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Heterotrophic microbial count during vemicomposting of municipal solid waste

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Vermicomposting is a non-thermophilic biodegradation of organic material through interaction between earthworm and micro-organisms resulting in production of vermicompost. This study emphasizes on the heterotrophic microbial count during vermicomposting of the organic content of municipal solid waste. The heterotrophic microbial count (bacteria, fungi and actinomycetes) of the vermicomposts of municipal organic solid waste (MOSW) and agricultural waste (AW) comprising of crop residue was done for about 90 days taking the microbial count at an interval of 15 days till 90 days. Two species of earthworms viz *Eisenia fetida* and *Eudrilus euginae* were used for vermicomposting. It was observed that there was a marked increase in the above mentioned microbial count after inoculation of earthworms. The bacterial count of MOSW increased steadily in both species of worms. A similar pattern was also followed in the case of AW. However, the fungal and actinomycetes count had fluctuations which differed in both species in the respective substrates.

Key words: Vermicomposting, earthworm, actinomycetes count, microbial count, municipal solid waste.

INTRODUCTION

The process of vermicomposting involves the bioxidation and stabilisation of organic matter by the compound action of earthworms and microorganisms. Thus, it results in the bioconversion of the waste stream into two useful products, earthworm biomass and vermicompost. The vermicompost does not contain any disease causing pathogens like *Salmonella*, *Escherichia coli* or parasitic worm eggs, or pathogenic bacteria. Vermicompost has been found to be a suitable system for studying the symbiotic relationships of earthworms with microorganisms (Aira et al., 2006). The earthworms not only digest and release the substances (Brown and Doube, 2004), which can be assimilated by plants, but also help in the dispersal of microorganisms (Tiunov and Scheu, 2000; Haynes et al., 2003; Edwards, 2004). Vermicompost is an excellent product since it is homogeneous, has reduced level of contaminates, has plant growth hormones, higher level of soil enzymes, greater microbial population and tends to hold more nutrients over a longer period without adversely affecting the environment. Compost application is found to be most important in increasing soil microbial biomass and

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enhancing microbial diversity (Nair and Ngouajio, 2012).

The earthworms modulate the substrate by digesting, thereby increasing the surface area for microbial activity (Lavelle and Spain, 2001; Kaushik et al., 2008). The mucus secreted by earthworms also has a stimulating effect on the microbes (Doube and Brown, 1998).

Bacteria, fungi, actinomycetes are the chief microorganisms that play a key role in the process of vermicomposting. The bacterium helps in the metabolisation of the raw organic material. Actinomycetes and fungi help in the degradation of complex organic molecules such as cellulose, lignin, chitin and proteins (Aira et al., 2006). These fungi play a crucial role in the mineralisation process; apart from the fact that they are associated with medicinal plants, mostly the endophytic fungi (Rajagopal et al., 2011). These microbes break down the complex organic compounds and absorb the simpler compounds into the cells. As a result of this, the nutrients such as nitrogen, phosphorus and potassium are released and recycled in various chemical forms.

In the present study, vermicomposting of the organic component of the municipal solid waste and vermicomposting of crop residue was done employing 2 species of earthworms viz *Eisenia fetida* (*Ef*) and *Eudrilus euginae (Ee)* for about 90 days. At an interval of every 15 days, the bacterial, fungal and actinomycetes count were done.

MATERIALS AND METHODS

Two types of samples were collected: the organic component of the MSW and the agricultural waste mainly comprising of crop residue. The municipal organic solid waste (MOSW) was collected from the main garbage dumping site of Cuttack, that is, Chakradharpur. The waste was sorted out and the organic matter was collected for the study. The agricultural waste was collected from the crop residue. Both substrates were air dried separately for 48 h and pre-composted prior to vermicomposting and composting process. Water was then sprinkled to maintain 40-50% moisture level of the substrate and was allowed to undergo preliminary microbial decomposition for 20 days.

From these pre-decomposed substrates, 100 g of sub-samples were drawn, homogenized and analysed for general and plant beneficial microbial communities. The process of vermicomposting was carried out for a period of 90 days. After about 30 days, the earthworms were inoculated in both substrates and the microorganisms count was done at an interval of every 15 days till 90 days. The detailed process of pre-composting and vermin-composting are given by Pattnaik and Reddy (2009).

Samples of 10 g (fresh weight) of substrates were serially diluted in 90 ml Ringer's solution up to 10-7 dilution and 1 ml of aliquot was pour plated in different media.Total heterotrophic bacteria, fungi and actinomycete present in the samples of MOSW and agricultural waste (AW) were isolated by pour plate technique and total counts were recorded. The samples were inoculated in different culture media; the media used for the growth of bacteria, fungi and actinomycetes were sterile nutrient agar (Allen, 1959), Sabouraud dextrose agar and actinomycete isolation agar (Hi media), respectively and incubated at different time intervals- incubation period being 24 h at 37°C for bacteria, four days for fungi and seven days for actinomycetes. All media were prepared in distilled water and autoclaved at 15 psi pressure at 121°C for 20 min and accordingly the dilution factor was prepared.

