



Full Length Research Article

EFFECTIVE MICROORGANISMS – AN INCREDIBLE ALTERNATIVE FOR WASTEWATER TREATMENT

***Victoria, J. and Uma Maheswari, N.**

Department of Microbiology, Sengamala Thayaar Educational Trust Women's College,
Sundarakkottai, Mannargudi-614001, Tamil Nadu, India

ARTICLE INFO

Article History:

Received 21st June, 2016
Received in revised form
16th July, 2016
Accepted 19th August, 2016
Published online 30th September, 2016

Key Words:

Waste Water,
EM-Solution,
Agar media,
Nitrate, Phosphate.

ABSTRACT

A study on the determination of the use of Effective Microorganisms (EM) was carried out. EM was purchased from the office of 'Ecopro' Auroville, Auroshilpam, Tamilnadu, India. The waste water was collected from STET Women's College, Sundarakkottai, Mannargudi, Thiruvarur Dt. The parameters that indicate the waste water treatment process such as odour, pH, DO, BOD, COD, TDS, TS, TSS, Nitrate and Phosphate were determined before and after the treatment of wastewater, to observe the efficiency of selected process. There was an appreciable reduction in the above mentioning values. All the parameters showed an elevated level in the raw sewage but after treatment there is a steady reduction after 5, 10, 15 and 20 days of incubation. No reduction was observed in the level of DO. All the parameters were reduced to tolerable environmental standard. Thus, this study provides valuable information regarding the application of microorganisms for the successful treatment of waste water.

Copyright©2016, Victoria and Uma Maheswari. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

INTRODUCTION

A major problem facing municipalities throughout the world is the treatment, disposal or recycling of sewage sludge. Generally sludge from municipal waste consists mainly of biodegradable organic materials with a significant amount of inorganic matter (Elliott, 1986). However, sludge exhibits wide variations in the physical, chemical and biological properties (Bruce, 1990, and Colin *et al.*, 1988). At the present time, there are a number of methods being used to dispose of sewage sludge from disposal of landfill to land application. Although there are many methods used, there are numerous concerns raised regarding the presence of constituents including heavy metals, pathogens and other toxic substances. This requires the selection of the correct disposal method focusing on efficient and environmentally safe disposal. New technologies are being produced to assist in the treatment and disposal of sewage sludge, conforming to strict environmental regulations. One of the new technologies being proposed is the use of Effective Microorganisms (EM) for the treatment of waste water.

***Corresponding author: Victoria, J.**

Department of Microbiology, Sengamala Thayaar Educational Trust
Women's College, Sundarakkottai, Mannargudi-614001, Tamil Nadu,
India

Effective Microorganisms (EM)

The technology of Effective Microorganism (EM) was developed during the 1970's at the University of Ryukyus, Okinawa, Japan (Sangakkara *et al.*, 2002). Studies have suggested that EM may have a number of applications, including agriculture livestock gardening and land-scaping, composting, bioremediation, clearing septic tanks, algal control and household uses (EM Technology, 1998). The practical application was developed by Professor Teuro Higa. He has devoted much of his scientific career to isolate and select different microbes for beneficial effects on soils and plants. He has found microbes that can coexist in mixed cultures and are physiologically compatible with one another. When these cultures are introduced beneficial effects are greatly magnified in a synergistic fashion (Colin *et al.*, 1988). A microbial inoculant containing many kinds of naturally occurring beneficial microbes called 'Effective Microorganisms' has been used widely in nature and organic farming (Diver, 2001). Effective Microorganisms technology respects nature and does not contain genetically engineered microbes. These microorganisms are collected from the natural environment and therefore are indigenous. Effective Microorganisms is a mixture of groups of organisms that has a receiving action on humans, animals and the natural

environment and has also been described as a multi-culture of coexisting anaerobic and aerobic beneficial microorganisms (EmTechnology. 1998). EM contains selected species of microorganisms such as lactic acid bacteria; *Lactobacillus casei*, Photosynthetic bacteria, *Rhodospseudomonas palustris*, Yeasts: *Saccharomyces cerevisiae*, *Candida utilis*, Actinomycetes: *Streptomyces albus*. All of these are mutually compatible with one another and coexist in liquid culture (Higa, 1995). The principle of EM is the conversion of a degraded ecosystem full of harmful microbes to a one that is productive and contains useful microorganisms. It is either used in activated form or by preparation of bokashi for the treatment of waste water. Many countries have adapted this method for the waste management (Outi Kaarela Bio 40006 2006). The application of EM will improve the quality of soil and irrigation water system. It can be used in seed treatment. It can be used to make organic sprays for the enhancement of photosynthesis and control of insects, pests and diseases.

