



# Quantitative and Qualitative Distribution of Bacteria in Vermicompost Produced by Different Organic Wastes

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## ABSTRACT

Earthworms are an important component of the soil macrofauna and represent 82% of the total biomass in tropical zones with a precipitation above 1000mm. Earthworms are soil vertebrates which play a key role in recycling of organic matter in soils. Anecic, epigeous and endogenous earthworms stimulate or inhibit the growth of bacteria of agriculture importance inside their digestive tracts. Aerobic and anaerobic bacterial count of viable microorganisms in vermicompost produced by exotic earthworm *Eisenia fetida* has been studied in the present paper. The vermicompost was produced by different types of wastes for example cow dung, kitchen waste, petha waste and agricultural waste. Number of bacteria was higher in earthworm casting than in ingested soil samples. Bacterial count was obtained by standard microbiological procedures on the basis of their morphological and biochemical characteristics. Results reveal that the maximum bacterial count obtained was through mixing cow dung and petha waste ( $75 \times 10^7$  cfu/g). Isolated bacteria were identified as belonging to genus *Bacillus*, *Pseudomonas*, *Flavobacterium*, *Vibrio*, *Clostridium*, *Mycobacterium* and *Azotobacter*. These bacteria inhabit the soil and develop considerably when there is easily degradable organic matter.

## INTRODUCTION

According to an estimate India produce about 3000 million metric tones of organic waste annually which is result of the rapidly increasing population and high rate of industrialization, and these wastes are disposed off by ocean dumping, incineration and land application (Tiwari 2002). Earthworms are soil invertebrates which play a key role in recycling organic matter in soils. They are also called as “ecosystem engineers” as they actively redesign the physical structure of soil environment by their activities of ingesting litter and soil particles, depositing casts on soil surface and translocating soil particles. Wastes from domestic agriculture, urban and industrial sources are the main cause of organic soil pollution (Gajalakshmi 2004). It is an environmentally sound way to reduce organic waste and produce organic fertilizer or soil conditioner. Like nutrients, depending on the type of crop residues and earthworm species used, the microbial load of the vermicompost also varies. Although vermicomposting is microbiological process, little is known about microorganisms involved and their activities during the specific phases of composting process. Standardized microbiological analysis may potentially be used for judging the quality and maturity of compost residue. A laboratory study was conducted to enumerate the bacterial count obtained from different kinds of waste residues at different time intervals by the earthworm *Eisenia fetida*.

## MATERIALS AND METHODS

The laboratory experiments were conducted in Microbiology and Nanotechnology Laboratory, Department of Botany, R.B.S College, Agra during 2009-10 by following standard techniques.

**Collection of waste:** The organic solid wastes selected for the study were kitchen waste, cow dung and agro-residues obtained from RBS College, Agra and from its farm campus. Petha waste was collected from petha manufacturing market lanes. Petha (*Benincasa hispida*) is a long creeper plant of Cucurbitaceae family. Petha (a kind of sweet) makers use the pulp of the fruit through 1.5 inch thick and 6-8 inch long peel, hard in texture and light green in colour. Only the mantle of 5 cm diameter is used for making petha and rest is thrown off with seeds (Fig. 1). Petha waste is the largest city waste of petha industry of Agra city. It produces enormous amount of organic waste approximately 20-40 tones daily and causes obnoxious putrefying odour in thickly populated area of the city and market. Methodology for successful conversion of petha waste into vermicompost was one of the most important aspects of the present work, for depolluting the city. It is felt that careful treatment of petha waste and other waste will give beneficial and cost effective manure.

**Collection of earthworms:** *Eisenia fetida* species of earthworm (Fig. 2) was obtained from Vermiculture Research



Fig. 1: Discarded part of petha fruit used for vermicomposting.

Fig. 2: *Eisenia fetida*.

Fig. 3: Worm bin used in the experiment.



Fig. 4: Vermicompost with earthworms in it.

Station, D.S. College, Aligarh, U.P, India.

