Eco. Env. & Cons. 26 (February Suppl. Issue) : 2020; pp. (S119-S123)
Copyright@ EM International
ISSN 0971–765X

Effect of the Earthworm E*udrilus eugeniae* on microbial and enzymatic activity during Vermicomposting of organic matter collected from Musiri area, Tamil Nadu, India

V. Senthil and R. Sivakami*

PG & Research Department of Zoology, Arignar Anna Govt. Arts College, Musiri 621 211, Tamil Nadu, India

(Received 3 October, 2019; accepted 2 November, 2019)

ABSTRACT

Vermiculture has been receiving considerable attention globally due to its potential role in organic farming and sustainable development. As characterizing microbial communities and enzyme activities during vermicomposting can provide vital information regarding the evolution of the process, the present study was undertaken in E*udrilus eugeniae* using various vermicomposts. Among the various vermicompost, the mixture containing cow dung, press mud and poultry waste (1:1:1 ratio) recorded maximal microbial population. Among the individual vermicomposts, poultry waste recorded the highest microbial count. Among the various enzymes, dehydrogenase activity was the highest in all vermicomposts. Nevertheless, other enzymatic activities (cellulase, protease and phosphatase) also showed increased activity in vermicomposts after vermicomposting. The present study suggests that a combination of vermicomposting mixtures are more efficient than individual vermicomposts.

Key words : *Eudrilus eugeniae, Vermicomposting, Enzymatic activity*

Introduction

Soil is the soul of the infinite life that promotes diverse microflora (Esakkiammal *et al*., 2015). Soil biota plays an important role in supporting nutrient cycling as well as creating and stablising soil structure (Thangaraj, 2015). Earthworms are extremely important in soil formation principally through their activities in consuming organic matter, fragmenting and mixing it infinitely with mineral particles to form aggregates (Muthukumaravel *et al*., 2008). During their feeding, earthworms promote microbial activity greatly which in turn accelerates the breakdown of organic matter and stabilization of soil aggregates.

Vermiculture has been receiving considerable attention in the recent years globally for its potential role in organic farming and sustainable development (Kale and Bano, 1986) as disposal of solid waste has become a major problem due to shortage of dumping sites and strict environmental laws. Various studies have shown that vermicomposting of organic wastes accelerates organic matter stabilization (Neuhauser *et al*., 1988; Frederickson *et al*., 1997) and gives a product rich in chelating and phytohormonal elements (Tomati and Galli, 1995) which has a high content of microbial agents and stabilized humic substances (Ferruzi, 1986).

Knowledge about the food requirement and digestive capability of the earthworms is essential to understand the decomposition process during vermicomposting. The enzymes and the enzyme activity in the gut of earthworms and in the casts are extremely important since they are responsible for the degradation of organic materials and to make the nutrients in available form for the plants (Devi and Prakash, 2015). As characterizing microbial communities and enzyme activities during vermicomposting process can provide valuable information regarding the evolution of the process, the rate of biodegradation and the maturity of the product, the present study was undertaken in the earthworm E*udrilus eugeniae* using various vermicomposts.

Materials and Methods

Organic waste and earthworm species

Poultry waste (PW) droppings was collected from Indian feeds farm, Namakkal district, Tamil Nadu, India. Press mud (PM) was obtained from effluent treatment plant of E.I.D. parry Sugar Mill located at Pettavaithalai, Tamil Nadu, India. Fresh cow dung (CD) was collected from the agricultural farm, Musiri, Tamil Nadu, India. The earthworm species *Eudrilus eugeniae* of different age groups were cultured and developed outside the laboratory on partially degraded cow dung as feed, respectively. Earthworm *E. eugeniae* (25-30 days) were randomly picked from the culture and used for the purpose of this experiment.

Experimental design

Six vermicomposters (cement tank) were established having 4 kg of feed mixture each containing Cow dung, Press mud and Poultry waste alone (control) and cow dung (CD), Fresh mud (FM) mixed with Poultry waste (PW) in different rations (Table 1). Each vermicomposter was established in triplicate. The feed mixtures were turned manually every day for 14 days Fiftyspecies of worms were introduced in each vermicomposter separately. The moisture content was maintained at 65-75% during the experiment. The vermicomposter were covered with moist jute to prevent moisture loss. The 0 day (Initial) refers to the day of inoculation of earthworms after stabilization of 14 days. Samples (initial substrate and vermicompost) for periodical analysis were taken before inoculating earthworms and at the end of experimentation.