The formulation method increased their persistence and dependability on the prevailing environmental condition and offered protection against unfavorable environmental condition. Moreover, EM can show better performance if they are mixed with suitable ingredients which may act as nutrients, adhesives or wet table agents (Karthick Raja *et al.*, 2014). The use of Effective Microorganisms (EM) for reducing volumes of sewage sludge has often been suggested as feasible in either wastewater treatment plants or onsite wastewater treatment systems such as septic tank and industrial effluents. In the present study, Effective microorganism was evaluated for the sewage water treatment efficacy under laboratory condition. EM has a sweet-sour taste and smell (pH below 3.5). It needs to be stored in an airtight plastic container. Gas needs to be released occasionally. For smaller quantities this can be done by opening the container. The best temperature for storage is between 15 and 20°C with little fluctuation (less than 10°C in 24 hours).

Waste Water

In India, the abundance of soils with low organic matter content, favours the use of industrial waste waters containing organic matter as an organic amendment and nutrient supply to soil. Although the benefits of wastewater use in irrigation are numerous but precautions should be taken to avoid short and long term environmental risks related. Earlier studies have shown that the effect of an industrial effluent vary from crop to crop. Waste water consists of approximately 99.9% water 0.02 to 0.08% suspended solids and other soluble organic and inorganic substances. In general the waste water is weak in nature that is BOD is normally not high. It is rich in nutrients like N and P. The advantage of municipal effluent for irrigational purpose namely supplement of nutrients, reducing the dependence of underground water as well as some mitigation of environmental pollution (Billore Reddy *et al.*, 1998), but Khatrt *et al.* (2003) observed some heavy metals in the sewage water which may accumulate to toxic levels in soil and plants tissues and may also inhibit a concomitant reduction in crop yield. When the waste water is released into the environment without proper treatment, it alters the characters of ecosystem. While irrigated, the growth and

productivity of the crop are also effected (Ayyasamy *et al.*, 2002).

Microorganisms in waste water

The waste water comprises of all types of microorganisms like bacteria and fungi. Bacteria are predominant when compared to fungal population. The widely distributed bacterial populations are *E.coli*, *Proteus sp*, *Bacillus sp*, *Pseudomonas sp*, *Acinetobacter sp*, *Stenotrophomonas sp*, *Rhodococcus sp.*, *Microbacterium sp*. which may be either aerobic or anaerobic in nature. Those organisms degrade the organic matters present in the wastewater and have always the BOD value of the waste water very low compared to drinking water (Yang *et al.*, 2008).

Uses of Recycled Waste Water

Water conservation measures, in particular, the use of recycled water, have forced landscapers to rethink ways to maintain landscapes and gardens. Recycled water is water that has been previously used, suffered a loss in quality and then treated to a point where it is of suitable quality for additional beneficial uses. With the increasing use of recycled water throughout the state, more knowledge is necessary to determine which plants, if any, can be grown and how irrigation must be managed. The use of recycled water landscape irrigation can, however, pose problems. Water always contains measurable quantities of dissolved substances collectively called salts. The suitability of water for irrigation purposes will depend on both the amount and the kinds of salt present in the waste water that may be quite variable from source to source and from time to time. Typically, as compared to potable water, recycled water in general will have higher total salt levels and specific ions may be present at higher concentrations especially for sodium, chloride, magnesium and sometimes calcium (Asano, 1981 and 1984). There is a potential of incurring damage to both plants and the soil structure when using recycled water in the landscape. Many landscapes and ornamental plants are grown in greenhouses or other controlled environmental conditions, additional considerations must be made. One of the first places in the country to use recycled water was San Francisco's Golden Gate Park. As early as 1912, recycled water was combined with ground water and used for irrigation, filling lakes and spillways. The effect of specific element and chemical compounds in recycled water needs to be identified in order to be able to manage recycled water irrigation for ornamental plants effectively. Most research concerning high concentrations of salinity effects by irrigation water on turf grass and crop plants has emphasized N^+ concentrations (Grieve and Maas, 1998).

