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RESEARCH ARTICLE

ENZYME ACTIVITIES AND MICROBIAL DYNAMICS OF VERMICOMPOST OF PAPERMILL SLUDGE BY THE EARTHWORM SPECIES - *EUDRILUS EUGENIAE*

[1,*]Ponmani, S. and [2]Udayasoorian, C.

[1]Dryland Agricultural Research Station, Tamil Nadu Agricultural University, Chettinad-630 102, India
[2]Department of Environmental Sciences, Tamil Nadu Agricultural University, Coimbatore-641 003, India

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ABSTRACT

Major enzyme activities of vermicompost of papermill sludge by earthworm species *Eudrilus eugeniae* and its simple compost were assessed across different periods in relation to their respective initiative substrates. In this experiment, the vermibed was prepared from different raw material viz., Mixed Liquor Suspended Solids (MLSS) and Leaf Litter (LL) were mixed with Cow Dung (CD) mixed with different ratios. All vermibeds expressed a significant increase in enzyme activities. Overall, the maximum value of enzyme activities was received in CD + MLSS + LL in 1:1:2 ratios. Further, these differences in overall enzyme activity agree with the variation found in extracellular enzyme activity suggesting certain dependence on substrate availability.

INTRODUCTION

In recent days, the cry of pollution is heard worldwide and international consensus can also be seen to protect the natural resources from various types of pollution. Now, the existence of mankind seems to be in threat because the problems of pollution have not only increased proportionally but also become more complicated and diversified due to the disposal of untreated and partially treated domestic and industrial effluents. Pulp and paper manufacturing is one of the oldest and 15th largest industry in India with an installed capacity of about three million metric tonnes of finished product per annum. It is a very water intensive industry and ranks third in the world, after the primary metals and the chemical industries, in terms of freshwater withdrawal (Tewari *et al.,* 2009). The Tamil Nadu Newsprint and Papers Limited (TNPL) situated at Kagithapuram, Karur District, Tamil Nadu, is the major paper industry in India established in 1984 with production capacity of 50,000 t of Newsprint and 40,000 t of writing paper every year. Later, it was expanded to meet the production capacity of 2,32,000 t $year^{-1}$ in 2007. Currently, it is discharge huge amounts of solid wastes [ETP sludge (10 t d^{-1}), fly ash (5 t d^{-1}), lime sludge (30 t d^{-1}), wet pith (3 t d^{-1}) and filter cake (5 t d^{-1})], which poses serious threat to the surrounding environment. Therefore, attempts were made to convert these solid wastes into value added biomanures for enhancing the soil fertility and crop productivity.

Corresponding author: Ponmani
Dryland Agricultural Research Station, Tamil Nadu Agricultural University, Chettinad-630 102, India.

Besides, enormous quantities of Mixed Liquor Suspended Solids (MLSS) (180 to 210 t d^{-1}) are being generated during wastewater treatment. MLSS is a mixture of raw or settled wastewater and activated sludge contained in an aeration basin in the activated sludge process which appears as slurry. Owing to presence of microbial inoculum, half the quantity of generated MLSS is recycled as seeding material in activated sludge process and rest of the quantity is dewatered in nearby sand filter beds. The dewatered MLSS is then disposed to near by farmers field after 2 to 3 months of natural curing for land application with out any value addition. Hence, bioconversion of MLSS into manure is of great interest since it contains carbohydrates, wood fibres such as hemi-cellulose, cellulose, lignins, as well as micro- and macro nutrients, trace metals, clays, water and with C: N ratios ranging from 10 to 300 (Price and Voroney, 2008). Conversion of MLSS into value added organic manures not only solves the disposal problems and pollution, but also replenishes the soil and reduces the fertilizer cost (Esse *et al.,* 2001).

Therefore, MLSS along with suitable bulking agent has to be standardized using composting technology. Composting is the best well known processes for stabilization of any organic solid wastes (Arvinder *et al.,* 2010). Enzyme activities have been used widely as an index of soil fertility or ecosystem status because they are involved in the biological transformations of native and foreign compounds in soils. Enzyme activities have been indirectly used as an index of microbial population on

organic matter decomposition. Thus, it was necessary to determine the enzymatic activity and microbial population during organic matter decomposition.

