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Influence of Aeration through Turning on Microbial Activity and Decomposition Rate in Vermicomposting

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

This study was implemented to investigate effects of turning (aeration) on microbiological activity and rate of decomposition in vermicomposting. Aeration is a key element in composting which provides oxygen to the aerobic organisms during composting. Oxygen also has the important function of controlling temperature as well as removing excess moisture and unhygienic gases. If the oxygen supply is limited, the composting process might turn anaerobic, which is much slower and odorous process. Such maintenance of aerobic condition has some additional significance for vermicomposting because the earthworms, which play vital role in this kind of composting, are essentially dependant on availability molecular oxygen in the system. Although some information is

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available on the beneficial effects of maintaining aerobic conditions in composting chambers through turning the piles during traditional composting yet such information with regard to vermicomposting is not available. In this experiment, therefore, the effect of turning the vermicomposting materials on nature and the behaviour of microorganisms and also on the rate of decomposition were studied. The result of the study will be hopefully helpful for maintaining aerobic condition in the vermicomposting system, thus improving the pace of vermicomposting and the quality of the product.

Keywords: Vermicomposting; aeration; unhygienicgas; cowdung; Eisenia foetida; microbial biomass carbon; microbial respiration; Aerobic bacteria; Anaerobic bacteria; cation exchange capacity.

1. INTRODUCTION

Composting may be defined as a biological process in which diverse and mixed group of microorganisms break down organic material to humus like substances [1]. Since aeration forms an important factor controlling the composition and behaviour of microorganisms in any system, the dynamics and magnitude of organic matter degradation during composting is also likely to be governed significantly by the degree of aeration in the composting substrate. Since aerobic microorganisms are plenty in numbers and diverse in nature in the environment and are also more active than the anaerobes, maintaining an aerobic environment in the composting system has been suggested by Gaur and Singh [2] and others. Such aerobic conditions not only accelerate the decomposition process but also maintains an hygienic condition during composting by restricting different reduction reactions [3]. Maintenance of an aerobic environment assumes more importance in case of vermicomposting owing to aerobic nature of the earthworms and also their gut microorganisms. Under traditional system of vermicomposting, epigeic earthworms, released to the wastes, do not generally enter deep into the organic materials owing to restricted availability of molecular oxygen [4,5]. This results in larger accumulation of the worms and also the cast microorganisms in the upper surface of the substrates. This behaviour is reflected in spatial variation in the rates of decomposition of the organic wastes at different depths. While the outer layer, which remains under aerobic condition is composted faster due to more intense microbiological activity, the deeper horizons, suffering from lack of oxygen, may undergo anaerobic decomposition which proceeds at much slower rate. Under such situation, periodic turning of the composting material has been suggested as an effective practice for providing aeration to entire mass of organic matter [3]. Beneficial effects of periodic turning of the organic materials on the rate of composting are long being utilized under "Indore" method of composting [2]. However, information on the effects of maintaining aerobic condition in the substrates through periodic turning under vermicomposting system is meager. In the present study, therefore, an attempt has been made to study the effects of turning of the substrate on microbial behaviour and maturation of the compost under vermicomposting system.

2. METHODOLOGY

The study was carried out in earthen pots under yard condition. Mixture of cow dung and straw at 1:1 (W:W) ratio was used as the substrate. To maintain different levels of aeration in the composting system, turning of the substrates at different time intervals viz. control (no turning) and turnings at 28, 21, 14 and 7 days intervals were practiced. Each treatment was incubated with epigeic earthworm *Eisenia foetida* @ 10 worms per kg of substrate for a period of 60 days. Microbial biomass carbon (MBC) and basal respiration rates were estimated at 15 days intervals for assessing the variations in microbiological activity. On the other hand, cation exchange capacity (CEC) of the composting medium was used as the index of the extent of humification. For developing a better understanding about the microbiological behaviour in the substrates, occurrences of aerobic and anaerobic bacteria were estimated on 30th day of incubation.