MATERIALS AND METHODS

Effective Microorganisms (EM)

Effective microorganisms (EM) used in this study was purchased from the office of 'Ecopro' Auroville, Auroshilpam, Tamilnadu, India as EM stock liquid culture containing a mixture of lactic acid bacteria, *Lactobacillus casei* (10^5), Photosynthetic bacteria, *Rhodospseudomonas palustris* (10^1) and Yeast, *Saccharomyces cerevisiae*. EM

solution is a yellowish liquid with a pleasant odour and sweet sour taste with a pH of 3 and stored in cool place without refrigeration.

Extension (EMe) and Activation (EMa) of EM Stock Solution (1)

For most applications EM stock solution is to be "extended or activated" prior to use. One litre of the EM stock solution and 1kg of jaggery were mixed with 20 liters of water. The water has to be clean and free from chlorine. The container should be of good- grade plastics. 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 (abbreviated as EMe or 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.

Collection of waste water sample

The waste water was collected in a sterilized plastic container from STET Women's College, Sundarakkottai, Mannargudi (Tk), Thiruvarur (Dt). Immediately after collection, the waste water was brought to the laboratory for further analysis. The collected waste water sample was subjected to physico-chemical and microbiological analysis.

Analysis of Physico-chemical Parameters of Waste Water Samples

The laboratory experiment was conducted to evaluate the effect of EM on waste water treatment with three replicates and untreated control. The physico-chemical properties such as pH, Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), Total Dissolved Solids (TDS), Dissolved Oxygen (DO), Total Solids (TS), Total Suspended Solids (TSS), Nitrate (N) and Phosphate (P) of the waste water samples were analysed before the addition of EM solution (2).

Isolation and identification of Microbial Populations

The collected waste water samples were subjected to serial dilution agar plating technique (Aneja, 2001), and the microbial populations on the nutrient agar and sabourad agar plates were counted using the digital colony counter. The bacterial and actinomycetes species were isolated and identified by Gram's staining and Biochemical tests and the fungal populations by lactophenol cotton blue staining.

Treatment of Waste Water samples

The set up consists of three 1liter Erlenmeyer flasks with 1 litre of waste water each. 100 ml of activated EM culture was added into the waste water sample. The setup was operated continuously for 20 days. EM was added each day at the dilution rate of 1:10,000 for five days. The effect of EM was assessed by changes in the odour, pH, Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), Total Dissolved Solids (TDS), Dissolved Oxygen (DO), Total Solids (TS), Total Suspended Solids (TSS), Nitrate (N) and Phosphate (P) after the incubation period.

RESULTS AND DISCUSSIONS

The total microbial populations mainly bacteria and fungi were observed before treatment. Bacterial population and fungal populations of 14.2×10^4 and 12.0×10^3 CFU/ml respectively were observed. No actinomycetes and Yeast were observed before treatment. Bacterial and fungal populations were increased gradually from 14.2×10^4 to 23.1×10^5 , 4.0×10^6 and 41.2×10^7 CFU/ml and 15.0×10^3 , 34.1×10^3 , 01.2×10^4 and 54×10^4 respectively at the time intervals of 5, 10, and 15 and 20 days. Actinomycetes and yeasts were observed only after the addition of solution into the wastewater. Actinomycetes and yeast populations of 3.4×10^2 , 34.1×10^2 , 45.3×10^2 and 51.1×10^2 CFU/ml and 41.1×10^3 , 54.1×10^3 , 6×10^4 and 27.1×10^5 CFU/ml were observed at 5, 10, 15 and 20 days of incubation with EM solution (Table 1).

Table 1. Microbial population (cfu/ml) of untreated and em treated wastewater

| S.NO | Microorganisms | Before Treatment | After Treatment | | | |
|------|----------------|-------------------------------|------------------------------|--------------------|--------------------|--------------------|
| | | Population of microbes CFU/ml | Incubation time(Days)/CFU/ml | | | |
| | | | 5 | 10 | 15 | 20 |
| 1 | Bacteria | 14.2×10^4 | 14.2×10^4 | 23.1×10^5 | 4.0×10^6 | 41.2×10^7 |
| 2 | Fungi | 12.0×10^3 | 15.0×10^3 | 34.1×10^3 | 01.2×10^4 | 54.1×10^4 |
| 3 | Actinomycetes | - | 3.4×10^2 | 34.1×10^2 | 45.3×10^2 | 51.1×10^2 |
| 4 | Yeast | - | 41.1×10^3 | 54.1×10^3 | 6.0×10^4 | 27.1×10^5 |