MATERIALS AND METHODS

Preparation of vermibeds

In this experiment, samples of raw materials *viz.,* Mixed Liquor Suspended Solid (MLSS), Leaf Litter (LL) and Cow Dung (CD) were collected from Tamil Nadu Newsprint and Papers Limited (TNPL). The LL consists of mainly mango and eucalyptus leaf. The LL were dried, chopped and sieved (<2 mm). The LL was amended with CD and MLSS in different ratios in order to produce eight feed mixtures. The substrate and mixed substrate material served as bedding as well as feed for the earthworms. One treatment was composed of pure CD (T_1); another treatment was composed of MLSS (T_2). The following mixed substrates (bedding) were prepared: (i) CD + MLSS in 1:3 ratios (T_3); (ii) CD + MLSS in 1:1 ratios (T_4); (iii) CD + MLSS in 3:1 ratios (T_5); (iv) CD + MLSS + LL in 0:1:3 ratios (T_6); (v) CD + MLSS + LL in 1:1:2 ratios (T_7); and (vi) CD + MLSS + LL in 1:2:1 ratios (T_8). The experiment was laid out in a Randomized Block Design and the experimental beddings were prepared in triplicate for each treatment. All beddings were kept for 2 months prior to experimentation for thermal stabilization, initiation of microbial degradation and softening of waste. About, 750 g of twenty two week old clitellated earthworms were collected from the stock culture and released into different beds containing 150 kg of substrate material. The moisture level of the substrates was maintained around 70 to 75 % throughout the study period by periodic sprinkling of adequate quantity of tap water.

Sampling and analysis

The samples were collected at different stages of the vermicomposting experiment *viz.,* 30, 60 and 90 days after initiation. The fresh samples were collected for enzyme analysis.

Enzyme analysis

Urease activity

Ten gram of dry and sieved compost sample was taken in a 100 ml volumetric flask. To this 1.5 ml of toluene was added, mixed well and incubated for 15 min. Then 10 ml of 10 per cent urea solution and 20 ml of citrate buffer were added, mixed thoroughly, stoppered and incubated for 3 h at 37^0 C. Then the volume was made up to 100 ml with distilled water, mixed by shaking immediately. The contents were filtered through Whatman No.1 filter paper and 1ml of filtrate was pipetted out into 50 ml volumetric flask. To this 9 ml of distilled water, 4 ml of phenate and 3 ml of NaOCl were added, mixed well and allowed to stand for 20 min. The volume was made up to 50 ml and mixed well. The bluish green colour developed was read at 630 nm. Simultaneously a blank was also prepared (without urea solution). The concentration of urease in the sample was obtained from the standard graph using diammonium sulphate (Bremner and Mulvaney, 1978).

Phosphatase activity

Five gram (w) of the fresh compost sample was taken in a boiling tube with 10 ml distilled water. To this 0.25 ml toluene and 1ml *p*-nitrophenol phosphate (PNPP) were added and incubated at room temperature for an hour. Then 5 ml of 0.5 M sodium chloride and 20 ml of 0.5 M sodium hydroxide was added to the sample and filtered through Whatman No.42 filter paper. The colour intensity was read at 420 nm and the concentration of phosphatase (X) in the sample was obtained from the standard graph (Tabatabai and Bremner, 1969).

Phosphatase activity of the sample (µg of PNPP/g) = X/w.

Dehydrogenase activity

Fresh compost sample (5 g) was taken in a boiling tube along with 1ml of 3 per cent 2,3,5-Triphenyl tetrazolium chloride and 1 ml of 1 per cent glucose. To this 2.5 ml distilled water was added and incubated for 24 h. Then 10 ml methanol was added to the setup and incubated for another 6 h. The sample was then filtered using Whatman No.1 filter paper. The filtrate obtained was red in colour. The colour developed was read at 485 nm and the concentration of dehydrogenase (X) in the sample was obtained from the standard graph drawn by using Tri Phenyl Formazan (TPF) as standard (Chendrayan *et al.,* 1980).

Dehydrogenase activity of the sample (µg of TPF/g) = X/5

Microbial population

The bacterial, fungal and actinomycetes population were determined using the standard dilution spread-plate method described by Allen (1953), Martin (1950) and Rangaswami (1966).

Statistical Analysis

Two way analysis of variance (ANOVA) was applied to determined any significant ($P<0.05$) difference among the parameters observed in vermicompost bed (Gomez and Gomez 1984). The treatment differences that are not significant were noted as Non Significant (NS).