2.1 Analytical Methods

Cation exchange capacity (CEC) values of the samples were determined by following the method of Harada and Inoko[1]. In addition to these common properties, several other parameters were studied for different studies, depending on the needs of the experiments. In each case, Microbial Biomass Carbon (MBC) was estimated by following the chloroform fumigation method as described by Vance *et al*. [6]. Here two parts of the organic waste sample, each weighing 2g were taken separately in a 50ml beaker and a 250ml conical flask fitted with stopper. The beaker was placed in a vacuum desiccator containing a vial of 10g soda lime and a beaker containing 25ml ethanol free chloroform. The desiccator was then evacuated until the chloroform boiled for 2 minutes. The desiccator with the content was then incubated at 25^0C for 24 hrs. After fumigation, the beaker containing chloroformed organic waste sample was then transferred to a 250 ml conical flask. Both the sets of fumigated and unfumigated samples were extracted with 100 ml 0.5M K_2SO_4 after shaking for 30 minutes and then filtered. The filtrates were finally used for determination of organic carbon with the help of standard potassium dichromate in presence of strong acid mixture (H_2SO_4 : H_3PO_4 :: 2:1 V/V). The difference between fumigated and unfumigated organic carbon value was used to calculate the microbial biomass carbon using a calibration factor and expressed as µg of MBC g^{-1} of sample. Microbial respiration was estimated by following a modified method of Alef [7]. For this purpose, 10g of the organic waste material was taken in 1 L conical flask and wetted to 50% of it's maximum water holding capacity. A vial containing 10ml of 0.5M NaOH was hanged inside the flask and the mouth of the flask was closed with a cork. The flask was incubated at 25^0C for 24 hrs. The CO_2 evolved during this period was determined by titrating the residual NaOH with 0.1M HCl. Occurrence of aerobic and anaerobic bacteria were carried out by following the method described by Dubey and Maheswari [8]. Compost samples were diluted by Serial Dilution technique and the inoculums from 10^{-5} dilution tube were aseptically speeded on solidified nutrient agar plates in triplicate using standard spread plate technique in a laminar air flow hood and the plates were placed in an incubator within a bell jar with a lighted votive candle inside and the bell jar was maintained by sealing the edges so as to ensure leak proof condition with paraffin wax .The burning of candle ensures presence of O_2 inside. As soon as the candle dips off, it indicates that all the O_2 in the jar has been utilized thus creating anaerobic condition there. The plates were then incubated at 30 ± 2^0C for 48 hrs and observed for growth of anaerobic bacteria. The occurrence of aerobic bacteria is estimated using similar method but without using the anerobic jar. Statistical analysis of the results were carried out by using MS-Office-Excel-2007 software packages.

3. RESULTS AND DISCUSSION

The values of microbial biomass carbon (MBC) indicating the gross occurrence of microorganisms in the composting media under different treatments have been presented in Table 1. As observed from the table, the MBC values were the highest during the initial phase of the study in all the treatments due to intense microbial activity and proliferation of the microorganisms in presence of sufficient amount of food materials. The values declined gradually as the composting proceeded due to consistent reduction in easy availability of food materials. However, there were considerable variations in MBC values among the treatments. The treatment with turning at 7 days interval showed maximum MBC value at 15^{th} day of observation and was followed by the treatment with turning at 14day interval. These two treatments received turning within 15 days of incubation and hence, maintained more aerobic condition than other treatments facilitating more abundance of microorganisms. The other treatments which did not receive any turning during this period showed lesser occurrence of microorganisms probably owing to lesser availability of molecular oxygen in the system. With 30 days of incubation, the treatments bearing turning at 7, 14 and 21 days showed higher MBC values, as compared to other two treatments. Although the treatment with turning at 28 days interval received a turning within this period, yet this treatment showed comparatively lesser MBC value. Since the turning was done only 2 days before the sampling, probably the proliferation of the microorganisms did not appear to the much apparent within this short period. With further extension of the incubation period, the MBC values declined in all the treatments owing to gradual humification of the substrates. Among the treatments, regular turning at 7 days interval maintained highest mean MBC values and this was followed by turnings at 14 days, 21 days and 28 days respectively (Fig. 1). The control treatment, which did not receive any turning,exhibited the lowest mean MBC value, showing that the magnitudes of aeration in the composting media, as were affected by periodic turnings of the substrate, exerted considerable influence on gross microbial biomass in the composts.