Table 2. Physico – Chemical Parameters of em Treated Waste Water

| S.No | Parameter | Untreated Waste Water | Treated Waste water | | | |
|------|----------------------------------|-----------------------|------------------------|------|------|------|
| | | | Incubation time (Days) | | | |
| | | | 5 | 10 | 15 | 20 |
| 1. | PH | 7.0 | 7.0 | 5.4 | 4.7 | 3.5 |
| 2. | Dissolved oxygen (mg/l) | 10.4 | 10.4 | 14.7 | 18.5 | 20.0 |
| 3. | Biochemical oxygen Demand (mg/l) | 28.4 | 28.4 | 18.4 | 15.0 | 11.5 |
| 4. | Chemical oxygen Demand (mg/l) | 54.4 | 54.4 | 50.4 | 42.5 | 32.4 |
| 5. | Total solids (mg/l) | 94 | 94 | 88.4 | 80.4 | 71.4 |
| 6. | Total dissolved solids (mg/l) | 60 | 60 | 58 | 52 | 48 |
| 7. | Total suspended solids (mg/l) | 34 | 34 | 30.4 | 28.4 | 23.4 |
| 8. | Nitrate (mg/l) | 6.7 | 6.7 | 5.9 | 2.8 | 1.6 |
| 9. | Phosphate (mg/l) | 3.2 | 3.2 | 2.7 | 2.2 | 1.0 |

New technologies are being produced to assist in the treatment and disposal of sewage sludge, conforming to strict environmental regulations. One of these new technologies being proposed is the use of Effective Microorganisms (EM). The technology of Effective Microorganisms (EM) was developed during the 1970's at the University of Ryukyus, Okinawa, Japan (Outi Kaarela Bio 40006 .2006). The isolation of gram negative organisms i.e., *E.coli*, *Pseudomonas sps*, *Bacillus sps*, from waste water collected from kitchen. The gram negative was identified as *E.coli* and *Pseudomonas sps* accounted for the majority (90%) of the strains (Khatrt *et al.*, 2003). In the present study, the microbial populations isolated from the wastewater before treatment were *Bacillus species*, the gram positive organism and gram negative organisms identified were *E.coli* and *Pseudomonas sp*. After treatment with EMa, different microorganisms such as bacteria, fungi, actinomycetes and yeast were isolated from the wastewater. The bacterial species such as *E.coli*, *Pseudomonas sp*, *Bacillus sp*, *Lactobacillus casei* and fungal species such as *Aspergillus niger*, *Aspergillus flavus*, *Rhizopus* and actinomycetes such as *Streptomyces albus* and yeast *Saccharomyces cerevisiae* were isolated and identified. The wastewater generated by the kitchen is heavily polluted with very high quantity of suspended solids high quantity of organic and inorganic materials (High values of BOD and COD) (Hussain *et al.*, 1999). In the present work, the polluted kitchen wastewater contains suspended solids, organic and inorganic materials which were removed by waste water treatment process using EM solution.

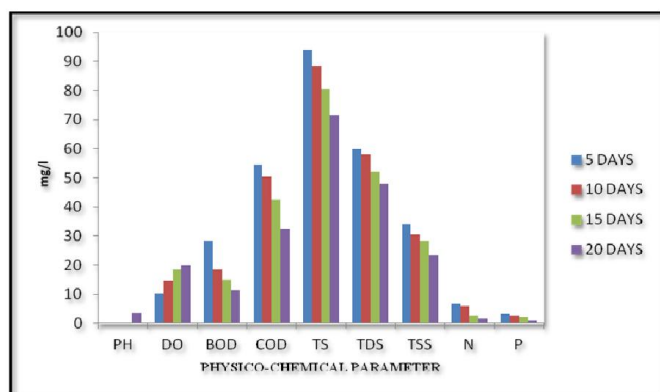


Fig. 1. Physico – chemical parameters of em treated waste water

The effective microorganism of wastewater treatment method is needed to determine the amount of oxygen demanding wastes in water. The amount of oxygen consumed by microorganisms in decomposing organic waste is proportional to the amount of waste water present. BOD indicates the level of organic matter pollution, the greater value; the greater the problem of decomposition; wastewater has BOD value of 28.4 mg/l (Blair *et al.*, 1978). EM treated domestic sewage showed distinct reduction in all the tested parameters under all the tested incubation period. Total dissolved solid was found to be reduced from 2160 mg/lit to 1012, 940 and 901 mg/lit. pH was also reduced from 9.0 to 8.4, 7.4 and 7.1 alkalinity was reduced from 59 mg/lit to 41, 37 and 21 mg/lit. The BOD was reduced from 2.8 to 2.1, 1.5 and 0.9. No reduction was observed in DO content. The COD was decreased from 164 to 141, 112 and 112 and 109 mg/lit at the respective incubation

