STUDIES ON THE MICROBIOGEOCOENOSE OF VERMICOMPOST AND ITS RELEVANCE IN SOIL HEALTH

Synopsis submitted to the University of Madras
in partial fulfilment of the requirements for the Degree of

DOCTOR OF PHILOSOPHY

By

PRISCILLA JEBAKUMARI
P.G. AND RESEARCH DEPARTMENT OF ZOOLOGY
THE NEW COLLEGE
CHENNAI - 600 014
INDIA

April 2004

Earthworm is nature's own litter, aerator, crusher, composter and master builder of the top soil (Talashilkar *et al.,* 2003). Charles Darwin (1881) pioneered the studies on the role of earthworms in soil fertility, followed by several researchers who studied the mechanisms of conversion of organic matter into humus through the action of earthworms and have highlighted their importance in the decomposition of organic wastes (Edwards, 1983; Sharma and Madan, 1983; Edward *et al.,* 1985; Ismail, 1994 a,b 1997; Piearce, *et al.,* 1995; Kale, 1998; Priya and Garg, 2003; Manna *et al.,* 2003). Earthworms are classified based on ecological strategies into epigeics or surface dwellers that are largely used in the composting process; anecics are sub-surface dwellers which create predominantly vertical burrows and are good for both composting and soil improvement. Endogeics predominantly create horizontal borrows and feed on organically rich soil and are called the soil formers. Earthworm feeding and burrowing activities modify soil physical, chemical and biological properties and enhance nutrient cycling by ingestion and by production of casts (Lee, 1985; Lavelle *et al.*, 1989; Blanchart *et al.*, 1990; Lavelle and Martin, 1992; Flegel *et al.*, 2000).

Earthworms are treasure-houses of microorganisms (Ismail, 1997) acting as vectors of beneficial microorganisms promoting growth of microorganisms in their guts. They effectively harness the beneficial soil microflora to convert organic wastes into humus through mutualistic associations, (Parle, 1963a; Atlavinyte and Pociene, 1973; Dash *et al.*, 1979; Lavelle 1989; Joy *et al.*, 1992; Piearce *et al.*, 1995; Sosamma, 1998; Lalitha, 1999; Haynes *et al.*, 1999) which is evident by the presence of microorganisms in their casts and gut (Parle, 1963 a, b; Lee, 1985; Lavelle and Martin 1992; Edwards and Bohlen, 1996; and Nagarathnam *et al.*, 2000). The intestinal mucus in the earthworm gut stimulates microbial activity and its growth thereby increases the decomposition of organic matter. That the passage of soil through the digestive tract is accompanied by qualitative and quantitative changes in the microflora is a well established fact (Parle, 1963; Dash and Senapati, 1980; Piearce and Philips, 1980; Edwards and Bohlen, 1996; Daniel and Karmegam, 1999).

Earthworms enhance nutrient cycling by producing casts which are rich in Ca, Na, Mg, N, P, K and exchangeable NH_4 (Atlavinyte and Vanagas, 1982; Edwards and Bohlen, 1996). Earthworms increase the availability of phosphate from rock phosphate, the mechanism behind the effects of worms are thought to be partly due to the enhanced microbial and phosphatase activity in the casts (Sharpley and Syers, 1976, 1977; Mba, 1977; Tiwari *et al.,* 1989; Palaniappan and Annadurai, 1999). Composting is a self heating, thermophilic and aerobic biological process which occurs naturally in heaps of bio-degradable organic matter such as manure, hay and straw. This dynamic process is brought about by the succession of mixed microbial population (bacteria, fungi, actinomycetes, algae, protozoans) with specific functions, all of which being interrelated in the total process. During the course of composting both quantitative and qualitative changes occur in the active microflora. Certain species of microorganisms multiply rapidly at first, change the environment and then disappear to allow others to succeed (Gaur *et al.,* 1968, 1982; Alexander 1979; Sosamma, 1998). The microorganisms derive their energy and carbon requirements from the decomposition of organic matter. For every 10 parts of carbon one part of nitrogen is required for building up their cell protoplasm (Biddlestone and Gray, 1985). The desirable C : N ratio is 25 to 35 and to arrive at the required C/N ratio, materials such as straw with high C/N ratio and dung or manure with low C/N ratio is mixed during composting (Dalzell *et al.*, 1987; Gaur, 1992).