Table 1. MBC (µg g^{-1}) of dry sample in different treatments during the period of incubation

Treatments	MBC at 15 days intervals	MBC at 30days intervals	MBC at 45 days intervals	MBC at 60 days intervals
No Turning (Control)	470.23	371.225	218.835	151.865
T. at 7 days intervals	689.0025	606.7125	490.1625	231.03
T. at 14 days intervals	660.4925	530.555	418.1125	221.8375
T. at 21 days intervals	442.8425	522.6625	389.3875	181.1025
T. at 28 days intervals	492.4	384.9025	374.4025	158.145
C.D.	**67.13825**	**58.54048**	**82.34**	**41.67328**

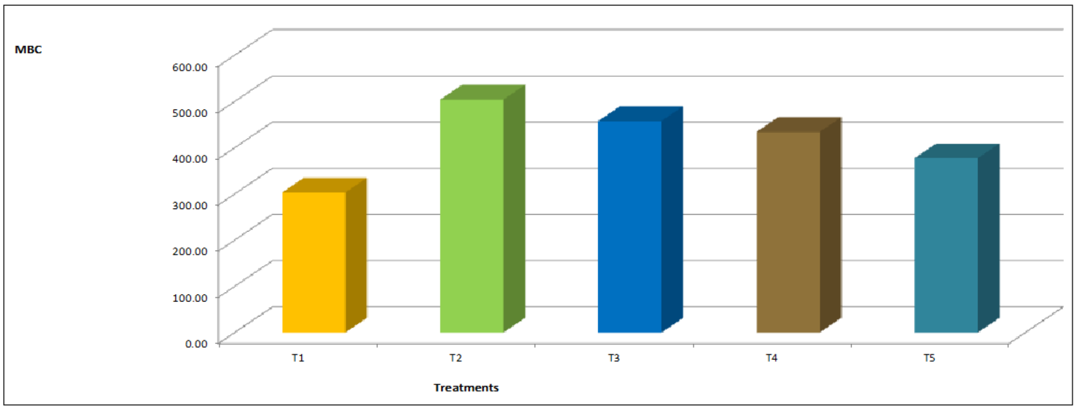

Fig. 1. Average MBC (µg g^{-1}) of oven dry sample in different treatments

The changes in basal respiration values, which indicate the rate of microbiological activity in the organic matter under composting [9] have been presented in Table 2 and in (Fig. 2). The values showed a close relationship with the MBC values of the substrates. This was obviously owing to the direct relationship between the biomass of the microorganisms and the oxygen consumed during their respiration. Highest rate of microbial respiration was observed in case of the treatment with turning at 7 days intervals. Such frequent turning of the substrate helped to maintain highly aerobic condition in the composting medium which was more congenial for microbial proliferation and activity. Since these aerobic microorganisms act in morefaster pace, the basal respiration values appeared to be considerably higher under this treatment. On the other hand, the control treatment, which did not receive only turning, showed the lowest basal respiration rates throughout the period of incubation. Other treatments, which received intermittent turnings, also showed basal respiration values in between these two values. The results support the observations on the MBC values of the substrates that microbial activity under vermicomposting became more prominent with the frequency of turning, thus promoting aeration in the vermicomposting systems [10].