time (Karthick Raja *et al.*, 2014). In the present study, the BOD in raw wastewater was 28.4 mg/l. After EM treatment, 5 to 20 days the level of BOD was decreased, from 28.0, 18.4, 15.0 and 11.5 mg/l (Table 2). COD gives a good indication of the amount of oxygen needed to stabilize the wastewater. It oxidizes ethanoic acid and polycarbohydrates that are not stabilized by bacteria. COD removals have been used as marks of efficiency or levels of waste stabilization in reactors. COD removal of 41 ton chemical oxygen demand/day in two treatment plants in Argentina and 100.8 ton COD/ day in one treatment plant in Kenya (Faxg and Lia 2001-15). In the present study, the COD of untreated waste water was 54.4mg/l. After EM treatment, the level of COD was reduced from 54.4 to 50.4, 42.5 and 32.4 mg/l. The kitchen wastewater is heavily polluted. The appropriate wastewater treatment facilities which will not only bring down the level of pollution but at the same time make the treated wastewater recyclable such as for agricultural purpose. Analysis of all the generated data of untreated and treated wastewater samples observed that pH, BOD, COD, TS, TDS, TSS, Nitrate and Phosphate contents of treated water were reduced to tolerable environmental standard and the DO level of the treated waste water was increased. The results obtained for all the parameters were better at the 20th day of incubation. Based on result obtained from liquid treatment it will be interpreted as one of the easy method which can be applied locally to convert the waste into byproduct which can help to reduce the environmental pollution. Thus, the present study revealed the necessity of treating wastewater in a cost effective and easiest method to reuse them in a proper utilizable way. Conventional treatment methods are more difficult and not economical for routine use. The usage of EM for domestic sewage treatment can be made routine on daily basis in the origin itself.

Conclusion

Economic development through industrialization, agriculture for the better future of our society results in environmental deterioration. The present study was undertaken to determine the use of one of such new technique i.e., Effective Microorganisms for the treatment of wastewater. The indicator like pH, DO, BOD, COD, TDS, TS, TSS, Nitrate and Phosphate were estimated before and after the treatment of wastewater, to observe the efficiency of selected process. There was appreciable reduction in the above mentioning values which has been observed by other liquid waste management.

Acknowledgement

I thank Dr.V.Dhivaharan, Dean of Lifesciences, Sengamala Thayaar Educational Trust Women's College, Sundarakkottai-614016, Mannargudi, Thiruvavur -Dt for providing me the facilities and greater support during the tenure of this study.

REFERENCE

- Aneja, K.R. 2001. Experiments in Microbiology, Plant pathology, Tissue culture and Mushroom Production Technology. 3rd Edition-New age International (p) Ltd., Publishers. New Delhi.

