

Conversion of leaf litter into compost by Effective Microorganisms (EM),
Bacillus subtilis, *Aspergillus niger* and their effect on
growth parameters of *vigna radiata* Linn

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ABSTRACT

In this present study, the Effective Microorganisms (EM) solution was prepared by using natural substrates such as banana, papaya, pumkin, egg and jaggery. It was extended and activated before used to plants. The extended EM was verified by pH (3.2) and by sweet sour smell. Leaf litter was used for composting, Effective Microorganisms (EM), *Bacillus subtilis*, *Aspergillus niger* were used for compost making in the six treatments (C, T1, T2, T3, T4 and T5). It was allowed for composting of 60-70 days. The compost samples from each treatment were collected. Physical and chemical properties of compost were analyzed using standard procedure. Then the effects of compost from each treatment were analyzed in the growth of green gram (*Vigna radiata*). From the results, the plants inoculated with the leaf litter treated with EM + *Aspergillus niger* and leaf litter treated with EM + *Bacillus subtilis* showed maximum effect.

Key words: Effective Microorganisms (EM), *Bacillus subtilis*, *Aspergillus niger*, Compost, *Vigna radiata* Linn.

1. INTRODUCTION

Farm technologies that sustain high productivity levels without impairing the soil health and water resources are the need of the day. However in the name of sustainable and environment friendly agriculture, some typical approaches of farming, collectively termed as organic farming are being promoted in the country advocating sole use of organic or biological nutrient resources and other inputs with complete exclusion on fertilizers and agrochemicals^[1]. A big significance for the process of composting represents the cell wall of microorganisms through which mass transfer is possible. Low molecular weight and water-soluble molecules can easily pass through the cell wall where they take part in the cell metabolism, providing energy and being built up into larger polymers, with the help of intracellular enzymes. To attack high molecular components, which cannot pass through the cell wall, microorganisms secrete extracellular enzymes. They break molecules down into the fragments that can be assimilated, while the rest is converted into a stable product, humus or compost^[2-3]. A microbial inoculants containing many kinds of naturally occurring beneficial microbes called Effective Microorganisms has been used widely in nature

and organic farming^[4]. The concept of Effective Microorganisms (EM) was developed in 1971 by Professor Teruo Higa, university of the Ryukyus, Okinawa, Japan. Effective Microorganisms (EM) solution is prepared from natural substrates and it can be used as an herbal insecticide to control insects and pathogenic microorganisms and can also used as plant growth inducers. The use of EM in crop production is either by direct application to the soil, to organic matter spread on the soil surface or to the plant. However, it can be used in composting organic matter at a different location and this material applied to the soil prior planting^[5]. Studies have suggested that EM may have a number of applications, including agriculture, livestock, gardening, and landscaping, composting, bioremediation, cleaning septic tanks, algal control and house hold uses. The present study was undertaken to investigate the conversion of leaf litter into organic compost by Effective Microorganisms (EM), *Bacillus subtilis*, *Aspergillus niger* and their effect on growth parameters of *Vigna radiata* Linn and to create an awareness among farmers about composting of organic waste and uses of EM solution in sustainable agriculture.

2. MATERIALS AND METHODS

2.1. Effective Microorganisms(EM) Preparation

250gm of Banana, Papaya and Pumpkin were chopped into small pieces and transferred into air tight container and mixed with one litre of bore well water, subsequently 250gm of jaggery and one egg was added. The barrel was closed tightly and incubated for 45 days. After 45 days, the white layer was formed on the surface. The fermented EM solution was collected by filtration. It is termed as EM stock solution.^[6]

2.1.1. Extension (EMe) and activation (EMa) of EM stock solution

One litre of EM stock solution and 1kg of jaggery were mixed with 20 litres of water. The water has to be clean and free from chlorine. The container should be of good-grade plastic. For the period of activation, the container was placed in shade at ambient temperature (20-40°C) without exposure to strong temperature fluctuations. Extended EM (EMe / EMa) will be ready after 5-10 days. It can be verified by a pH of 3.5 or lower and a pleasant sweet sour smell.^[7]

2.2. Composting

2.2.1 Sample Collection

Leaf litter was collected from Mannargudi, Thiruvavur District and allowed in the direct sunlight for reducing other microbes present. It was used for composting.

2.2.2. Experimental Design

The Effective Microorganisms (EM), *Aspergillus niger*, *Bacillus subtilis* were used for compost making in the following treatments.

Control: 5 kg of Leaf litter + urea (g/kg of waste)

T1: 5kg of leaf litter + 100ml of *Bacillus subtilis* culture + urea (g/kg of wastes)

T2: 5 kg of leaf litter + 100 ml spore suspension of *Aspergillus niger* + urea (g/kg of waste)

T3: 5kg of leaf litter + 100ml of EM solution

T4: 5 kg of leaf litter + 100 ml of *Bacillus subtilis* culture + 100ml of EM solution

T5: 5 kg of leaf litter + 100ml spore suspension of *Aspergillus niger* + 100 ml of EM solution

Composting was done by sandwich method in a pit (45cm height and 30 x 20 cm in diameter). The pits were covered with polythene sheet. Moisture content was maintained during composting by spraying water regularly. The compost will be ready after 60-75 days.