The rate of progress towards the mature end product is dependent upon several process factors like nutrient content, moisture, aeration and heap acidity.

All organisms require water for life and at a moisture content below 30%, compost heap slows down, the optimum moisture content being between 50 to 60% (Sinha, 1994; Dalzell, *et al.*, 1987; Sosamma, 1998; Lalitha, 1999). Aeration is achieved by turning the pile or by forced draft of air. The energy released during composting is in the form of heat and this causes a rise in temperature. Generally heap passes through stages of warming up, a peak temperature of about 60-70°C, cooling down and maturing. During the thermophase weed seeds and pathogens get destroyed (Thambirajah *et al.*, 1995; Itavaara *et al.,* 2002; Ellorrieta *et al.*, 2002), and complex organic compounds like lignin are degraded (Tuomela *et al.*, 2000).

During composting, as the microorganisms begin their biochemical degradation, the most available substrate such as sugars and starches are attacked (Sinha, 1994) resulting in a drop of pH value. pH usually rises as the next level of substrate, the protein or nitrogen containing organic material is tackled by the microorganisms (Nakasaki *et al.*, 1993; Diaz *et al.*, 2002).

Microorganisms that carry out composting processes are generally those that are naturally indigenous to the soil and the refuse environment (Sinha, 1994; Sosamma, 1998; Lalitha, 1999).

The events of sequential appearance of microorganisms on substrate with respect to time is called succession (Garret 1981). Primary colonizers include weak parasites and primary saprophytic saccharolytic microorganisms. Secondary colonizers include cellulose decomposers and lastly lignin decomposers. During the course of decomposition the water soluble components are metabolised first, followed by cellulose, hemicellulose and lignin. During the course of disappearance of one component and start of the other the spectrum of microflora decomposers gets changed (Bhagyaraj 1988; Subba Rao, 2000).

Microorganisms associated with organic matter decomposition

Bacteria are the most abundant and most diversified group of microorganisms. Since the bacterial activity is not necessarily related to the taxonomy of the groups involved, it is preferable to classify them from a physiological standpoint as autotrophs and chemotrophs/heterotrophs. (Bear, 1964). Bacteria play an important role in organic matter decomposition and mineralisation (Yokoyama *et al.*, 1991; Sakadevan *et al.*, 1993, Subba Rao 2000). They play a prominent role in carbon cycle, through the bacterial respiration and through degradation of organic matter and the carbon is returned back to the atmosphere. Microorganisms especially the nitrogen bacterial forms play a very important role in soil fertility, not only because of their ability to carry out biochemical transformation but also due to their importance as a source and sink for mineral nutrients (Jenkinson and Ladd, 1981; Lovelle and Jarvis, 1996).

Mineralisation of N is a microbial process by which organic forms of nitrogen are converted to inorganic forms like NH_4, NO_2 and NO_3. Mineralisation takes place in three step-by-step reaction - aminisation, ammonification and nitrification. The first two are carried out by heterotrophic microorganisms while the third step is mediated by autotrophic bacteria.

Ammonification converts nitrogen to ammonium ions, while nitrification group of autotrophs - convert ammonium to nitrite and there after to nitrate by the nitrifiers (*Nitrosomonas* and *Nitrobacter*). pH and temperature have a distinct effect on nitrification (Todhi and Ruess, 1988; Prasad and Power 1997; van Heerden *et al.*, 2002).

The presence of ammonium ions and nitrate ions in a compost sample has been suggested as an index of maturity (Chakrabarti *et al.*, 2003). N_2 release begins when the C to N ratio decreases to between 25 and 30 (Blair, 1988) Phosphorus like nitrogen is required for better crop yields. As most Indian soils are deficient in phosphorus, application of phosphatic fertiliser has become essential, but phosphate fertilisers form complexes with soil and become unavailable (Bolan and Hedley, 1989). The phosphate solubilising microorganisms help in mobilizing this locked up phosphorus and make it available to the plants.