Determination of total microbial populations and activity

The different microbial colonies developing on the plates were estimated by counting. Microbial biomass was analyzed by the chloroform fumigation – extraction method (Vance *et al.* (1987). The number of colony forming unit (CFU) on the surface on the media was counted and expressed as CFUx10^6g^{-1}, according to the method described by Baron *et al*. (1994) to determine the microbial activity (in terms of all the vermicomposters and worm gut.) Dehydrogenase activity was according to the method described by Stevenson, (1959). Cellulse, protease, urease and phosphatase activities were calculated according to the method described by Garcia *et al.* (1994).

Results and Discussion

The various vermicomposters used for the present study are presented in Table 1, while Table 2 records the total microbial population during the initial stages in various vermicomposters along with the gut microbial count in the earthworm. As evident from the table, the total microbial population was found to differ in all the vermicomposters with the highest being recorded in the mixture containing cow dung + press mud + poultry waste (1:1:1 ratio), followed by a mixture containing press mud + poultry waste (1:1 ratio) and cow dung + press mud (1:1 ratio). When a comparison between individual combinations are taken, poultry waste recorded the highest microbial count followed by fresh mud and cow dung respectively. A perusal of literature reveals that Meharaj and Mannivannan (2015) while studying the vermicomposting ability of *E. eugeniae* also reported maximum microbial content in a mixture containing cow dung, press mud and pig manure. Pramanik *et al*. (2009) suggested that microbes are the key factor in nutrient transformation and addition of bulking material in initial inorganic waste resulted in enrichment in the nutrient status of vermicomposts. Fracchia *et al.* (2006) proposed that there was a symbiotic relationship between the earthworms and their gut flora which enhanced the nutrient content of vermicomposts. In the present study, the increased microbial levels and enhanced activity in the final product could be due to the higher nutrient concentration in the initial substrate material and vermicompost, multiplication of microbes while passing through gut of earthworm, optimal moisture and large surface area of casts ideally suited for better feeding, multiplication and activity of microbes as suggested by Meharaj and Manivannan (2015). It appears that among the various vermicomposts, the mixture ofcowdung, fresh mud and poultry waste provided the above conditions better resulting in increased microbial population.

The various enzymatic activities during vermicomposting of various combinations are recorded in Tables 3 and 4. Wong and Fang (2002) and Sen and Chandra (2009) reported that dehydrogenase activity is considered as a parameter for microbial activity which is related to a group of enzymes that catalyze metabolic reactions producing ATP through the oxidation of organic matter and has been often used to monitor the biological activity of composting and vermicomposting process. Results of the present study clearly shows that the combination containing cow dung + press mud + poultry waste recorded the maximum dehydrogenase activity followed by the mixture containing press mud + poultry waste and cow dung + press mud. Among the individual combinations, the vermicompost containing poultry waste recorded maximum dehydrogenase activity followed by fresh mud and cow dung.

Meharaj and Manivannan (2015) while studying the vermicomposting ability of *E.eugeniae* also re-

Table 1. Description of Vermicompost used for experimentation of the present study.

S. No.	Vermicompost	Ratio	Description
1.	Control (CD)	-	100% Cow Dung
2.	Fresh Mud (FM)	-	100% Fresh Mud
3.	Poultry Waste (PW)	-	100% Poultry Waste
4.	Control+ Fresh Mud	1:1	1 Part Cow Dung + 1 Part Fresh Mud
5.	Fresh Mud + Poultry Waste	1:1	1 Part Fresh Mud + 1 Part Poultry Waste
6.	Control + Fresh Mud + Poultry Waste	1:1:1	1 Part Cow Dung + 1 Part Fresh Mud + 1 Part Poultry Waste

Table 2. Total Microbial Population Count (CFU/106^{-6}) in initial Substrate, Gut of worms and Vermicompost of Different Vermicomposter of Poultry Waste amends with different organic waste

S. No.	Vermicomposter	Initial Substrate	*Eudrilus eugeniae*	
			Gut Worms	Vermicompost
1.	Cow dung (CD)	4.2 ± 0.26	5.8 ± 0.64	5.6 ± 0.67
2.	Fresh mud (FM)	5.6 ± 0.52	5.2 ± 0.28	5.8 ± 0.64
3.	Poultry waste (PW)	2.8 ± 0.64	6.2 ± 0.28	6.2 ± 0.58
4.	Control + Fresh Mud (1:1 ratio)	4.6 ± 0.26	6.8 ± 0.34	6.4 ± 0.5
5.	Fresh Mud + Poultry Waste (1:1 ratio)	4.3 ± 0.28	6.7 ± 0.26	7.0 ± 0.64
6.	Control + Fresh Mud + Poultry Waste (1:1:1 ratio)	4.2 ± 0.42	8.6 ± 0.64	8.4 ± 0.24