2.2.3. Compost Analysis

The representative compost samples were collected and air-dried. The following physical and chemical properties of the compost were analyzed.

- Organic carbon^[8]
- Total Nitrogen^[9]
- Cellulose^[10]
- Reducing sugar^[11]
- pH

2.3. Pot Cultivation

The seedlings of *Vigna radiata* Linn were transplanted in six pots of equal size, 30cm in height. Garden soil was used as the culture medium. The pots were provided with water facilities. The plant chosen for this study was green gram [*Vigna radiata* Linn]. Six treatments were done. The pots were maintained in the open shade at the temperature of 27-30°C.^[12]

2.3.1. Analysis of morphological Parameters

After 2 months of growth, 3 plants per pot were removed from all samples and studied for the following morphological parameters such as height of the plant (in cm), number of leaves (per plant), number of roots (per plant), and number of flowers (per plant).

2.4. Statistical Analysis

Mean was calculated to facilitate the comparison of the data of various physicochemical parameters of compost and growth parameters of green gram in all samples.^[13]

3. RESULT AND DISCUSSION

The effect of Effective Microorganisms (EM), *Bacillus subtilis*, *Aspergillus niger* on the composting of leaf litter was reflected on the total organic carbon and nitrogen content of the organic manure. The compost was ready after 60-70 days. After composting, the effect of compost on the growth of *Vigna radiata* was reflected on the maximum leaf number, maximum height, maximum root number and maximum number of flowers of the plants.

3.1. Compost Analysis

3.1.1. Effects of the EM, *Aspergillus niger*, *Bacillus subtilis* on C:N. ratio of compost

From the results, it is evident that there was a gradual decrease in C:N ratio from Control to Treatment – 5. The reduction of C:N ratio was high in Treatment – 5 (1.37±0.04) and Treatment 4 (1.44 ±0.05) compared to control (2.8±0.10). This is due to the fact that decomposing microorganisms such as *Bacillus subtilis*, *Aspergillus niger* along with EM actively utilize the carbon and nitrogen content compared to that of the control. In the case of fungal composting, C:N ratio of the harvested compost fall within.

Table -1: Effects of Effective Microorganisms (EM), *Bacillus subtilis* and *Aspergillus niger* on the C:N ratio of the compost.

Treatment	Organic Carbon (%)	Nitrogen (%)	C:N Ratio
Control	16.8 ± 1.41	5.8 ± 0.25	2.8 ± 0.10
T1	10.8 ± 1.25	6.0 ± 0.25	1.8 ± 0.08
T2	10.1 ± 0.67	6.0 ± 0.25	1.68 ± 0.06
T3	10.0 ± 0.53	6.2 ± 0.25	1.61 ± 0.06
T4	9.8 ± 0.20	6.8 ± 0.30	1.44 ± 0.05
T5	9.6 ± 0.20	7 ± 0.41	1.37 ± 0.04

Results are expressed as mean ± standard deviation

Table -2: Effects of Effective Microorganisms(EM), *Bacillus subtilis* and *Aspergillus niger* on cellulose, reducing sugar and pH of the compost.

Treatment	Organic Carbon (%)	Nitrogen (%)	C:N Ratio
Control	16.8 ± 1.41	5.8 ± 0.25	2.8 ± 0.10
T1	10.8 ± 1.25	6.0 ± 0.25	1.8 ± 0.08
T2	10.1 ± 0.67	6.0 ± 0.25	1.68 ± 0.06
T3	10.0 ± 0.53	6.2 ± 0.25	1.61 ± 0.06
T4	9.8 ± 0.20	6.8 ± 0.30	1.44 ± 0.05
T5	9.6 ± 0.20	7 ± 0.41	1.37 ± 0.04

Results are expressed as mean ± standard deviation

Table-3: Effects of compost on the morphological parameters of *Vigna radiata* Linn.

Treatment	Cellulose (%)	Reducing Sugar mg/g	pH	
			Before composting	After composting
Control	25.2 ± 0.40	11.67 ± 0.45	5.3	6.8
T1	19.12 ± 0.38	11.20 ± 0.43	5.2	7.5
T2	18.0 ± 0.38	10.72 ± 0.40	4.9	7.2
T3	15.2 ± 10.35	8.27 ± 0.40	3.8	7.2
T4	11.0 ± 0.27	6.63 ± 0.39	4.0	7.1
T5	9.13 ± 0.23	4.93 ± 0.37	4.2	7.3

Results are expressed as mean ± standard deviation

recommended C:N ratio of 20:1 reported by Reneickle et al., 1986 It is very important that the plants cannot assimilate mineral nitrogen unless the organic matter added to the soil.

3.1.2. Effects of the EM, *Aspergillus niger*, *Bacillus subtilis* on cellulose content of the compost

Decrease in cellulose content denotes the maturity of compost. Among all treatments, Treatment 5 showed low cellulose content (9.13%) than that of other treatments. Control showed maximum cellulose content (25.2%) (Table-2).