Phosphate solubilising microorganisms predominantly comprising of *Bacillus megatherium* var *phosphobacterium* and *Pseudomonas sp* are credited for mineralising organic phosphorus and rock phosphate (Singh and Amberger 1991; Prabhakara *et al.*, 2000; Ravichandran *et al.*, 2003). The enzymatic decomposition of organic phosphorus is due to the presence of phosphomonoesterases both acidic and alkaline (Illmer and Schinner, 1992). Three gram positive and four gram negative phosphate solubilising bacteria were isolated from compost samples (Jugnu *et al.*, 1993).

Organic sulphur is generally associated with proteins in plants and in their residue. Sulphur is therefore as much an integral part of soil organic matter as nitrogen. Many heterotrophic microorganisms are capable of oxidising elemental sulphur and produce thiosulfate as a major product (Germida *et al.,* 1985). The oxidation of sulphur is brought about by *Thiobacilli* which are a group of autotrophic bacteria that derive their energy from the oxidation of sulphur to sulphate and for the fixation of CO_2 into organic matter while *Thiobacillus thiooxidans* produces Sulphuric acid and *Thiobacillus thioparus* produces sulphate (Prasad and Power, 1997).

Fungi are known to be active degraders of organic matter in soils (Alexander, 1979). Fungi are chemoorganotrophic and are dominant agents in the organic matter decomposition. Cellulolytic activity of fungi are not only important in soil cellulolysis, but also in saccharification of agricultural and other wastes (Mandels and Sternberg, 1976). As heterotrophs, the fungi are exceptionally well equipped to undertake the rapid decomposition of virtually all major plant constituents such as celluloses, hemicelluloses, starch and lignin.

Aspergillus sp, Penicillium sp, Fusarium sp, and *Rhizopus sp.* show cellulolytic, lipolytic proteolytic activities, (Rao, 1977; Saddler, 1982; Gomes *et al.,* 1989; Durand *et al.*, 1984; Mishra, *et al.*, 2004).

Mixed cultures of fungi were shown to enhance cellulolytic and pectinolytic activity (Pericin *et al.,* 1982), *Aspergillus niger* and *Aspergillus awamorii* play an important role in phosphate solubilising (Lakshminarasimhan and Seshadri, 2004). *Trichoderma sp* produce mycolytic enzymes like chitinase which attribute to its biocontrol potential (Prabhavathy, *et al.*, 2004)

The constant recurrence of thermophilic fungi on composting plant materials suggest that they are actively involved in the decomposition process. *Mucor pussilus, Humicola sp, and Thermomyces sp* reveal to have strong enzymatic activities towards cellulose and lignin degradation (Cooney and Emerson, 1964; Johri, 2001) Thermophilic microorganisms like *Scytalidium sp* play an important role during compost production. *Chaetomium thermophile* var *coprophile* has been isolated from municipal wastes (Sharma and Lyons, 2001). *Aspergillus fumigatus* a thermotolerant has been recorded from various types of composts (Domsch *et al.*, 1980).

The actinomycetes are chemoorganotrophic, filamentous bacteria. The fact that they are found in large numbers during the later stages of decomposition has suggested that the main role is the breakdown of more resistant components of plant and animal tissues (Bear, 1964; Bagyaraj, 1988). 90% of all actinomycete isolates from soils are *Streptomyces*, in addition the genera encountered often in organic matter during the decomposition are *Thermomonospora sp* and *Thermoactinomyces sp.* Thermotolerant actinomycetes are found in large number during the thermophase stage of composting, the forms isolated include *Streptomyces sp, Nocardia sp, Thermoactinomycetes sp* and *Thermomonospora sp* (Paul and Clark, 1996). Majority of the thermophilic actinomycetes produce xylanase, an enzyme playing a vital role in pulp degradation (Gupta 2001); *Streptomyces sp, Nocardia sp, Streptoverticillium sp and Micromonospora sp,* reported in compost heaps are known to have phosphate solubilising activities (Banik and Dey, 1982; Deshmukh, 2003).

Algae are particularly dependent on high moisture levels, their major role is an outcome of their synthetic rather than their degradative abilities and in barren soils they are the primary invaders. Their main role is the production of nitrogen and oxygen (Watanabe, 1962; Bear, 1964). Cyanobacteria, also referred to as blue green algae, due to the presence of heterocyst and nitrogenase enzyme, act as potential nitrogen fixers even under water logged conditions (Paul and Clark 1996). Blue green algae add lots of organic matter and improve soil aggregation (Bagyaraj 1988; Venkataraman and Tilak 1990). The predominant genera of nitrogen fixing blue green algae are *Anabaena sp, Nostoc sp, Calothrix sp, Tolypothrix sp,* and *Westiellopsis sp*, suitable for lowland rice cultivation (Palaniappan and Annadurai, 1999).