**Experimental Setup:** The experiments were conducted on the farm campus of R.B.S College, Agra in earthen pots having 5 kg of waste with different amendments in ratios 1:1. In the treatment pots (Fig. 3), 100 earthworms were introduced after 8-10 days of partial decomposition of the organic wastes.

Moisture level was maintained at 60-70% and to avoid direct sunlight the worm bins were covered by gunny bags. There were three replicates for each feed mixture. The vermicompost samples (Fig. 4) were analysed for their total bacterial count and were determined by serial dilution plate count method (Johnson et al. 1959) on Nutrient Agar medium. The bacterial colonies appeared (Fig. 5) were purified by streak plate method and maintained on the nutrient agar slants by keeping them in refrigerator. Isolates were identified by various biochemical tests and observing the colonies under microscope.

## RESULTS AND DISCUSSION

Three amendments, i.e. petha waste : cow dung, kitchen waste : cow dung and agricultural waste : cow dung were selected.

The results revealed that the vermicompost prepared from the amendment 1, i.e. petha waste : cow dung showed maximum total bacterial count, which was  $75 \times 10^7$  cfu/g as showed in Fig. 6 and Tables 1-3. It varied from  $29 \times 10^7$  to  $75 \times 10^7$  cfu/g with minimum in the beginning and maximum at 45<sup>th</sup> day. In amendment 2, i.e., kitchen waste : cow dung showed total bacterial count of  $31 \times 10^7$  to  $60 \times 10^7$  cfu/g with minimum on the first day and maximum between 35-45 days. In amendment 3, i.e. agricultural waste : cow dung total bacterial count varied from  $32 \times 10^7$  to  $55 \times 10^7$  cfu/g with minimum in the beginning and maximum at 45<sup>th</sup>

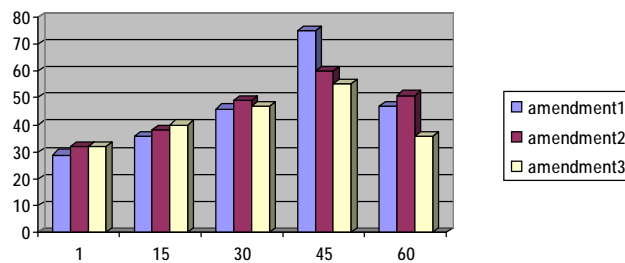


Fig. 1: Comparative biodiversity of the three amendments at 1<sup>st</sup>, 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup> and 60<sup>th</sup> day of observation.

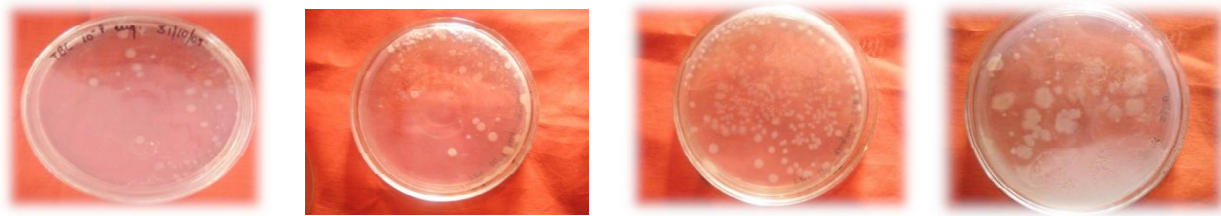


Fig. 5: Diversity of bacterial population at 0<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup> and 60<sup>th</sup> day respectively during vermicomposting.

Table 1: Quantitative and qualitative spectrum of bacterial population during vermicomposting through amendment 1.