Table 3. Cellulase and Protease during Vermicompost of Poultry-waste with different Organic-waste using *Eudrilus eugeniae*

S. No.	Vermicomposter	Cellulase (24 hr)		Protease (24 hr)	
		Initial substrate	Vermicompost	Initial substrate	Vermicompost
1.	Cow dung (CD)	5.2 ± 0.72	6.4 ± 0.32	4.8 ± 0.62	5.3 ± 0.48
2.	Fresh mud (FM)	5.6 ± 0.64	6.8 ± 0.28	5.2 ± 0.28	6.3 ± 0.56
3.	Poultry waste (PW)	5.8 ± 0.72	7.0 ± 0.28	4.8 ± 0.32	6.4 ± 0.64
4.	Control + Fresh Mud (1:1 ratio)	4.8 ± 0.28	7.2 ± 0.34	3.8 ± 0.36	7.2 ± 0.72
5.	Fresh Mud + Poultry Waste (1:1 ratio)	5.2 ± 0.34	7.4 ± 0.64	4.9 ± 0.62	8.0 ± 0.64
6.	Control + Fresh Mud + Poultry Waste (1:1:1 ratio)	5.8 ± 0.64	7.8 ± 0.52	5.2 ± 0.24	8.6 ± 0.72

Table 4. Phosphatase and Dehydrogenase activity during Vermicompost of Poultry-waste amended with different organic-waste using *Eudrilus eugeniae*

S. No.	Vermicomposter	Phosphatase (mL/5 g substrate)		Protease (mL/5 g substrate)	
		Initial substrate	Vermicompost	Initial substrate	Vermicompost
1.	Cow dung (CD)	3.2 ± 0.36	4.9± 0.64	8.6± 0.26	14.4 ± 0.82
2.	Fresh mud (FM)	2.6 ± 0.46	3.6 ± 0.34	8.9± 0.72	16.7± 0.42
3.	Poultry waste (PW)	3.2± 0.38	3.8± 0.28	8.2± 0.64	17.2± 0.24
4.	Control + Fresh Mud (1:1 ratio)	3.6± 0.64	4.4± 0.82	8.8± 0.62	17.4± 0.42
5.	Fresh Mud + Poultry Waste (1:1 ratio)	3.4± 0.28	4.8 ± 0.26	8.9± 0.28	18.6± 0.64
6.	Control + Fresh Mud + Poultry Waste (1:1:1 ratio)	3.6± 0.39	5.2± 0.26	9.2± 0.64	19.6± 0.26

ported highest dehydrogenase activity in both the worms. This is in line with the present observation. However, Devi and Prakash (2017) while studying the vermicomposting ability of *E.eugeniae* recorded invertase as the enzyme which showed maximal activity which was followed by dehydrogenase. In the present study invertase activity has not been assessed.

A comparison of other enzymatic activities (cellulase, protease and phosphatase) also suggests an increase in their activities in all the vermicomposts after vermicomposting. Similar results were also obtained by Devi and Prakash (2015) and Meharaj and Manivannan, (2015) while working on other earthworms. The reasons for the increased enzymatic activities in the casts could be due to greater consumption rate, enhanced gut microbial population (Parthasarathi, 1997; Parthasarathi and Renganathan, 2002; Parthasarathi *et al*., 2007), enhanced microbial population in the casts, availability of adequate oxygen, moisture, temperature, pH, the quantity and quality of organic matter and the amount of elemental nutrients essential for microbial growth and activity during vermicomposting (Oleink and Byzov, 2008). Hence it can be suggested that the specific environment in vermicomposting, organic matter composition and earthworm gut condition as well as selective effects of the earthworm gut fluid and surface excreta are the major dynamic forces for the observed pattern of microbial community and enzyme activity in the vermicomposts as already reported by Neuhauser *et al*. (1980); Byzov *et al*. (2007) and Meharaj and Manivannan (2015). Results of the present study further suggest that combination of mixtures is more efficient than individual vermicomposts.

References

Baron, J.E., Peterson, R.L. and Finegold, M. S. 1994. *Diagnostic Microbiology*, 9th edition, Chap. 9, Mosby, London, pp. 79-96.