Thus a complete diversity of microorganisms comprising of bacteria, fungi, actinomycetes and algae occur during the different phases of the composting process making the compost a haven of microbial diversity. Vermicompost further supports a distinguished diversity of microorganisms to enhance soil fertility and support soil health.

Objectives of the present research

- To monitor the changes in the physical, chemical and microbiological parameters during the composting process.
- To trace the intrinsic relationship, between temperature, pH & moisture during composting.
- To understand the N, P & S nutrient dynamic during composting.
- To characterise the compost at different stages of the composting process.
- Taxonomic identification of fungi, actinomycete and algae and physiological identification of bacterial forms during the composting process.

The current investigation proceeds in the following lines:

Biodung composting units were set using cattledung slurry, vegetable waste and straw. Representative samples were collected periodically. After six weeks this biodung was introduced into a vermibed which contained both aneceic and epigeic varieties of earthworms (*Lampito mauritii* Kinberg and *Perionyx excavatus* Perrier respectively). Compost samples before introduction into the vermibed and after formation of vermicompost were collected and subjected to the following analysis.

I. Physical Parameters

a. pH

b. Temperature

c. Moisture.

II. Chemical parameters

a. Nitrogen
b. Phosphorus
c. Potassium
d. Carbon
e. Sulphur
f. Calcium
g. Magnesium

Extracellular production of nutrients by microorganisms isolated from compost such as

a. Phosphate
b. Thiosulphate
c. Sulphuric acid
d. Nitrite
e. Nitrate
f. Ammonium
g. Nitrogen

III. Microbial Flora

i) Isolation of heterotrophic bacteria and quantification.

ii) Identification and quantification of bacteria from a physiological stand point.

a. Ammonifiers

b. Nitrifiers

c. Phosphate solubilisers

d. Sulphur bacteria

iii) Isolation, Identification and quantification of fungi and thermophilic fungi

iv) Isolation, identification and quantification of actinomycetes

v) Isolation, identification and quantification of algae

Suitable statistical packages have been applied and the results are discussed based on previous reports.

REFERENCES

Alexander, M. 1979. *Introduction to Soil Microbiology*, 2nd Edition, Wiley Eastern Ltd., New Delhi, pp : 1 - 467.

Atlavinyte, O. and Vanagas, J. 1982. The effects of earthworms on the quality of barley and rye grain. *Pedobiologia.,* **23** : 256 - 262.

Atlavinyte, O. and Pociene, C. 1973. The effect of earthworms and their activity on the amount of algae in soil. *Pedobiologia.,* **13** : 445 - 455.

Bagyaraj, D.J. 1988. Soil Microorgansims, In: *Applied Soil Biology and Ecology* (Veeresh. G.K. and Rajagopal. D. eds.) IInd Edition, published by Mohan Primlani for Oxford Pub. Co. New Delhi, India. pp:1-25.

Banik, S. and Dey, B.K. 1982. Available phosphate content of an alluvial soil as influenced by inoculation of some isolated phosphate solubilising microorganism. *Plant and Soil.,* **69** : 353 - 364.

Bear, F.E. 1964. *Chemistry of the Soil.* Oxford and IBH Publishing Co., 2nd Edition, New Delhi, India. pp : 24 - 34.

Biddlestone, A.J. and Gray, K.R. 1985. Composting, *In : Comprehensive composting*. Vol. 4 (Robinson, C.N. and Howell, J.A. ed) Pergamon Press, Oxford, U.K.).

Blair, J.M. 1988. Nitrogen, Sulfur and Phosphorus dynamics in decomposing deciduous leaf litter in the Southern Appalachians. *Soil Biol. Biochem.* **20(5)** : 693-701.

Blanchart, E., Lavelle, P. and Spain A.V. 1990. Effects of biomass and size of *Millsonia anomala* on particle aggregation in a tropical soil in the presence of *Panicum maximum. Biol. Fertil. Soils,* **10** : 113-120.