- APHA. 1989. Standard methods for the examination of water and wastewater, 20th edition, Eds Eaton A.D., Cleseri L.S. and Greenberg A.E. American Public Health Association. Washington. DC. *Journal of Experimental Sciences*, 2(7):30-32.
- Asano T, Smith R.G. and Tchobanglous G., 1984. In: Pottygrove, G, Asano, T. 1984 Irrigation with reclaimed municipal wastewater a guidance. Manual report. NO-1 wr.calif.
- Asano, T. 1981. Evaluation of agricultural irrigation projects using reclaimed water. Agreement 8:179-215. Office of water recycling, California State Resource Control Board, Sacramento CA
- Asia-Pacific Natural Agriculture Network. 1995. EM Application manual for APNAN countries, Shintani, M. (ed) 1st Asia-Pacific Natural Agriculture Network, Bangkok, Thailand, 34.
- Ayyasamy, P.M., Banuregha, R., Vivekanandhan, G., Savitamani, K. and Lakshamanaperumalsamy, P. 2002. Treatment of Sago factory effluent by aerobic microbial consortium. *Indian J. Environ. Protect.* 22:554-558.
- Billore, S.K. and Reddy, K.B. 1998. Feasibility of nitrogen removal from municipal waste water by application to land coverer with a fodder grass. In: Environmental Issue and Researches in India. (eds.). K. Agarwals & B. K. Garg. Himanshu Public., Udaipur. 293-304.
- Blair, Jr J.C., Moeller, T., Kleinberg, J., Guess, C.O., Catellion, M.E. and Metz, C. 1978. Chemistry. Academic Press. New York, San Francisco, London. 348-382.
- Bruce A.M. 1990. Sewage Sludge Processing: Progress and Problems. Sewage Treatment and Use of sewage Sludge and Liquid Agricultural Wastes, ed P.L'Hermitte, Elsevier Applied Science. Published by Lanfax Laboratories Armidale. ISBN 0-9579438-1-4 :347-354.
- Colin F., Leschber R., and Mininni G. 1988. Physical and chemical characterization of sewage sludge, in Sewage Sludge Treatment and Use: New Developments, Technological Aspects and Environmental Effects, eds AH Dirzwager, PL' Hermitte), Elsevier Applied Science.
- Crawford, J. H. 2002. Review of composting. *Process. Biochemistry.* 18:14-15.
- Diver S. 2001. Nature Farming and Effective Microorganisms'. Retrieved from Rhizosphere Publications, Resource Lists and Web Links from Steve Diver, <http://ncatark.uark.edu/steved/Nature-Form-EM.html>.
- Elliott, H.A. 1986. Land Application of Municipal sewage', *Journal of Soil and Water Conservation*, 41 : 5-10.
- EM Technology. 1998. (updated 10 May 1999, accessed 31, Oct 2001), 'Effective Microorganisms for a Sustainable agriculture and Environment', EM Tech Product 1, <http://emtech.org/prodol.htm>.
- EM Trading. 2000. Effective Microorganisms (EM) from sustainable community development. Effective Microorganisms@emtrading.com. <http://www.emtrading.com.html>.
- Grieve C.M., and Maas E.V. 1998. Differential effects of sodium and calcium ratio on *Sorghum* genotypes *Crop. Science* 28:639-665.
- Higa, 1995. Effective microorganisms : their role in Kyusei Nature Farming and Sustainable Agriculture. In: Parr J.F., Hormick S.B. and Simison M.E., (ed). Proceeding of the Third International Conference on Kyusei Nature Farming, U.S. Department of Agriculture, Washington, D. C., U. S. A.
- Hussain, T., Javaid, T., Puaar, J.R., Jilani, G. and Haq, M.A. 1999. Rice and wheat production in Pakistan with effective microorganisms. *Americ. J. Agricult.* 14 : 30-36.
- Jainmin Hua Pinglin A.N., Josef, Winter, Claudi and Gallert. 2003. Elimination of COD, Microorganisms and Pharmaceuticals from sewage by trickling through sandy soil below leaking sewers. *Wat Res.*, 37 .18 : 4395-4404.
- Javaid A., Bajwa R. and Anjum T. 2008. Effect of heat sterilization and EM (Effective Microorganisms) application of wheat (*Triticum aestivum* L.) grown in organic matter amended soils. *Cereals Res. Commun.* 36:489-499.
- Karthick Raja Namasivayam, S., Shunmugaraj, M., Arvind Bharani R.S. and Francis A.L. 2014. Evaluation of phytotoxicity of Effective Microorganisms treated distillery industry effluent. *Biosci. Biotech. Res. Asia.* 11 (2): 587-592.
- Khattr, S., Dhankhar, R. and Dhayia, J.S. 2003. Impact of sewage wastewater on seed germination and seedling growth of some Rabi and Kharif crops of Haryana. *Indian J. Environ. Protect.* 23:1161-1165.
- Kriss, A.E. 1971. Oceanic microbiology: Ecology and geography of microorganisms. *Mikrobiologia.* 40:904-911.
- Outi Kaarela Bio 40006. 2006. Water Chemistry, Laboratory Exercise, Tampere University of Technology Environmental Engineering Biotechnology. *Journal of Pure and Applied Microbiology.* 6 (1): 333-338.
- Sangakkara U.R. 2002. The Technology of Effective Microorganisms : case studies of application. Cirencester, UK : *Royal Agricultural College. Exp. Sci.* 2(7) :30-32.
- Yang J.S., Liu W.J., Wu J.L. and Yuan H.L. 2008. Application of biological aerated filter in treating biomass wastewater and its microbial population characteristics. *Huan Jing Ka Xue.* 29 (1): 3133-3137.