Byzov, B. A., Khomyakov, N. V., Kharin, S. A. and Kurakov, A. V. 2007. Fate of soil bacteria and fungi in the gut ofearthworms. *Eur. J. Soil Biology*. 43: 149-156.

Devi, J. and Prakash, M. 2015. Nutrient Status, Micoflora and Enzyme activities in Vermicompost of three different organic substrates. *Int. J Curr. Res. Aca. Rev*. 3 : 443-448.

Esakkiammal, B., Esaivani, C., Vasanthi, K., Lakshmi Bai, L. and Shanthi Preya, N. 2015. Microbial diversity of Vermicopost and Vermiwash prepared from *Eudrilluseuginae. Int. J. Curr. Microbiol. Application. Sci*. 4 : 873-883.

Ferruzi, C. 1986. *Manual de Lombricultura*. Ed. Munidiprensa, Madrid. pp. 45 - 55

Fracchia, L., Dohrmann, A. B., Martinotti, M. G. and Tebbe, C. C. 2006. Bacterial diversity in a finished compost and vermicompost: differences revealed by cultivation independent analyses of PCR-amplified 16S rRNA genes. *Applications. Microbiol. Biotechnology*. 71: 942-952.

Frederickson, J., Butt, K. R., Morris, R. M. and Daniel, C. 1997. Combining vermiculture with traditional green waste composting systems. *Soil Biol. Biochem*. 29 : 725-730.

García, C. Hernández, T. Costa, F. and Ceccanti, B. 1994. Waste Manage. *Research, Soil Biol*. 12 : 457–466.

Kale, R. D. and Bano, K. 1986. *Org. Waste Uti. by Vermicomposting*. GK VK Agricultural University, Bangalore, India.

Meharaj, I. and Manivannan, S. 2015. Influence of poultry waste amended with different organic food sources on growth and reproduction performance of indigenous earthworms *Lampito mauritii* (Kinberg) and *Perionyx excavatus* (Perrier). *European Journal of Experimental Biology*. 5 : 1-6.

Muthukumaravel, K., Amsath, A. and Sukumaran, M. 2008. Vermicomposting of vegetable wastes using cow dung. *Chem*. 5(4) : 810-813.

Neuhauser, E. F., Hartenstein, R. and Kaplan, D. L. 1980. Growth of the Earthworm Foetida in relation to population density and food rationing. 35 : 93-98.

Neuhauser, E. F., Loehr, R. C. and Malecki, M. R.1988. The potential of earthworms for managing sewage sludge. In: Edwards, C. A. and Neuhauser, E. F.(ed.), *Earthworms in Waste and Environmental Management.* SPB Academic Publishing, The Hague. pp. 9-20.

Oleink, A. S. and Byzov, B. A. 2008. Response of bacteria to earthworm surface excreta. *Microbiology*. 77 : 765-773.

Parthasarathi, K. and Renganathan, L. S.2002. Supplementation of press mud vermicasts with NPK enhances growth and yield in leguminous crops (*Vigno mungo* and *Arachis hypogaea). Journal of Current Science.* 2 : 35-41.

Parthasarathi, K., Renganathan, L. S., Anandi, A. and Zeyer, J. 2007. Diversity of microflora in the gut and casts of tropical composting earthworms reared on different substrates. *J. Environ. Biol.* 28 : 87-97.

Pramanik, P., Ghosh, G. K., Ghosal, P. K. and Banik, P. 2009. Effect of microbial inoculation during vermicomposting of different organic substrates on microbial status and quantification and documentation of acid phosphatase. *Bioresour. Technology*. 98: 2485-2494.

Sen, B. and Chandra, T. S. 2009. Do earthworms affect dynamics of functional response and genetic structure of microbial community in a lab-scale composting system? *Bioresour. Technology*. 100 : 804-811.

Stevenson, I.L., *Canadian J. Microbiology.* 1959, 5: 229-235.

Thangaraj, R. 2015. Leaf litter waste management by vermicomposting using local and exotic Earthworm species. *Journal of Science.* 5 : 314-319.

Tomati, U. and Galli, E. 1995. Earthworms, soil fertility and plant productivity. *Proceedings of the International Colloquium on Soil Zoology. Acta Zoological Fennica.* 196: 11-14.

Vance, E.D., Brookes, P.C. and Jenkinson, D. S. 1987. *Soil Biol. Biochemistry.* 19 : 703–707.

Wong, J. W. C. and Fang, M. 2002. *J. Applications. Microbiology*. 92 : 764-775.