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Nutrient Status, Microflora and Enzyme Activities in Vermicomposts of Three Different Organic Substrates

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KEYWORDS

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A B S T R A C T

Twenty one days pre-decomposed substrates, teak leaf litter (TLL), paper mill sludge (PMS) and pressmud (PM) in combination with cowdung was subjected to vermicomposting using the epigeic earthworm, *Perionyx ceylanensis* for 60 days. The enzymes, amylase (EC 3.2.1.1), cellulase (EC 3.2.1.4.), protease (EC 3.4.0), phosphatase (EC 3.1.3.1, 2) and invertase (EC 3.2.1.20) were analysed in vermicomposts of TLL, PMS and PM on 60th day. The physico-chemical characteristics and microbial availability in vermicompost clearly showed that the manurial value of vermicompost is of good quality; the enzymatic analyses in earthworm gut, vermicasts and vermicomposts showed that the vermicomposts produced were biologically active and can be used as manure for crop plants and to improve soil fertility status.

Introduction

Vermicompost is an aerobically degraded and humified organic matter which has undergone chemical degradation by enzymes in the gut of earthworms and by the associated microbial population. In several studies higher enzyme activities were measured in worm casts than in surface soil (Tiwari *et al.*, 1989). Ranganathan and Vinotha (1998) reported that posterior gut of reproductively active, clitellate stage of the compost worm, *Eudrilus eugeniae* when reared in press mud exhibits enhanced amylase, protease, acid and alkaline phosphatase and cellulase activity compared to the activity of these enzymes observed in immature, pre-clitellate stage worms reared in the same media. Acid phosphatase activity in earthworm is due to microorganisms and alkaline phosphatase is regarded as typical of earthworms (Lee, 1985). The expression of alkaline phosphatase in developing embryo and mature stages of the earthworm, *Eisenia*

andrei was investigated by Park *et al.*, (1996). The embryonic stages examined in this study appeared to have only one slow-moving form of alkaline phosphatase which had a different mobility from the intestinal alkaline phosphatases of the mature worm, suggesting that intestinal alkaline phosphatases of embryos may be different from mature forms.

It has been reported that cellulase, amylase, invertase, protease and phosphatase activities in pressmud and vermicasts of fresh, 15- and 30-day-old casts of *Lampito mauritii* and *Eudrilus eugeniae* decreased considerably with reference to ageing (Parthasarathi and Ranganathan, 2000). Aira et al. (2005) found that increased in extracellular enzyme activity with reference to the rate of pig slurry application. In both rates of pig slurry applied, the presence of earthworms in young layers stimulated microbial growth which decreased once earthworms left the slurry and the layers aged. This increase was related to the initial activation of the microbial enzymes studied as correlations between microbial biomass and enzymes showed, which indicated an increase of intracellular enzyme activity. In the present study, physico-chemical characteristics, microbial population and enzyme activities in vermicomposts of three different organic substrates were analysed.

Materials and Methods

Teak leaf litter (TLL) was collected from an agroform near KSK College, Kanchipuram. The paper mill sludge (PMS) was procured from a private mill near Kanchipuram. The filter mud or pressmud (PM) was collected from Cheyyar Co-operative Sugar Mills Ltd., located in Thenthandalam, Anakkayur, Thiruvannamalai District, Tamil Nadu. The cowdung was collected from nearby cattle sheds in fresh form and allowed to stabilize for one week and used for the study. The organic substrates, TLL, PMS and PM were subjected to initial decomposition in rectangular draining cement tanks of 75cm×60cm×45cm size by sprinkling water, regular mixing and turning of the substrates for 20 days. The earthworm, *Perionyx ceylanensis* Mich. for the study, originally collected from culture bank of the Department of Biology, Gandhigram Rural Institute- Deemed University, Tamil Nadu, India was mass multiplied in cow dung and used for vermicomposting studies. Based on the studies reported by Karmegam and Daniel (2009a and 2009b) and Prakash and Karmegam (2010) on vermicomposting of different organic substrates using *P. ceylanensis*, the ratio of organic substrate mix, i.e., 1:1 (50:50) proportion on dry weight basis was used in the present study.

Accordingly, the pre-decomposed organic substrates were mixed with cowdung in 1:1 ratio on dry weight basis, transferred to vermibeds and moistened to hold 60-70% moisture content. The vermicomposting studies were carried out for 60 days using *P. ceylanensis* in three replicates twice under controlled conditions. The vermibeds without earthworms were also maintained as control. The microbial population count, i.e., bacteria, fungi and actinomycetes and the characteristics of vermicomposts were analysed as described in Prakash and Karmegam (2010).

The enzymes, amylase (EC 3.2.1.1), cellulase (EC 3.2.1.4.), protease (EC 3.4.0), phosphatase (EC 3.1.3.1, 2) and invertase (EC 3.2.1.20) in the composts and vermicomposts harvested from three different substrates were analysed. The activity of amylase, cellulase and invertase was done according to method described by Galstyan (1965). Protease and phosphatase activities were analysed adopting the

methods of Sarath et al. (1989) and Jannossy (1963), respectively.

Results and Discussion

The characteristics of vermicompost obtained from three different substrates in combination with cowdung prepared using *P. ceylanensis* are given in Table 1. The nutrient contents in the vermicomposts of TLL+cowdung, PMS+cowdung and PM+cowdung is found to be in elevated levels. The organic carbon content and C/N ratio in PM+cowdung was 27.00% and 19.29 which was the lower than the vermicompost obtained from TLL and PMS vermibed substrates. The total nitrogen, phosphorus and potassium contents were in the range of 1.16-1.40%, 0.98-1.30% and 0.87-1.05% respectively. Similar reports on the various parameters of the vermicompost obtained at ICRISAT, Hyderabad were made by Nagavallemma et al. (2004) and Prakash and Karmegam (2010).