3.1.3. Effects of the EM, *Aspergillus niger*, *Bacillus subtilis* on reducing sugar of the compost

Treatment-5 showed low amount of reducing sugar (4.93mg/g) than that of other treatments (T4-6.63, T3-8.27, T2-10.72, T1-11.2mg/g). Control have maximum reducing sugar (11.67mg/g) respectively (Table-2).

3.1.4. Effects of the EM, *Aspergillus niger*, *Bacillus subtilis* on pH of the compost

The final pH of degraded sample of leaf litter was 6.8, 7.5, 7.2, 7.2, 7.1 and 7.3 for T1, T2, T3, T4 and T5 respectively. In this study, results regarding pH change indicated that pH of decomposed sample was changed from acidic to neutral value. The pH of decomposed sample is increased gradually during composting by microbes (*Bacillus subtilis*, *Aspergillus niger*) due to the degradation soluble organic nitrogen, containing materials to soluble organic nitrogen, the formation of ammonium ion and release of hydroxide ion by hydrolysis (Table-2).

3.2. Effects of the compost on the morphological growth parameters of *Vigna radiata* Linn

The results from the morphological analysis of *Vigna radiata* grown with compost treated with EM, *Bacillus subtilis*, *Aspergillus niger* increased the growth, height of the plant, leaf number, root number and number of flowers. Increase in the number of leaves result to higher rates of photosynthesis hence increased plant growth. These results indicate the significant role of microorganisms in composting and enrichment of plant nutrients.

Biodegradation of leaf litter by microorganisms is dependent on the plant and microbial species. Residues reduction varied markedly with plant species. However, there is a relatively good relation between residues biodegradation and microbial lignocellulytic enzyme activities. In this study, comparatively

higher level of enzymatic activity was observed in the Treatment 5 and 4 (Table-2). Our finding similar to the work in which EM plus xylanase enzyme treated compost has significant role in seed germination, shoot length, leaf length of green gram.^[15]

4. CONCLUSION

There is an urgent need to manage the bulk organic wastes effectively and economically. At the same time, it is also necessary to generate value added products from these wastes. Composting is an alternative technology for a sustainable solid waste management. EM can be used to promote soil fertility and soil quality, reduces or eliminates the use of inorganic inputs, enhances crops yields and quality fertilizer by promoting fermentation and decomposition of waste and organic matter used in agriculture and lowers the hazards of continued cropping in open and greenhouse environments.

5. REFERENCES

1. Chhonkar PK and Dwivedi BS. Organic farming and its implications on India's food security, Fertilizer News, 2004; 38-49.
2. Biddlestone J and Gray KR. Composting In: Comprehensive Biotechnology. Edited by Moo Young. M, Pergammon press, New York. 1985; 1059-1070.
3. Haug RT. Compost: Engineering Principles and Practice, Ann Arbor Science Publishers, Inc., Ann Arbor, MI. 1980.
4. Xu HL. Nature Farming: History, Principles and Perspectives, Journal of Crop Production, 2000a; 3:1-10.
5. Shintani M. EM application manual for APNAN countries, Asia Pacific Natural Agriculture Network, Bangkok, Thailand, 1995; 1st edn: 34.
6. Ahmed John S, Ahamed Jeelani Basha A, Packialakshmi N and Lawa sheela devi A. Application of Effective Microorganisms for the purification of different kinds of water, Asian Jr. of Microbiol. Env.Sc., 2007; 9:337-340.
7. Asia Pacific Natural Agriculture Network. EM application manual for APNAN countries. Edited by Shintani M. Bangkok, Thailand, 1995; 1st edn: 34.
8. Walkley A and Black JA. An estimation of Degjareff method of determining soil organic matter and a proposed modification of chromic acid titration method, Soil Science, 1934; 32:29-39.

9. Jackson ML. Soil chemical analysis, Prentice Hall of India, Newdelhi. 1973,page no..?
10. Updegroff DM. Semimicro determination of cellulose in biological materials, Analytical Biochemistry, 1969; 32:420-444.
11. Miller GL. Use of dinitrosalicylic reagent for determination of reducing sugars, Analytical Biochemistry, 1959; 31:426-428.
12. Parvathi K Venkateswarlu K and Rao AS. Effect of pesticides on development of *Glomus musseae* in groundnut (*Arachis hypogea*), Pans Br. Mycol.Soc., 1985; 42:421-438.
13. Bose S. Statistical analysis for biological data. In:Biostatistics in elementary biophysics, Jytoi Book, Madurai. 1982; 127-128.
14. Reneicke AJ, Viljoean and Sauyam RJ. The suitability of *Eudrilus eugineae*, *Perionyx excavatus* and *Eisenia foetida* for vermicomposting in Southern Africa in terms of their temperature requirements, Journal of Soil Biology and Biochemistry, 1986; 24:1295-1307.
15. Rubini M and Sashi V. Biowaste composting by Effective Microorganisms and crude xylanase and its effect on the growth of *Vigna radiata*, Journal of Ecobiology, 2011; 29:135-140.