The results of the microbial analysis were given as the CFU/g fresh weight of samples. Each CFU value was the average of 3 sample replicates. The number of colony forming units per gram of dry weight (CFU/g dwt) was calculated for the total microflora. The data was further processed into logarithmic form for standardisation and then analysis was done.

Statistical analysis of the microbial populations was carried out through analysis of variance (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

In the MOSW, there was a gradual increase in the bacterial count from the 0^{th} day onwards. It almost increased to 2 folds on the 30^{th} day. After the 30^{th} day when the earthworms were inoculated, both species had a steady increase till 90 days (Figure 1a). In the case of AW, it was also observed that the count increased to 1.5 folds on the 30th day. The bacterial count followed the same pattern as in the case of MOSW in both species depicting a steady increase (Figure 1b).

The fungal count in the case of MOSW increased to 2 folds and 1.88 folds in case of AW. However, in MOSW, the count had a fluctuation. In the case of *Ef*, there was an increase from 45^{th} day to 60^{th} day, and then a slight increase was marked on the 75^{th} day which again increased on the 90^{th} day. However in the case of *Ee*, same pattern was followed till 75^{th} day but it progressively decreased on the 90^{th} day (Figure 2a). On the other hand, in the case of AW, it showed a decreased count from 45^{th} to 60^{th} day, but then increased till the 90^{th} day. But *Ee* had a different pattern; from 45^{th} day till 75^{th} day there was a gradual decrease which later on increased on the 90^{th} day (Figure 2b).

The actinomycetes count in the case of MOSW showed a steady increase from the 30^{th} day onwards till 90^{th} day in both species of earthworms. A similar pattern was observed in the case of *Ef* in AW. However, the actinomycetes count in *Ee* fluctuated. It first showed a decreased pattern from 45^{th} to 60^{th} day and then progressively increased till 75^{th} day but suddenly decreased thereafter (Figure 2c).

It was observed that the microbial biomass (bacteria, fungi and actinomycetes) was higher after the inoculation of earthworms in both substrates (Subler et al., 1998; Edwards, 2004). This can be attributed to the fact that the high levels of ammonia and partially organic matter in casts and mucus of earthworms that was excreted along with the casts, stimulates the microbial growth and activity in the vermicomposts (Aira et al., 2006).

Conclusion

The present study demonstrates that vermicomposting boosts the growth of beneficial microorganisms. The bacterial count showed a steady increase in both substrates and with both the species. Thus, this shows that the microorganisms play a pivotal role in the

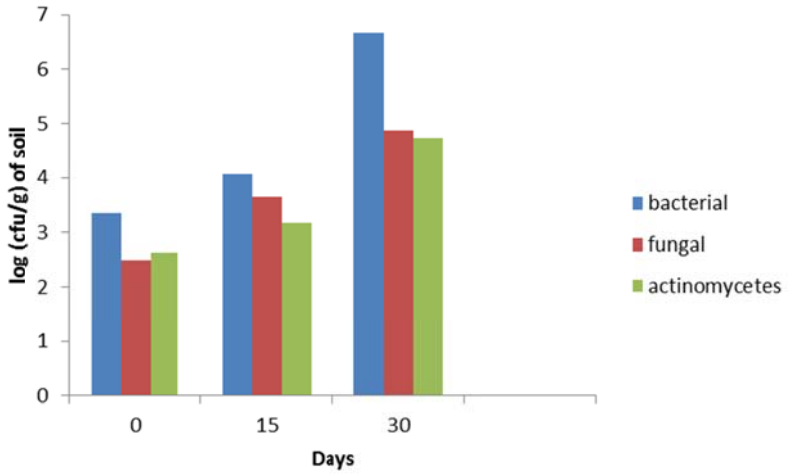

Figure 1a. Microbial count in the MOSW before inoculation of earthworms.

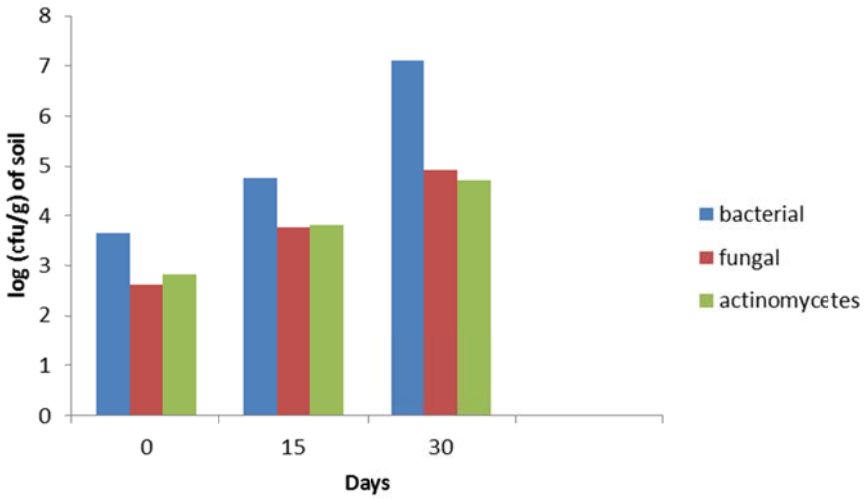

Figure 1b. Microbial count in the AW before inoculation of earthworms.