S. No	Bacterial parameters	1 day	15 day	30 day	45 day	60 day
<b>Bacterial isolates (cfu/g)</b>						
1	<i>Acromobacter</i> sp.	4	2	-	-	-
2	<i>Azotobacter</i> sp.	2	4	5	10	6
3	<i>Bacillus bacilli</i>	4	7	6	8	7
4	<i>Bacillus subtilis</i>	-	-	-	-	6
5	<i>Bacillus</i> sp.	-	-	8	10	-
6	<i>Micrococcus</i> sp.	6	8	10	12	11
7	<i>Pseudomonas</i> sp.	-	4	8	12	10
8	Unidentified Gram +ve	7	5	5	8	3
9	Unidentified Gram -ve	6	6	4	5	4
~	Total population (cfu/g* × 10 <sup>7</sup> )	29	36	46	75	47

\* cfu/g - colony forming unit/g

Table 2: Quantitative and qualitative spectrum of bacterial population during vermicomposting through amendment 2.

S. No	Bacterial parameters	1 day	15 day	30 day	45 day	60 day
<b>Bacterial isolates (cfu/g)</b>						
1	<i>Acromobacter</i> sp.	4	3	-	-	-
2	<i>Azotobacter</i> sp.	2	3	5	7	8
3	<i>Bacillus bacilli</i>	3	5	6	8	7
4	<i>Bacillus subtilis</i>	-	2	-	2	6
5	<i>Bacillus</i> sp.	1	-	9	7	-
6	<i>Micrococcus</i> sp.	5	8	10	11	9
7	<i>Pseudomonas</i> sp.	-	5	8	8	10
8	Unidentified Gram +ve	9	6	5	9	5
9	Unidentified Gram -ve	8	6	6	8	6
~	Total population (cfu/g* × 10 <sup>7</sup> )	32	38	49	60	51

\* cfu/g - colony forming unit/g

day. Hence, through this we can conclude that when cow dung is mixed with petha waste then we obtained maximum bacterial count.

Though information about the microbial load of vermicompost is meagre, Giraddi (2007) reported the beneficial microflora in vermicompost such as fungi, bacteria and actinomycetes ( $2.65 \times 10^4$ ,  $11.37 \times 10^7$  and  $10.43 \times 10^4$  cfu/g, respectively), which resembles our data for bacteria.

Meenatchi et al. (2009) reported the bacterial count obtained from *Perionyx excavatus* with having waste such as paddy straw, soybean post harvest waste, kitchen waste and weed waste ( $16 \times 10^6$ ,  $65 \times 10^7$ ,  $18 \times 10^7$  and  $12 \times 10^7$  cfu/g).

*Achromobacter* sp. is found in initial stages on 0<sup>th</sup> and 15<sup>th</sup> day. Some bacteria like *Azotobacter*, *Micrococcus* and *Bacillus* were found throughout the experiment and some bacteria in the last stages of experiment like *Bacillus subtilis*. Some unknown species of Gram +ve and Gram -ve bacteria were also found. *Pseudomonas* sp. is not found initially but later found throughout the experiment.

In the absence of sufficient number of reports, comparisons of either substrate effect or earthworm influence on beneficial microflora of vermicompost cannot be made. However, the present results will act as baseline information for future study to throw much light on this aspect

Table 3: Quantitative and qualitative spectrum of bacterial population during vermicomposting through amendment 3.

S. No	Bacterial parameters	1 day	15 day	30 day	45 day	60 day
<b>Bacterial isolates (cfu/g)</b>						
1	<i>Acromobacter</i> sp.	4	3	-	-	-
2	<i>Azotobacter</i> sp.	2	5	6	9	3
3	<i>Bacillus bacilli</i>	3	5	7	9	5
4	<i>Bacillus subtilis</i>	-	-	-	-	6
5	<i>Bacillus</i> sp.	1	-	8	7	-
6	<i>Micrococcus</i> sp.	5	8	9	8	5
7	<i>Pseudomonas</i> sp.	-	5	7	10	3
8	Unidentified Gram +ve	9	6	4	6	6
9	Unidentified Gram -ve	8	8	6	6	8
~	Total population (cfu/g* $\times 10^7$ )	32	40	47	55	36

\* cfu/g - colony forming unit/g

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