Bolan, M.S. and Hedley, M.J. 1989. Dissolution of PO_4 rocks in soils. *Fert. Res.,* **19** : 65-75.

Chakravarti, K., Bhattacharya, P. and Chakrabarti, A. 2003. Assessing compost quality by stability and maturity parameters. *Science and Culture,* **69(8)** : 262 - 265.

Cooney, G.D. and Emerson, R. 1964 *Thermophilic fungi. An account of their biology, activities and classification.* W.H. Freeman and Co; San Francisco, U.S.A pp.128-140.

Dalzell, H.W., Riddlestone, A.J., Gray, K.R. and Thurairajan, K. 1987. *Soil management compost production use in tropical and sub tropical environments*. FAO soils bulletin **56.** pp : 14 - 27.

Daniel, T. and Karmegam, N. 1999. Bioconversion of selected leaf litters using an African epigeic earthworm, *Eudrilus eugeniae. Ecol. Enn. Cons.* **5(3)** : 27-275.

Darwin, C. 1881. *The formation of vegetable mould through the actions of worms with observations on their habits.* John Murray, London, 326 pp.

Dash, M.C., Mishra, P.C. and Behera. N. 1979. Fungal feeding by tropical earthworm. *Trop. Ecol.,* **20** : 9-12.

Dash, M.C. and Senapati, B.K. 1980. Cocoon morphology, hatching and emergence pattern in tropical earthworms. *Pedobiologia,* **20** : 317 - 324.

Deshmukh, A.M. 2003. *Biofertilisers to Biopesticides*, ABD Publishers, Jaipur, India, pp : 1997 - 2001.

Diaz, M.J., Madejon, E., Lopez, F., Lopez, R. and Cabrera, F. 2002. Optimisation of the rate vinasse / grape, marc for co-composting process, *Process Biochemistry* **(37)** : 1143 - 1150.

Domsch, K.H., Cams, W. and Anderson, T.H. 1980. *Compendium of soil fungi*, Vol. 1 : Academic Press, London, pp : 1 - 859.

Durand, H., Soucaille, P. and Tiraby, G. 1984. Comparative study of cellulases and hemicellulases from four fungi. Mesophilic *Trichoderma reesei* and *Penicillium sp,* and thermophiles *Thielavia terrestris* and *Sporotrichum cellulophilum. Enzyme Microb. Technol.,* **6** : 175 - 180.

Edwards, C.A., Burrows, I., Fletcher, K.E. and Jones, B.A. 1985. The use of earthworms for composting farm wastes. *In : Composting of Agricultural and other wastes* (Gasser, J.K.R.ed.) Elsevier. Netherlands, pp : 229 - 242.

Edwards, C.A., Bohlen, P.J. 1996. *Biology and Ecology of Earthworms*, 3rd Edition, Chapman and Hall London, 426 pp.

Edwards, C.A. 1983. Earthworm ecology in cultivated soils. In : *Earthworm Ecology - from Darwin to Vermiculture* (Satchell, J.E. ed.), pp : 123 -127.

Elorrieta, M.A., Suarez - Estrella, F., Eshella, M., Lopez, J., Vargas - Garcia, M.C. and Moreno, J. 2002. *Agricultural Ecosystems and Environments,* **96(1-3)** : pp : 141 - 146.

Flegel, M. and Schrader, S. 2000. Importance of food quality on selected enzyme activities on earthworm casts. *Soil Biol. Biochem.,* **32** : 1191 - 96.

Garret, S.D. 1981. *Soil fungi and soil fertility*, 2nd Edition. Pergamon Press, Oxford.

Gaur, A.C., Sadasivam, K.V., Mathur, R.S. and Magu, S.P. 1982. Role of mesophilic fungi composting. *Agricultural Wastes*, **14** : 453 - 460.

Gaur, A.C., 1992 Bulky Organic manures and crop residues. In : *fertilizers, Organic manures, recyclable wastes and biofertiliser,* (Tandon, H.L.S. ed.) F.D.C.O. New Delhi, pp : 36 - 53.

Gaur, A.C., Mathur, R.S. and Bhardwaj, K.K.R. 1968. Some aspects of influence of humic substances on soil microflora in *Proc. Natl. Acad. Sci., India,* **38** : 25.

