.52 ± 0.16
Total Available Phosphorous (%)	0.28 ± 0.06	0.55 ± 0.04	0.85 ± 0.06	1.25 ± 0.04	1.59 ± 0.16	1.90 ± 0.12
Total Potassium (%)	0.04 ± 0.01	0.08 ± 0.02	0.12 ± 0.01	0.16 ± 0.04	0.19 ± 0.04	0.20 ± 0.02
C:N ratio	12.00 ± 0.30	13.02 ± 0.28	14.71 ± 0.33	14.60 ± 0.21	16.88 ± 0.56	16.78 ± 0.54
Fecal Coliforms (MPN/g)	< 0.2 ± 0.2	< 0.2 ± 0.2	0.2 ± 0.2	0.4 ± 0.2	0.4 ± 0.2	0.7 ± 0.2

Values are means of three replicates ±; Standard deviation

Table 5: Concentration of Heavy metals (mg kg^{-1}) in initial feed mixtures

Feed mixture No.	Cr	Cu	Ni	Pb	Zn	Fe	Cd	Mn
1	123.48 ± 10.65	78.60 ± 5.80	88.21 ± 3.08	54.93 ± 4.10	83.74 ± 8.23	431.50 ± 14.65	43.89 ± 1.63	321.40 ± 13.16
2	101.20 ± 5.25	86.33 ± 3.26	71.62 ± 2.91	50.50 ± 2.05	106.92 ± 5.49	582.63 ± 20.37	33.20 ± 2.18	267.65 ± 21.48
3	82.90 ± 3.93	90.55 ± 3.51	58.64 ± 2.72	47.12 ± 2.02	127.94 ± 5.33	664.94 ± 21.91	23.68 ± 1.94	218.23 ± 13.02
4	61.10 ± 3.39	95.73 ± 3.70	43.47 ± 1.59	42.10 ± 2.35	146.96 ± 12.13	739.34 ± 31.78	16.10 ± 0.60	166.52 ± 10.63
5	40.50 ± 3.04	101.30 ± 4.14	30.49 ± 1.44	39.31 ± 1.63	168.98 ± 15.37	885.72 ± 33.63	6.65 ± 0.31	116.88 ± 6.26
6	28.43 ± 2.34	103.79 ± 3.91	19.64 ± 1.06	38.85 ± 3.93	185.96 ± 13.92	935.00 ± 41.79	BDL	73.72 ± 5.08

Values are means of three replicates ±; Standard deviation, BDL = Below Detection Limit

Table 6: Concentration of Heavy metals (mg kg^{-1}) in final vermicompost

Feed mixture No.	Cr	Cu	Ni	Pb	Zn	Fe	Cd	Mn
1	109.62 ± 4.03	72.50 ± 3.06	81.83 ± 5.75	51.90 ± 3.73	73.65 ± 4.51	378.88 ± 13.71	39.90 ± 2.71	288.24 ± 12.31
2	83.92 ± 2.61	78.79 ± 3.12	61.40 ± 3.06	40.57 ± 2.72	84.48 ± 2.80	470.29 ± 17.66	27.55 ± 1.23	228.14 ± 15.81
3	63.40 ± 2.55	74.27 ± 4.29	42.81 ± 2.29	33.45 ± 1.62	92.41 ± 2.98	487.36 ± 21.04	18.22 ± 0.51	168.29 ± 10.14
4	42.45 ± 2.48	65.89 ± 2.13	29.35 ± 3.27	26.48 ± 0.98	92.21 ± 3.74	493.71 ± 24.67	10.61 ± 0.48	113.86 ± 11.78
5	24.45 ± 1.01	66.09 ± 3.03	18.75 ± 1.09	21.38 ± 0.98	95.26 ± 5.49	529.12 ± 29.77	3.22 ± 0.22	69.48 ± 5.41
6	15.38 ± 1.69	56.18 ± 2.68	12.69 ± 0.87	20.25 ± 0.77	94.49 ± 4.58	460.33 ± 19.90	BDL	37.63 ± 5.64