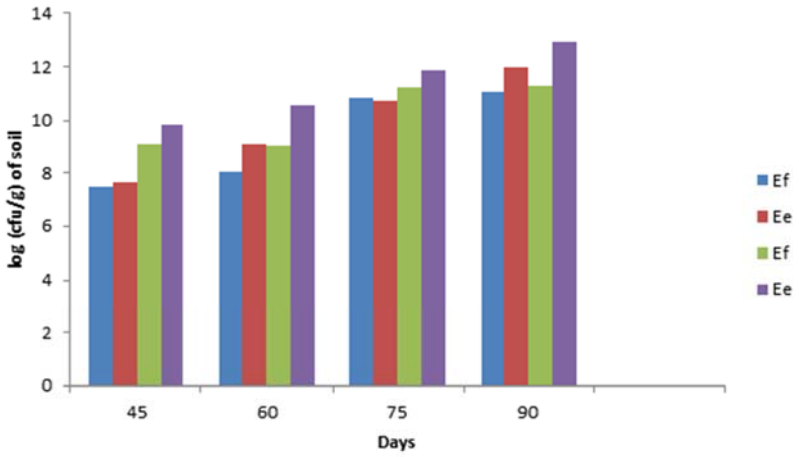

Figure 2a. Bacterial count in both the substrates (MOSW and AW) after inoculation of 2 types of earthworms from 45th day up to 90th day at an interval of 15 days.

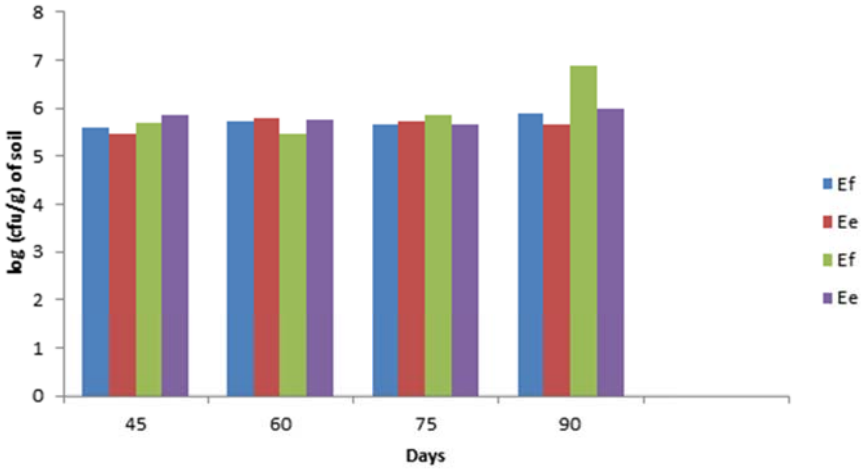

Figure 2b. Fungal count in both the substrates (MOSW and AW) after inoculation of 2 types of earthworms from 45th day up to 90th day at an interval of 15 days.

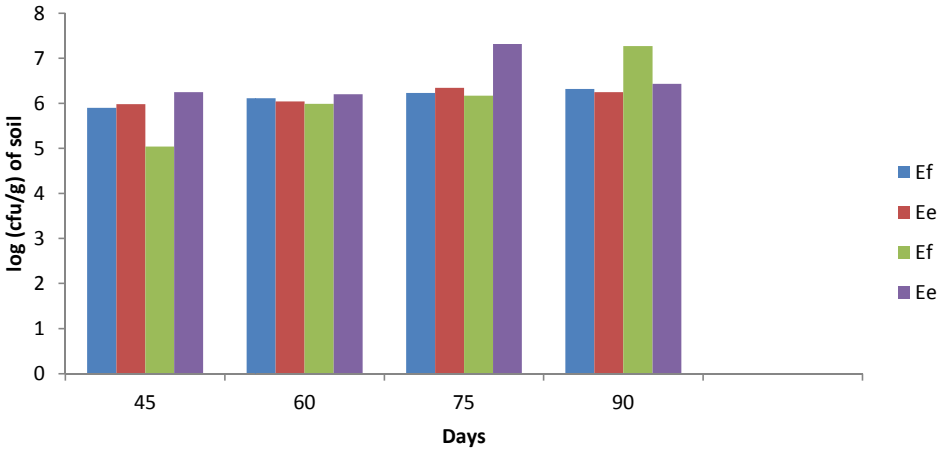

Figure 2c. Actinomycetes count in both the substrates (MOSW and AW) after inoculation of 2 types of earthworms from 45th day up to 90th day at an interval of 15 days.

breakdown of the complex organic compounds to simpler forms along with the earthworms. The microbes in a way help in alleviating the toxic properties rendered by the MSW during vermicomposting, thus making it more suitable for other fields of application.

Conflict of interests

The authors did not declare any conflict of interest.

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