RESULTS AND DISCUSSION

Temperature

Significant differences in temperature were observed throughout the period of composting. In vermicomposting process, the temperature increased in all the treatments up to 60^{th} day of composting. Then it decreased and reached the ambient temperature at 90^{th} day of composting. Invariably, at all the stages of vermicomposting process, the highest value (46.4, 59.5 and 32.0 ^{0}C) was recorded in T_5 [CD+ MLSS (3:1)] followed by T_7 [CD+ leaf litter + MLSS (1:1:2)] and the lowest value of 40.4, 50.8 and 30.3 ^{0}C was recorded in T_2 (MLSS alone). The interaction effect was found to be significant. This was due to the heat released by the oxidative action of intensive microbial activity on the organic matter resulted in

Table. Changes in temperature (^{0}C) during fortification of MLSS through vermicomposting process

Treatments	Temperature (^{0}C)				
	Sampling period (days)				
	Initial	30	60	90	Mean
T_1	29.9	42.1	55.6	31.3	39.5
T_2	29.3	40.4	50.8	30.3	38.0
T_3	30.8	45.3	52.9	31.3	40.1
T_4	31.3	45.4	53.5	30.5	40.2
T_5	32.4	46.4	59.5	32.0	42.6
T_6	30.1	45.3	52.4	30.4	39.6
T_7	31.4	46.3	59.3	31.6	42.2
T_8	31.3	46.1	56.5	31.5	41.4
Mean	30.8	44.7	55.1	31.1	40.4
	SEd			(P<0.05)	
T	0.85			1.71	
D	0.60			1.21	
T x D	1.71			3.41	

Treatments
T_1- CD T_2- MLSS SEd- Standard
T_3- CD + MLSS (1:3) T_4- CD + MLSS (1:1) Error deviation
T_5- CD + MLSS (3:1) T_6- CD+ LL + MLSS (0:1:3) T- Treatments
T_7- CD+ LL + MLSS (1:1:2) T_8- CD+ LL + MLSS (1:2:1) D-Days

Table. Changes in urease activity during fortification of MLSS through vermicomposting process

Treatments	Urease activity (µg N g^{-1})				
	Sampling period (days)				
	Initial	30	60	90	Mean
T_1	72	161	206	131	143
T_2	85	182	248	154	167
T_3	80	170	224	140	154
T_4	84	176	240	148	162
T_5	90	212	265	184	188
T_6	78	154	252	160	161
T_7	94	221	278	196	197
T_8	86	198	260	178	181
Mean	84	184	247	161	169
	SEd			(P<0.05)	
T	5.13			10.3	
D	3.62			7.24	
T x D	10.3			20.5	

Treatments
T_1- CD T_2- MLSS SEd- Standard
T_3- CD + MLSS (1:3) T_4- CD + MLSS (1:1) Error deviation
T_5- CD + MLSS (3:1) T_6- CD+ LL + MLSS (0:1:3) T- Treatments
T_7- CD+ LL + MLSS (1:1:2) T_8- CD+ LL + MLSS (1:2:1) D-Days

the rise in temperature during the first mesophilic phase of composting process (Peigne and Girardin, 2004). The temperature of the following thermophilic phase reaching about 60 ^{0}C when most of the organic matter was degraded with the help of thermophilic bacteria and fungi, consequently depleting most of the oxygen. The thermophilic phase was followed by cooling phase, when compost maturation stage occurred and compost temperature dropped to that of the ambient (Zibilske, 1999). Then, the decreasing trend of temperature with the progress of composting process occurred, which was probably due to the decreased bacterial activity. It may also be attributable to regular sprinkling of water. According to Tiquia *et al.* (1997), temperature is a good indicator for maturity of the compost.

Enzyme activity

The enzyme activity in the vermicompost initially increased and then decreased gradually as shown in table. The results corroborates with the findings of Tiquia (2002), who reported that during composting, the soluble organic matter in the starting material is initially assimilated by the microorganism and once the soluble organic matter is used up, the microorganisms produce hydrolytic enzymes to depolymerize the larger compounds (*ie* lignin, cellulose and hemicellulose) to smaller fragments that are water soluble. The water soluble components dissolve in water and are finally assimilated by the microorganisms.

Urease activity

The urease activity was observed to increase on 30th day, later it showed a decreasing trend for all the treatments. During 30th day of vermicomposting process, significantly higher urease activity of 221 µg N g^{-1} of compost was recorded in T_7 [CD+ leaf litter+ MLSS (1:1:2)], which was on par with T_5 [CD+MLSS (3:1)]. At 60 and 90th day of vermicomposting process, significantly higher urease activity (278 and 196 µg N g^{-1} of compost) was received in T_7 followed by T_5. Invariably T_1 (CD alone) recorded the least urease activity (161, 206 and 131 µg N g^{-1} of compost) at all the three stages of vermicomposting process. Among the interaction effect, the treatment and days were significantly different from each other.