Germida, J.J., Lawrence, J.R. and Gupta, V.V.S.R. 1985. *Microbial oxidation of 'S' in Saskatchewan Soils.* In : Proceedings of S-84. (Terry J.W. ed.) pp : 703 - 710.

Gomes, J., Esterbauer, H., Gomes, I. and Steiner, W. 1989. Screening of some wild fungal isolates for cellulolytic activities. *Lett. Appl. Microbiol.,* **8** : 67-70.

Gupta, S., Kapoor, M., Rustagi, R., Kubad, R.C , 2001. Biodiversity & potentiality of thermophilic actinomycetes, *Proceedings of symposium on Thermophiles 2001, Biology and Biotechnolog of thermophilic microbes,* University of Delhi, New Delhi, India. 70 pp.

Haynes, R.J., Fraser, P.M., Tregutha, R.J., Piercy, J.E., Diaz - Cosin., Jesus, J. B., Trigo, D. and Garvin, M.H. 1999. Size and the activity of the microbial biomass and N, S, and P availability in earthworm casts derived from arable and pastoral soil amended with plant residues. 6th Intl - Symposium on Earthworm Ecology, Vigo, Spain. *Pedobiologia,* **43** : 567-573.

Illmer, P. and Schinner, F. 1992. Solubilisation of inorganic phosphates by M.O. isolated from forest soils. *Soil Biol. Biochem.* **24(4)**: 189 - 195.

Ismail, S.A. 1994a. Vermitech : Harnessing the earthworm for the benefit of mankind. *Wake up India,* **17** : 18-20.

Ismail, S.A. 1994b. *Vermiculture in India. Organic Matters,* **17** : 23-28.

Ismail, S.A. 1997. *Vermicology : The Biology of Earthworms.* Orient Longman, India. 92 pp.

Itavaara, M., Karjomar, S., and Selin, J.F. 2002. Biodegradation of polylactide in aerobic and anaerobic thermophilic conditions. *Chemosphere,* **46(6)** : 879-885.

Jenkinson, D.S. and Ladd, J.N. 1981. Microbial biomass in soil measurement and turnover. In : *Soil Biochemistry* (Paul, E.A. and Ladd, J.N. ed.) 5, pp: 415 - 471.

Johri, B.N. 2001. Thermophilic fungi revisited: Do they hold a future. *Proceedings of International Symposium on thermophiles 2001, Biology and Biotechnology of thermophilic microbes,* University of Delhi, New Delhi. India. 8 pp.

Joy Dorcas, E.K., Reddy, M.K. and Reddy, S.M. 1992. Mycoflora associated with the body surface, gut and casting of earthworm (*Lampito mauritii* Kinberg) in relation to soil in Semi-Arid Tropical Grassland. In *Envn. and Ecology,* **10(4)** : 914 - 918.

Jugnu, Varsha, N. and Patel, H.H. 1993 : Inorganic PO_4 solubilisation by certain soil bacteria. *Indian. J. Exp. Biol.* **31**: 743 - 746.

Kale, R.D. 1998. Earthworms : Nature's gift for utilisation of organic wastes. In : *Earthworm ecology* (Edwards, C.A. ed.) CRC Press. The Netherlands, pp: 355 - 376.

Lakshminarasimhan, C. and Seshadri, S. 2004. Possible role of *Aspergillus niger* in improving the phosphate solubilisation in Thanjavur delta area Tamil Nadu. *Proceedings of National symposium on recent trends in biology biodiversity and biotechnology of fungi,* Mycological Society of India. pp 26-27

Lalitha, R. 1999. *Influence of earthworm activity on soil processes, microbial density and crop growth.* Ph.D. Thesis, University of Madras, 168 pp.

Lavelle, P., Barois, I., Martin, A., Zaidi, Z. and Schafer, R. 1989. Management of earthworm populations in agro-ecosystems : A possible way to maintain soil quality? In : *Ecology of arable land* (Clarholm, M. and Bergstrom, L. ed.), Khluwer Academic Publishers. The Netherlands, pp: 109-122.

Lavelle, P. and Martin, A. 1992. Small-scale and large-scale effect of endogenic earthworms on soil organic matter dynamics, soils of the humic topics. *Soil Biol. Biochem.* **24** : 1491 - 1498.