The results of the present study showed that the vermicomposts of three different organic material combinations with *P. ceylanensis* is rich in nutrients, suitable to be used as organically rich source of biofertilizers for any crop. Different ratios of organic materials such as leaf litter, pressmud and paper mill sludge with cowdung for vermicomposting have been successfully used by Kaviraj and Sharma (2003) and Prakash and Karmegam (2010). The elevated level of Zn, Mn and Fe in vermicompost indicates accelerated mineralization with selective feeding by earthworms on materials containing these metals. Increased levels of macro- and micro-nutrients in vermicomposts were also observed by Suthar (2007). The population of bacteria, fungi and actinomycetes in the vermicomposts of TLL, PMS and PM are given in Fig. 1 which shows enormous population build up of microbes in vermicompost. The existence of symbiotic relationship between earthworms and microorganisms and also the presence of increased number of microorganisms in the vermicompost has been reported by certain workers (Parthasarathi 2007; Jayakumar *et al.*, 2009).

Table 2 shows the activities of enzymes, namely amlylase, cellulose, invertase, phosphatase and protease in vermicomposts of three different vermibed substrates. All the enzymes analysed in the study showed higher levels in vermicompost than in composts (control sets without earthworms).

Table.1 Physico-chemical Characteristics of Vermicomposts of TLL, PMS and PM Substrates

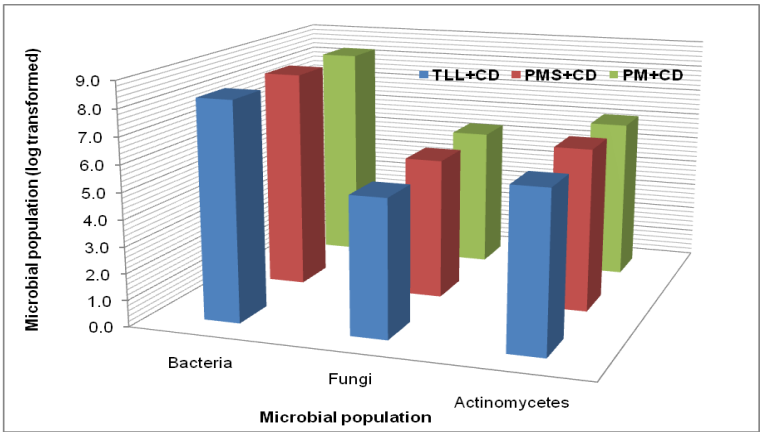
Parameter	TLL+Cowdung	PMS+Cowdung	PM+Cowdung
pH (at 25ºC)	7.21	7.30	7.10
E.C. (mS/cm)	3.20	4.30	5.10
Ca (%)	1.20	1.50	2.00
Na (%)	0.22	0.18	0.54
K (%)	0.95	0.87	1.05
Mg (%)	0.34	0.45	0.50
Org. carbon (%)	26.50	28.00	27.00
N (%)	1.16	1.30	1.40
P (%)	1.30	0.98	1.10
C/N Ratio	22.84	21.54	19.29

Table.2 Enzyme Activities in the Vermicomposts of Different Substrates

Enzymes analysed	Vermibed substrates					
	TLL + CD (1 : 1)		PMS + CD (1 : 1)		PM + CD (1 : 1)	
	Compost	Vermicompost	Compost	Vermicompost	Compost	Vermicompost
Amylase	2.40	5.31	2.18	3.70	3.10	4.33
Cellulase	4.33	8.04	3.89	6.36	4.25	7.67
Invertase	3.82	6.36	2.53	5.05	4.83	7.01
Phosphatase	2.11	7.13	4.18	6.33	3.65	5.45
Protease	2.90	5.67	2.45	4.61	2.78	6.16

(Activities of amylase, cellulase and invertase are expressed as mg of glucose/g of oven dry samples for 24 hrs of incubation; Protease activity: mg of glutamic acid/g of oven dry substrates for 24 hrs of incubation; Phosphatase activity: mg/phenol/g of oven dry substrates for 24 hrs of incubation).

Fig.1 Total Microbial Population in the Vermicompost of Three Different Substrates



The activity of amylase in TLL+CD vermicompost was 5.31 mg of glucose/g of oven dry samples for 24 hrs of incubation; whereas, the compost of the same substrate was lower (2.40 mg of glucose/g of oven dry samples for 24 hrs of incubation. A maximum of 8.04 mg of glucose/g of oven dry samples for 24 hrs of incubation was recorded for cellulase in TLL+CD than the others. The maximum activity for invertase, phosphatase and protease was 7.01 mg of glucose/g of oven dry samples for 24 hrs of incubation in PM+CD vermicompost, 7.13 mg/phenol/g of oven dry substrates for 24 hrs of incubation in TLL+CD vermicompost and 6.16 mg of glutamic acid/g of oven dry substrates for 24 hrs of incubation in PM+CD vermicompost respectively (Table 2). Lee (1985) reported increased enzyme activities and microbial populations in worm casts as compared with the underlying soil. Digestive enzymes of *Perionyx millardi* have been assayed by Mishra (1993) and her result shows the presence of protease, cellulase, amylase, invertase and urease. The earthworm showed maximum activity for protease and amylase, minimum activity for invertase, cellulase and urease in comparison to other tropical earthworms. Further, the activities of microorganisms in earthworm gut and vermireactors are related to enzyme activities in vermicompost (Kumar et al. 2011b). The nutrient contents, NPK, microbiological status and enzymatic

activities observed in the vermicomposts produced from three different substrates, teak leaf litter, paper mill sludge and press mud in combination with cowdung (1:1) for 60 days using *P. ceylanensis* show that the vermicomposts can be used as manure for plant growth.

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