The increase in urease activity in vermicompost might be attributed to increase in extra cellular enzyme activity during controlled biodegradation of wastes. This was due to the continuous accumulation of cell-released (extra cellular) enzymes in humic matter, which became stabilized and resistant to physical and microbial degradation (Banitez *et al.,* 2000). The enzyme urease was responsible for breakdown of urea into CO_2 and NH_3. During the composting process, urease activity was found to be increased and reached maximum during the thermophilic period but declined to a constant low level at the end of the composting process. Reithel (1971) reported that the increase in urease activity depends on the nature of the starting material, which is rich in excreted urea. Thus it is favourable for the development of an ureolytic flora which later reduces, or stops producing the enzyme when urea becomes exhausted. Maximum urease activity was recorded in the treatment T_7 [CD+ leaf litter+ MLSS (1:1:2)]. This might be due to higher microbial population and temperature. Skujins (1967) reported that the temperature, organic matter and population of microorganisms also affect the urease activity. Frankkenberger and Dick (1982) reported that urease activity was significantly correlated with organic carbon and total nitrogen.

Phosphatase activity

During the process of vermicomposting, the phosphatase activities were maximum on 60th day and remain roughly static till the vermicomposting process was completed. Among the treatments, the urease activity showed a significant difference with maximum activity (29.3 and 36.8 µg of PNPP g^{-1} of compost) in the treatment T_7 [CD+ leaf litter+ MLSS (1:1:2)],

Table. Changes in phosphatase activity during fortification of MLSS through vermicomposting process

Treatments	Phosphatase activity (µg of PNPP g^{-1})				
	Sampling period (days)				
	Initial	30	60	90	Mean
T_1	14.1	16.1	19.2	17.7	16.8
T_2	19.8	22.2	28.5	21.3	23.0
T_3	19.2	21.4	27.1	20.5	22.1
T_4	19.5	22.7	27.3	20.9	22.6
T_5	21.0	29.0	36.5	22.4	27.2
T_6	18.8	20.5	22.2	20.1	20.4
T_7	23.7	29.3	36.8	25.6	28.9
T_8	19.8	25.1	29.2	21.2	23.8
Mean	19.5	23.3	28.4	21.2	23.1
	SEd			(P<0.05)	
T	0.50			0.99	
D	0.35			0.70	
T x D	0.99			1.97	

Treatments

T_1- CD	T_2- MLSS	SEd- Standard Error deviation
T_3- CD + MLSS (1:3)	T_4- CD + MLSS (1:1)	T- Treatments
T_5- CD + MLSS (3:1)	T_6- CD+ LL + MLSS (0:1:3)	D-Days
T_7- CD+ LL + MLSS (1:1:2)	T_8- CD+ LL + MLSS (1:2:1)	

Table. Changes in dehydrogenase activity during fortification of MLSS through vermicomposting

Treatments	Dehydrogenase activity (µg of TPF g^{-1} 24 h)				
	Sampling period (days)				
	Initial	30	60	90	Mean
T_1	19.0	22.8	24.1	19.2	21.3
T_2	20.1	25.9	27.5	21.7	23.8
T_3	18.2	24.2	26.8	20.3	22.4
T_4	19.6	25.3	27.0	21.0	23.2
T_5	21.3	26.8	28.4	22.6	24.8
T_6	18.9	23.1	25.8	20.3	22.0
T_7	21.7	27.8	29.5	23.1	25.5
T_8	21.0	26.2	28.0	22.2	24.4
Mean	20.0	25.3	27.1	21.3	23.4
	SEd			(P<0.05)	
T	0.48			0.97	
D	0.34			0.68	
T x D	0.97			1.94	

Treatments

T_1- CD	T_2- MLSS	SEd- Standard Error deviation
T_3- CD + MLSS (1:3)	T_4- CD + MLSS (1:1)	T- Treatments
T_5- CD + MLSS (3:1)	T_6- CD+ LL + MLSS (0:1:3)	D-Days
T_7- CD+ LL + MLSS (1:1:2)	T_8- CD+ LL + MLSS (1:2:1)	

Table. Changes in bacterial population during fortification of MLSS through vermicomposting