Lee, K.E. 1985. *Earthworms - Their ecology and relationships with soil and land use.* Academic Press, London 411 pp.

Lovelle, R.D. and Jarvis, S.C. 1996. Effect of cattle dung on soil microbial biomass C & N in a permanent pasture soil. *Soil Biol. Biochem,* **28(3)** : 291 - 299.

Mandels, M. and Sternberg, D. 1976. Recent advances in cellulose technology. *Ferment. Technol.,* **54** : 267 - 285.

Manna, M.C., Iha, S., Ghose, P.K. and Acharya, C.L. 2003. Comparative efficacy of three epigeic earthworms under different deciduous forest litter decomposition. *Bioresource technol.,* **88** : 197 - 206.

Mba, C. 1997. Rock phosphate solubilising *Streptosporanguim* isolates from casts of tropical earthworms. *Soil Biol. Biochem.,* **29(3-4)** : 381 - 385.

Mishra, P., Singh, S.K, and Karekar, P.P. 2004. *Proceedings of National symposium on recent trends in biology, biodiversity and biotechnology of fungi,* Mycological Society of India, pp : 56 - 58.

Nagarathnam, B., Karmegan, N. and Thilagavathy, D. 2000. Microbial changes in some organic materials subjected to earthworm action. *J. Ecobiol.,* **12(1)** : 45 - 48.

Nakasaki, K., Yaguchi, H., Sasaki, Y., and Kubota, H., 1993. Effects of pH control on composting of garbage, *waste management and research II (2)* : 117 - 125.

Palaniappan, S.P. and Annadurai, K. 1999. *Organic farming Theory and Practice.* Scientific Publishers, India, pp : 30 - 111.

Parle, J.N. 1963a. Microorganisms in the intestines of earthworms. *J. Gen. Microbiol.,* **31** : 1-11.

Parle, J.N. 1963b. A microbial study of earthworms casts. *J. Gen. Microbiol.,* **31** : 13-22.

Paul, E.A. and Clark, F.E. 1996. *Soil microbiology and biochemistry*, II edition. Academic Press Inc., California, U.S.A. pp : 289 - 313.

Pericin, D., Jarak, M., Antov, M., Vujicic, B. and Keveresan, S. 1992. Effect of inorganic phosphate on the secretion of pectinolytic enzymes by *Aspergillus niger. Lett. Appl. Microbiol.,* **14** : 275-278.

Piearce, T.G. and Philips, M.J. 1980. The fate of ciliates in the earthworm gut. An invitro study. *Microb. Ecol.,* **5** : 313 - 319.

Piearce, T.G., Moody, S.A., Briones, M.J.I. and Dighton, J. 1995. Earthworm *Lubricus terrestris, L., Allolobophora chlorotica* - Used in straw composting. *Soil Biol. Biochem.,* **27(9)** : 1209-1213.

Prabhakara, K.S., Gomathisivam, P., Leemarose. H.P., Leemarose, Y.M., Sudha, S., Josephine, T. and Manjunath, K. 2000. Solubilisation of rock PO_4 by an endorhizospheric bacterium *Pseudomonas sp. J. Ecotoxicol Environ. Moni,* **10(2)** : 137-140.

Prabhavathy, V.R., Mathivanan, N., Murugesan, K. and Lalithakumari, D. 2004. Strain improvement in *Trichoderma* by protoplast fusion for multienzymes production and enhanced biocontrol. *Proceedings of National Symposium on recent trends in biology, biodiversity and biotechnology of fungi* : Mycological Society of India. 45 pp.

Prasad, R., and Power, J.F. 1997. *Soil fertility for sustainable agriculture.* Lewis Publishers, New York, U.S.A. pp : 115 - 127.

Priya, K. and Garg, V.K. 2003. Vermicomposting of mixed solid textile mill sludge and cow dung with the epigeic earthworm *Eisenia fetida. Bioresource Technol.,* **90** : 311-316.

Rao, R.R. 1977. Studies on cellulase production by an *Aspergillus strain. Indian. J. Microbiol.,* **17** : 123-128.

Ravichandran, V.K., Devarani, N. and Balasubramanian, T.N. 2003. Dissolution of low grade rock PO_4 in the soil amended with organic manure and phosphobacteira. *J. Ecobiology,* **15(1)** : 55-59.