Values are means of three replicates ±; Standard deviation, BDL = Below Detection Limit

Table 7: Concentration of heavy metals accumulated by earthworms (mg/kg)

Feed mixture No.	Cr	Cu	Ni	Pb	Zn	Fe	Cd	Mn
1	15.29 ± 0.58	6.84 ± 0.23	6.80 ± 0.25	4.01 ± 0.26	10.40 ± 0.34	52.89 ± 1.87	4.45 ± 0.20	31.90 ± 1.02
2	20.61 ± 0.77	10.22 ± 0.30	10.42 ± 0.52	10.21 ± 0.32	25.47 ± 1.20	286.39 ± 11.72	5.53 ± 0.18	49.61 ± 1.71
3	31.72 ± 1.43	33.56 ± 1.15	22.05 ± 1.12	21.11 ± 0.94	57.37 ± 1.82	308.28 ± 10.47	10.36 ± 0.43	97.50 ± 4.07
4	25.25 ± 1.30	40.69 ± 1.32	18.42 ± 0.97	23.72 ± 0.82	76.30 ± 4.10	339.73 ± 11.52	8.52 ± 0.34	85.86 ± 4.34
5	19.44 ± 0.69	40.03 ± 2.13	11.60 ± 0.44	19.43 ± 0.62	73.17 ± 2.40	442.49 ± 10.70	4.18 ± 0.18	62.78 ± 3.03
6	14.53 ± 0.68	52.43 ± 1.71	7.51 ± 0.43	19.10 ± 0.66	78.92 ± 3.35	471.56 ± 22.60	0.72 ± 0.10	31.47 ± 1.14

Values are means of three replicates ±; Standard deviation

Table 8: Percentage (%) reduction of heavy metals in different feed mixtures after vermicomposting

Feed mixture No.	Cr	Cu	Ni	Pb	Zn	Fe	Cd	Mn
1	11.73 ± 2.23	7.30 ± 2.34	7.71 ± 3.50	7.30 ± 3.59	12.41 ± 3.91	12.26 ± 3.12	10.14 ± 3.73	9.93 ± 2.09
2	17.30 ± 3.19	11.84 ± 3.79	14.54 ± 4.09	20.21 ± 4.10	20.82 ± 5.17	19.15 ± 3.50	16.65 ± 3.60	14.54 ± 3.04
3	23.09 ± 3.52	18.06 ± 3.90	27.60 ± 4.68	28.80 ± 3.29	27.84 ± 4.15	26.36 ± 3.30	23.75 ± 4.43	22.67 ± 3.97
4	35.15 ± 4.44	31.50 ± 3.87	32.37 ± 3.66	36.34 ± 4.59	36.92 ± 4.32	33.14 ± 4.30	32.92 ± 3.75	31.56 ± 6.40
5	40.49 ± 3.60	34.52 ± 3.09	38.05 ± 4.08	44.42 ± 3.17	43.30 ± 5.14	39.95 ± 3.83	42.86 ± 4.66	40.71 ± 4.38
6	43.79 ± 4.36	45.51 ± 3.65	38.23 ± 3.57	48.16 ± 4.92	49.43 ± 4.53	50.43 ± 4.46	0.72 ± 0.16	49.68 ± 5.95

Values are means of three replicates ±; Standard deviation

Table 9: Heavy metal limits (mg kg^{-1}) for compost in USA and European countries

Heavy metal	EU limit range	USA biosolids limit
Chromium	70 – 200	1200
Copper	70 – 600	1500
Cadmium	0.7 – 10	39
Mercury	0.7 – 10	17
Nickel	20 – 200	420
Lead	70 – 1000	300
Zinc	210 - 4000	2800