Treatments	Bacterial population (× 10^6 CFU g^{-1})				
	Sampling period (days)				
	Initial	30	60	90	Mean
T_1	36.6	42.3	45.3	17.3	35.4
T_2	38.0	55.3	58.0	33.6	46.2
T_3	37.6	49.0	51.6	29.3	41.9
T_4	38.0	51.6	54.3	30.6	43.6
T_5	40.3	61.0	64.0	38.0	50.6
T_6	37.0	45.6	47.0	19.0	37.2
T_7	41.3	63.6	67.6	38.6	52.8
T_8	38.3	56.6	60.3	35.3	47.6
Mean	38.4	53.1	56.0	30.1	44.4
	SEd			(P<0.05)	
T	0.56			1.12	
D	0.40			0.80	
T x D	1.12			2.25	

Treatments

T_1- CD	T_2- MLSS	SEd- Standard Error deviation
T_3- CD + MLSS (1:3)	T_4- CD + MLSS (1:1)	T- Treatments
T_5- CD + MLSS (3:1)	T_6- CD+ LL + MLSS (0:1:3)	D-Days
T_7- CD+ LL + MLSS (1:1:2)	T_8- CD+ LL + MLSS (1:2:1)	

Table. Changes in actinomycetes population during fortification of MLSS through vermicomposting

Treatments	Actinomycetes population (× 10^2 CFU g^{-1})				
	Sampling period (days)				
	Initial	30	60	90	Mean
T_1	20.6	19.0	41.0	20.0	25.2
T_2	27.3	32.0	42.6	29.0	32.7
T_3	25.6	26.3	41.3	22.6	29.0
T_4	28.6	31.3	42.0	25.3	31.8
T_5	34.3	39.3	48.6	35.0	39.3
T_6	23.3	23.6	41.3	21.3	27.4
T_7	37.0	42.3	52.6	37.0	42.2
T_8	30.3	38.6	50.0	32.3	37.8
Mean	28.4	31.6	44.9	27.8	33.2
	SEd			(P<0.05)	
T	0.56			1.12	
D	0.40			0.80	
T x D	1.12			2.25	

Treatments

T_1- CD
T_2- MLSS
T_3- CD + MLSS (1:3)
T_4- CD + MLSS (1:1)
T_5- CD + MLSS (3:1)
T_6- CD+ LL + MLSS (0:1:3)
T_7- CD+ LL + MLSS (1:1:2)
T_8- CD+ LL + MLSS (1:2:1)

SEd- Standard Error deviation
T- Treatments
D-Days

which was on par with T_5 [CD+MLSS (3:1)] in both 30 and 60th day of vermicomposting process. At 90th day of vermicomposting process, significantly higher phosphatase activity (25.6 µg of PNPP g^{-1} of compost) was recorded in T_7 and it was closely followed by T_5. The treatment T_1 (CD alone) registered the least phosphatase activity (16.1, 19.2 and 17.7 µg of PNPP g^{-1} of compost) at all the stages of composting. The interaction was significantly different from each other. Phosphatase plays an important role in transforming the organic phosphorus into the available form of phosphorus (Pallab De *et al*., 1990). In all the compost treatments, the level of phosphatase enzyme increased during the early stage and remained constant during the later periods of the process. The increase in phosphatase activity might be attributed to high temperature and bacterial population (Chonkar and Tarafdar, 1984). The treatments that received cow dung, leaf litter and MLSS with the ratio of 1:1:2 (T_7) recorded maximum phosphatase activity. The greater phosphatase activity in vermicompost from cow dung might be due to greater nitrogen content of the substrate, which in turn resulted to higher microbial activity.

Dehydrogenase activity

The dehydrogenase activity showed significant difference among the treatments with the highest value in (T_7) CD+ leaf litter + MLSS (1:1:2) (27.8 and 29.5 µg of TPF g^{-1} of compost for 24 h)) at 30 and 60th day of vermicomposting process. At the end of the vermicomposting process, similar to 30th and 60th day the T_7 also received the higher dehydrogenase activity of 23.1 µg of TPF g^{-1} of compost for 24 h, which was on par with T_5 and T_8. Invariably T_1 (CD alone) registered least dehydrogenase activity (22.8, 24.1 and 19.2 µg of TPF g^{-1} of compost for 24 h) at all the three stages of vermicomposting process. The interaction was significantly different from each other. Biological oxidation of organic compounds is generally a dehydrogenation process, therefore dehydrogenases is a very important enzyme group in evaluating composting process (Fang *et al.,* 1998). Dehydrogenases activity of all treatments showed a similar trend with a marked increase during initial stages of composting and thereafter gradually decreased. This might be due to increase in microbial population in the initial stage which decreased at the later stages of composting. The result was in accordance with the findings of Singh and Ganguly (2005), who reported that higher value of dehydrogenase activity in conventional compost indicated that the compost was still being decomposed and hence it exhibited higher microbial activity. In fully matured compost decomposition is slowed down and the dehydrogenase activity is thus lower. The dehydrogenase activity was maximum in the cow dung +leaf litter + MLSS (1:1:2). Dhruva Kumar *et al*. (1992) also reported that dehydrogenase activity showed a significant positive correlation with number of microorganisms and amounts of organic carbon.