Table 2. Microbial respiration (mg. CO_2 g^{-1} of dry sample hr^{-1} at 25° C) in different treatments during the period of incubation

Treatments	Microbial respiration at 15 days intervals	Microbial respiration at 30days intervals	Microbial respiration at 45 days intervals	Microbial respiration at 60 days intervals
No Turning (Control)	1.19	0.44	0.14	0.79
T. at 7 days intervals	2.34	2.48	2.53	1.56
T. at 14 days intervals	2.22	2.34	2.38	1.98
T. at 21 days intervals	1.20	2.14	2.25	1.47
T. at 28 days intervals	1.24	2.06	2.00	1.43
C.D.	**0.17**	**0.36**	**0.31**	**0.28**

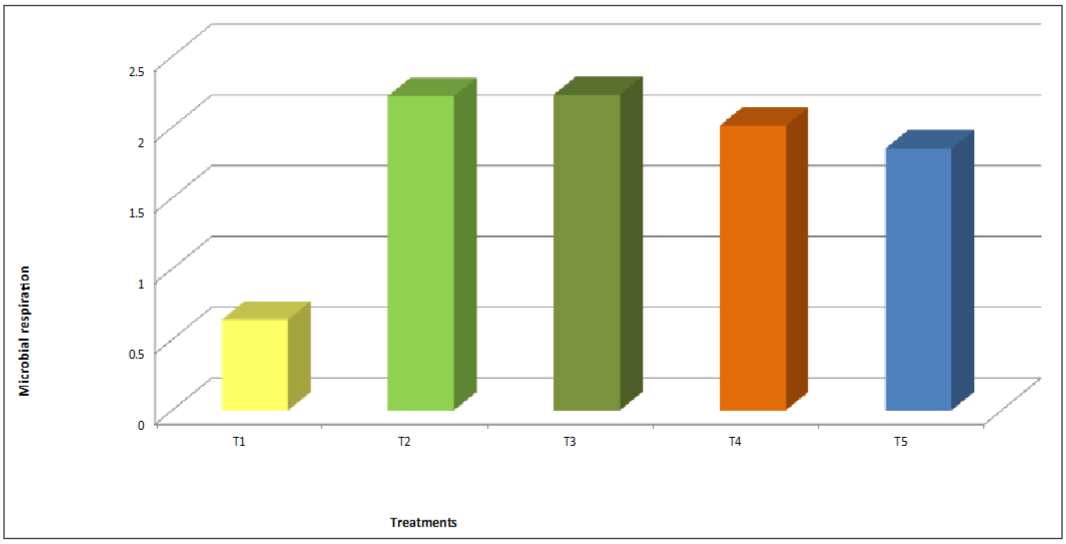

Fig. 2. Microbial respiration (mg. CO_2 g^{-1} of oven dry sample hr-1 at 25^0C) in different treatments

To support these observations further, a midlevel study was carried out on 30th day of incubation to assess the relative occurrence of aerobic and anaerobic bacteria in the substrates under different treatments (Fig. 3). As observed from the figure, total colony forming units of the bacteria were the highest in case of the treatment with turning at 7 days interval. Dominance of aerobic bacteria was also found to be more prominent under this treatment and this was followed closely by the treatment receiving turning at 14 days interval. The control treatment and the treatment with 28 day turning, on the other hand, showed comparatively lower bacterial population. The ratios between aerobic and anaerobic bacteria were also much narrower under these two treatments obviously owing to dominance of anaerobic condition in these two treatments. It may be mentioned once again, that the MBC values and basal respiration rates were also lowest in these two treatments.

The effects of such variations in microbial activity in the organic wastes under different treatments were reflected in the rate of humification also, as indicated by the cation exchange capacity (CEC) values of the substrates (Table 3). CEC values increased consistently with the period of incubation under all the treatments owing to gradual humification of the organic wastes, opening more cation exchange sites. The control treatment, without any turning of the substrate, showed the lowest CEC values, obviously owing to its slow rate of decomposition, as was evidenced by lower microbiological activity under this treatment. On the other hand, the treatment with 7 days interval which exhibited highest microbial activity and also maintained highest degree of aeration, as shown by the relative occurrence of aerobic: anaerobic bacteria, resulted in largest amount of humification, indicated by the CEC values. The treatments with turnings at 14 and 21 days intervals also showed appreciably high CEC values indicating good rate of humification. It has already been reported that microbiological activity as well as degree of aeration were also moderately high in these treatments. It was interesting to observe

Table 3.CEC (C.mol.(P^+) kg^{-1}) in different treatments during the period of incubation

