



ISSN: 0975-833X

RESEARCH ARTICLE

EFFECTIVE MICROORGANISMS (EM) IN WASTE WATER TREATMENT AND ITS EFFECT ON THE GROWTH PARAMETERS OF *VIGNA RADIATA*

*Victoria, J. and Uma Maheswari, N.

Department of Microbiology, STET Women's College, Sundarakkottai-614016, Mannargudi, Thiruvarur Dt, Tamil Nadu, India

ARTICLE INFO

Article History:

Received 12th October, 2016
Received in revised form
28th November, 2016
Accepted 16th December, 2016
Published online 31st January, 2017

Key words:

Commercial biofertilizer,
Effective Microorganisms (EM),
Vigna radiata.

ABSTRACT

Effective microorganisms (EM) is a commercial biofertilizer that contains a mixture of co-existing beneficial microorganisms collected from natural environments. Predominantly it consists of species of photosynthetic and lactic acid bacteria, yeast, and actinomycetes. The present study was undertaken to determine the efficiency of Effective Microorganisms (EM). EM was obtained from the office of 'Ecopro' Auroville, Auroshilpam, Tamilnadu, India. The waste water was collected from the women's hostel of STET women's College, Sundarakkottai, Mannargudi, Sundarakkottai. 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. Soil samples were collected from Thiruthuraiipoondi, Thiruvarur District, Tamilnadu, India which consists of Nedumbalam (Ndb) soil series, one of the soil series of Thiruvarur District. The experimental set up was designed randomly having EM alone (T1), Treated waste water alone (T2), EM plus Treated waste water (T3) and control (C). 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. EM treated waste water was utilized for the cultivation of *Vigna radiata*. Among the treatments studied, T3 showed the highest results when compared to other treatments proving the efficiency of EM in recycling of waste water. Key words: *Vigna radiata*, Effective microorganisms (EM), Secondary crop, Treatments, Sewage, Incubation.

Copyright©2017, 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.

Citation: Victoria, J. and Uma Maheswari, N. 2017. "Effective Microorganisms (EM) in waste water treatment and its effect on the growth parameters of *Vigna radiata*", *International Journal of Current Research*, 9, (01), 45302-45309.

INTRODUCTION

The practice of using large amounts of chemical fertilizers and pesticides has been followed by the farmers to increase crop yield. Nowadays, they became aware of the drastic effects of loading the soil with chemicals. They heavily affect the production and completely degrade the environment (Nishio, 1996). In 1989, the National Research Council of National Academy of Sciences negotiated about the future of the chemical-based agrosystem and issued a report on their consequences. Sustainable agriculture aims to create systems which are environment friendly, energy saving, highly productive and economically profitable, conserving natural resources and promising the safety and quality of food (Parr *et al.*, 1994). Poincelet (1986) also reported the same about the usage of chemical fertilizers and pesticides to conserve the natural resources. Incorporating composts to the soil improve

the fertility and also pave the way to manage the organic wastes (Piqueres *et al.*, 2005). Biofertilizers are one of the ways to improve the crop yield and quality and also reduces the production cost. It has more advantages than chemical fertilizers which can clean the environment and increase the production (Pham, 2004). The most important concern for an agriculturist is to save the environment for agriculture use (Levai *et al.*, 2006). The reason for the reduction in the fertility of soil were the nutrient loss due to soil erosion, salts and toxic elements accumulation due to water logging and improper nutrient balancing. Nitrogen and phosphate requirements are compensated by using large quantities of chemical fertilizers which has the disadvantage of high cost and environmental issues. The growth of the plant was improved by the organic fertilizers amended to the soil (Ghosh *et al.*, 2004). Plants require macro and micronutrients for their growth in which nitrogen was slowly mineralized and it does not available in required amount resulting in lower yield (Zaman *et al.*, 2004). The soil quality was improved by the organic fertilizers proving their capacity when used for a longer time (Ahmad *et al.*, 2013). Soil microbes are the main factors for the cycling of

*Corresponding author: Victoria, J.