Microbial dynamics

Microbial succession plays a key role in composting process and appearance of some microorganisms reflects the quality of maturing compost (Ishii *et al.,* 2000; Ryckeboer *et al.,* 2003). The microbial population increased from the beginning of the composting period (30th day) and then decreased during the later stage (90th day). This might be due to abundant degradable organic matter present during the initial stages. Afterwards, the exhaustion of these readily decomposable substances caused by the intense microbial activity and by stabilization of organic matter into humic substances. Bhardwaj (1999) and Mini *et al.* (1999) reported that microorganisms developing in the composting system at primary stage are greater in number than those appearing in later stages.

During the period of vermicomposting, the bacterial population increased up to 60th day of vermicomposting and decreased subsequently during the vermicomposting process. At 30 and 60th day of vermicomposting process, bacterial population was higher (63.6 and 67.6 x 10^6 CFU g^{-1} of compost) in T_7 [CD+ leaf litter+ MLSS (1:1:2)], followed by T_5 [CD+MLSS (3:1)]. At 90th day of vermicomposting process, significantly higher bacterial population of 38.6 x 10^6 CFU g^{-1} of compost was noticed in T_7, which was on par with T_5. During all the stages of composting, the treatments differed significantly from each other. Invariably, T_1 (CD alone) exhibited lower bacterial population (42.3, 45.3 and 17.3 x 10^6 CFU g^{-1} of compost) at all the three stages of vermicomposting process.

The fungal population was increased up to 60th day of vermicomposting and thereafter there was a significant reduction in population till the end of composting process (90th day of composting). At 30 and 60th day of vermicomposting process, T_7 [CD+ leaf litter+ MLSS (1:1:2)] registered significantly higher fungal population (32.0 and 55.6 x 10^4 CFU g^{-1} of compost) and it was closely followed by T_5 [CD+MLSS (3:1)]. At 90th day of composting, higher fungal population of 16.6 and x 10^4 CFU g^{-1} of compost was recorded in T_7, which was on par with T_5. In all the three stages of vermicomposting process, T_1 (CD alone) noticed the lowest fungal population (18.3, 41.3 and 13.0 x 10^4 CFU g^{-1} of compost). The interaction was significantly different from each other. As in the case of bacterial and fungal, the actinomycetes population was also increased up to 60th day of vermicomposting and decreased subsequently as the vermicomposting progressed. At 30, 60 and 90th day of composting, T_7 [CD+ leaf litter+ MLSS (1:1:2)] was noticed significantly higher actinomycetes population (42.3, 52.6 and 37.0 x 10^2 CFU g^{-1} compost) and it was closely followed by T_5 [CD+MLSS (3:1)]. The treatment T_1 (CD alone) registered the least actinomycetes population (19.0, 41.0 and 20.0 x 10^2 CFU g^{-1} compost) at all the stages of composting. The interaction effect was found to be significant. The microbial population was maximum in T_7 [cow dung +leaf litter + MLSS (1:1:2)]. This might be due to addition of MLSS, which are rich in microbial population and receiving high temperature during the process of composting. The population of bacteria outnumbered other microorganisms. This is in accordance with the findings of Davis *et al.* (1992), who observed that the number of bacterial colony forming units were higher than fungal colony forming units during composting process. The reduction of microbial population was observed in the end of the vermicomposting process. This result is in confirmation with the findings of Aira *et al.* (2007), who reported that the reduction in microbial biomass by earthworms has no effect on microbial activity and the passage through earthworms gut favoured the appearance of a reduced but more catabolically active microflora.

Conclusion

The results demonstrated vermicomposting as an alternate technology for the management of biodegradable organic wastes. Earthworms increased enzyme activity during vermicomposting of MLSS independently of the rate of application of MLSS and cow dung. This increase was indicating substrate availability for extracellular enzyme activity.

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