Treatments	CEC at 15 days intervals	CEC at 30days intervals	CEC at 45 days intervals	CEC at 60 days intervals
No Turning (Control)	50.62	54.25	70.60	95.97
T. at 7 days intervals	127.57	130.07	133.52	143.82
T. at 14 days intervals	99.62	114.37	118.17	134.37
T. at 21 days intervals	88.78	104.25	107.72	124.10
T. at 28 days intervals	95.22	94.7	93.32	101.76
C. D.	**13.61**	**13.69**	**20.96**	**22.03**

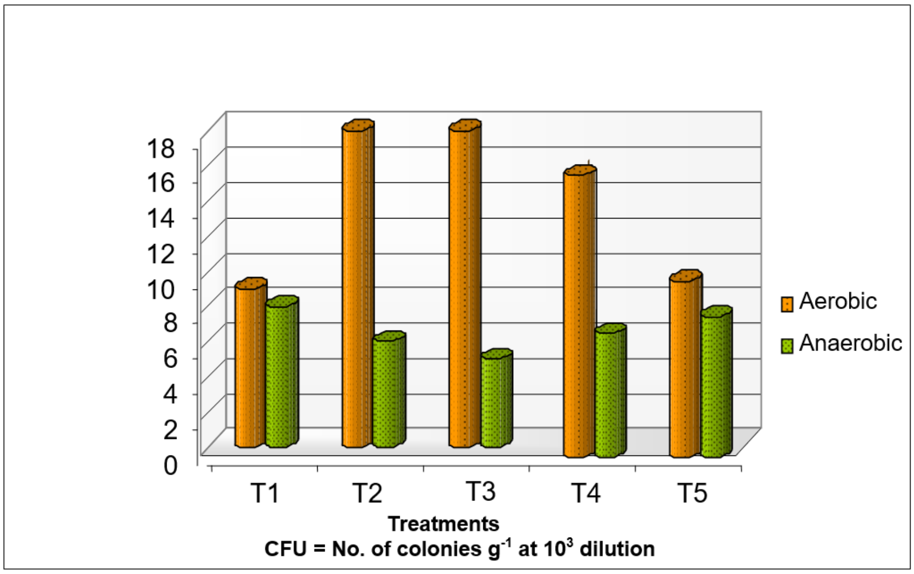

Fig. 3. CFU of aerobic and anaerobic bacteria in the substrate after 30 days of incubation

that the difference between CEC values under the treatment with 7 day turning and other treatments were statistically significant during earlier period of study. However, the differences became narrower during later phase, as the treatments with 14 and 21 days turnings also received several turnings and maintained moderate aerobic condition to accelerate the microbial activity and, thereby, the decomposition.

The results of the study thus indicated increase in the level of aeration through periodic turning of the organic substrates to be a major factor for encouraging the microbial activity and, thereby, the rate of humification in the vermicomposting system.

4. CONCLUSION

Vermicomposting is an aerobic system where earthworms and aerobic microorganisms exhibit synergistic activities. Maintaining a gross aerobic environment in the vermicomposting system, therefore, appears to play significant role in carrying out the degradation process. Since periodic turning of the substrates is likely to help in inducing aeration in the vermicomposting system and, thereby, to influence the decomposition of the wastes, another yard study was carried out during the first phase of the work programme to study the effect of periodic turning on the pace of vermicomposting. For the purpose of the study, organic wastes incubated in earthen pots were inoculated with epigeic earthworm *Eisenia foetida* and were subjected to different intervals of turning of the substrates viz. 0, 7, 14, 21 and 28 days. The microbial activity in terms of MBC and microbial respiration also the pace of vermicomposting with regard to changes in CEC values were assessed at periodic intervals. In addition, a midway estimation of the relative occurrence of aerobic and anaerobic bacteria in the vermicomposting system under different treatments was also carried out. The study revealed the frequency of turning of the substrate to influence the microbial activity and also the rate of degradation of the waste material in the vermicomposting system. More frequent turning helped in larger maintenance of aerobic condition in the composting chamber resulting in dominance of aerobic bacteria which, in turn, resulted in faster rate of vermicomposting.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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