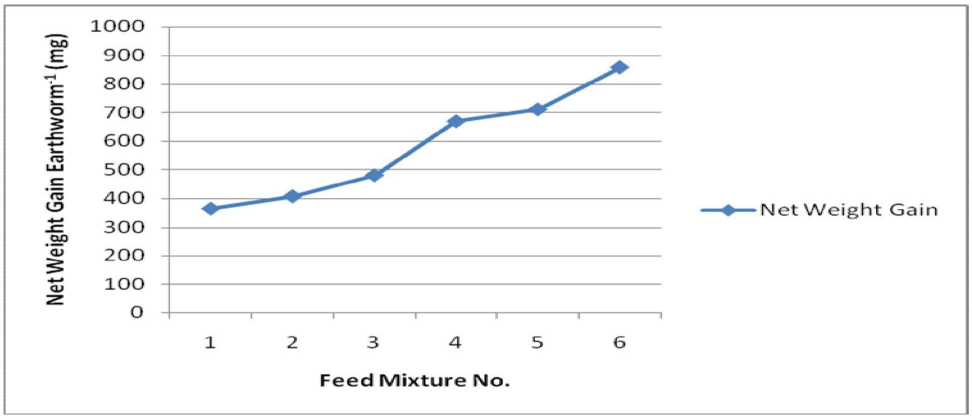

Fig. 1: Biomass Growth of *E. foetida* in different feed mixtures during vermicomposting.

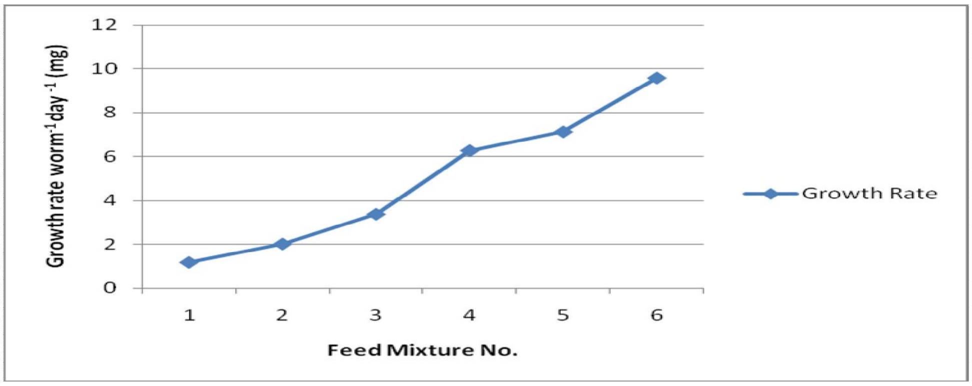

Fig. 2: Growth rate of *E. foetida* in different feed mixtures during vermicomposting

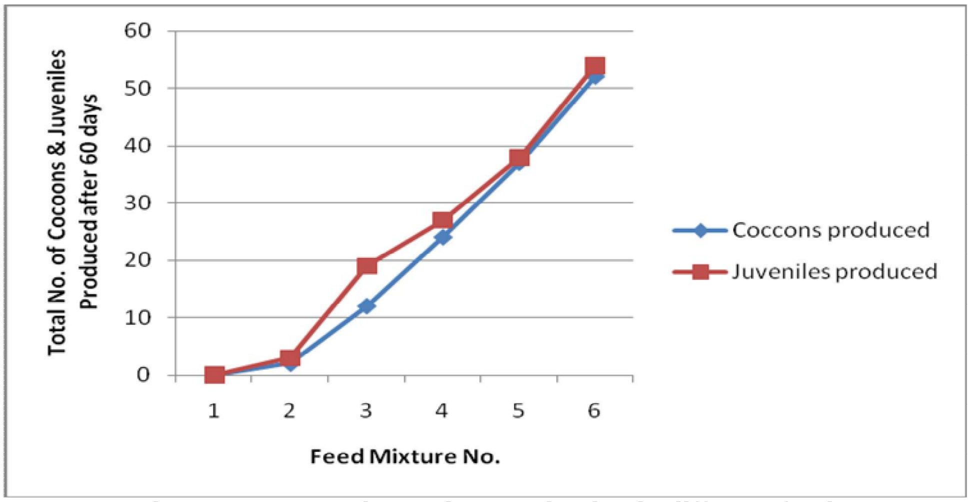

Fig. 3: Cocoons and Juveniles production in different feed mixtures during vermicomposting

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