Saddler, J.N. 1982. Screening of highly cellulotyic fungi and the action of their cellulose enzyme systems. *Enzyme Microb. Technol.,* **4** : 414-418.

Sakadevan, K., Mackay, A.D. and Hedley, M.J. 1993. Influence of sheep excreta on pasture uptake and leaching losses of sulphur, nitrogen and potassium from grazed pastures. *Aust. J. Soil. Res.,* **31** : 151-162.

Sharma, N. and Madan, M. 1983. Recycling of organic wastes through Vermicomposting *Bioenergy Newsletter,* **2** : 30-31.

Sharma, S. and Lyons, G. 2001. Role of thermophilic microorganisms in the production of mushroom compost. *Proceedings of Symposium on thermophiles, biology and biotechnology of thermophilic microbes.* University of Delhi, New Delhi. India. 18 pp.

Sharpely, A.N. and Syers, J.K. 1976. Potential role of earthworm casts for the 'p' enrichment of run off waters. *Soil Biol. Biochem.,* **8** : 341-346.

Sharpley, A.N. and Syers, J.K. 1977. Seasonal variation in casting activity and in the amounts and release to solution of 'p' forms in earthworm casts. *Soil Biol. Biochem.,* **9** : 227-231.

Singh, C.P. and Amberger, A. 1991. Solubilisation and availability of phosphate during decomposition of rock PO_4 enriched straw and urine. *Biological Agriculture Horticulture*, **7** : 261 - 269.

Sinha, R.K. 1994. *Environmental science - disinfection, desalination, composting*. Common Wealth Publishers, New Delhi. India pp : 138-147.

Sosamma, K.O. 1998. *Studies on bio-processing of organic solid wastes applying vermitech and efficacy of vermicompost and vermiwash in soil fertility*. Ph.D. Thesis, M.G. University, Kerala, India. 110 pp.

Subha Rao, N.S. 2000. *Soil microbiology and plant growth*. Oxford and IBU Publishing Co. Pvt. Ltd., New Delhi, pp : 252 - 280.

Talashilkar, S.C., Kadam, R.G., Todkari, A.A., Inamdar, P.V. and Dhopavkar, R.V. 2003. Vermiculture Biotechnology. In : *Vermisource Technology* (Tripathi. G. ed.,) Discovery Publishing House, New Delhi. India, pp : 81-156.

Thambirajah, J.J., Zulkali, M.D. and Hashim, M.A. 1995. Microbiological and biochemical changes during the composting of oil palm empty-fruit-bunches. Effect of nitrogen supplementation on the substrate. *Bioresource Technol* : **52(2)** pp : 133-144.

Tiwari, S.C., Tiwari, B.K. and Mishra, R.R. 1989. Microbial populations, enzyme activities and nitrogen - phosphorus - potassium enrichments in earthworm casts and in the surrounding soil of a pineapple plantation. *Biol. Fertili. Soils.,* **8** : 178-182.

Todhi, M.A.K. and Ruess, R.W. 1988. Variation in soil nitrifiers and leaf nitrate reductase activity of selected tree species in a forest community. *Soil Biol. Biochem.*, **20 (6)** : 939 - 943.

Tuomela, M., Vikman, M., Halakka, A. and Itavaara, M. 2000. Biodegradation of lignin in a compost environment. *Bioresource Tech., 72(2):* 169-183.

van Heerden, I., Cronje, C., Swart, S.H. and Kotze, J.M. 2002. Microbial, chemical and physical aspects of citrus waste composting. *Bioresource technology*, **81(1)** : 71-76.

Venkataraman, G.S. and Tilak, K.V.B.R. 1990. *Soil fertility and fertiliser use.* IV.IFFCO, New Delhi, India. pp : 138-148.

Watanabe, A. and Yamamoto, Y. 1971. Algal nitrogen fixation in the tropics, *Pl. Soil.* Special Volume : 403 - 413.

Watanabe, A.J. 1962. Ecology of microorganism in the soil. *Gen. Appl. Microbiol.,* **8** : 85.

Yokoyama, K., Kai, H., Koga, T. and Aibe 1991. Nitrogen mineralisation and microbial population in cow dung, dung balls and underlying soil affected by paracoprid dung beetles. *Soil Biol. Biochem.*, **23** : 649 - 953.