Department of Microbiology, STET Women's College, Sundarakkottai-614016, Mannargudi, Thiruvarur Dt, Tamil Nadu, India

plant nutrients and replace the chemical fertilizers. Soil quality, plant growth and yield were enhanced by the soil microorganisms (Zaman and Chang, 2014). The essential role of beneficial microorganisms were the fixation of atmospheric nitrogen, decomposition of organic wastes to exit plant nutrients, detoxification of pesticides, plant disease and pathogen suppression, improving nutrient cycling and production of many bioactive compounds including vitamins, hormones and enzymes which induces the plant growth (Kuperin *et al.*, 2014). Nutrient supply to the soil can be improved by using the inoculums of Effective microorganisms. This is a newer technology developed by Prof. Teruo Higa from the University of Ryukus, Okinawa, Japan in 1970's (Sangakkara, 2002). EM have a lot of applications such as agricultural livestock, gardening and landscaping, composting, bioremediation, septic tank clearing, algal control and household uses (EM Technology, 1998). Prof. Teruo Higa explained the practical application of EM. He was the first to isolate and screen microbes from soil and make them to coexist compatible with one another when introduced into soil, it produce many beneficial effects in a synergistic fashion (Crawford, 2002).

Effective microorganisms is an inoculum consisting of many naturally occurring beneficial microorganisms used widely in nature and organic farming (Diver *et al.*, 2001). a group of microorganisms having an attractive effect on living forms and environment and explained as the multiculture of aerobic and anaerobic microorganisms that exist compatibly (EM Trading, 2000). It contains selected species of microorganisms such as *Lactobacillus casei*, the lactic acid bacteria, *Rhodospseudomonas palustris*, the photosynthetic bacteria, *Saccharomyces cerevisiae*, the yeast and *Streptomyces albus*, the actinomycete coexist in a liquid media mutually compatible with each other. Sweet sour taste and smell with a pH of below 3.5 were the major characteristics which should stored in an air tight good grade plastic container with provisions to release gas periodically or by opening the container for small quantities. The container should be stored in the temperature between 15 and 20 ° C. Little fluctuations cannot affect the solution. EM improves the quality of the soil and irrigation water system, for seed treatment, as organic sprays which enhance the photosynthesis and control of pests, insects and diseases. Its persistence and dependability on the existing environmental condition was increased and protection was offered against unfavorable condition. Better results from EM can be obtained when mixed with suitable ingredients (Javaid *et al.*, 2008). Cost of the chemical fertilizers makes it out of reach for most poor farmers. Loss of biodiversity occurs due to the usage of man-made agro chemicals. These observations increased the need of organic farming. EM improves the crop yield thereby increasing the food security (Chrispaul *et al.*, 2010).

Waste water contains organic materials from living forms or synthetic organic compounds enter through a number of ways as human wastes, detergents and industrial sources. In the treatment of domestic waste water microorganisms plays an important role in the breakdown of many organic pollutants (Taylor *et al.*, 1997). The major role of microbes in on site systems (septic tanks) is the degradation of organic wastes however some microbes such as bacteria and viruses can cause health problems to humans (Harris *et al.*, 1998). The reason for using EM species is the production of organic acids, enzymes, antioxidants and metallic chelates (Higa and Chinen, 1998).

Antioxidant environment created by EM enhances the solid-liquid separation, the basis for cleaning waste water. In India, soils with low organic content favour the usage of waste water with organic matter as organic amendment and nutrient supply to soil. Though there are numerous benefits in using waste water precautions should be taken to avoid environmental risk. From the earlier studies, the effect of effluents varies from crop to crop. Waste water approximately consists of 99.9% water, 0.02 to 0.08% suspended solids and other soluble organic and inorganic substances. It is weak in nature that is the BOD level is normally low and rich in N and P.

MATERIALS AND METHODS

Collection and Analysis of Waste Water Sample

The waste water was collected in a sterilized plastic container from the women's hostel of 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 parameters analysis. 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 and after the addition of activated EM solution (APHA, 1989).

Collection of Soil Sample

Soil samples were collected from Thiruthuraiipoondi, Thiruvarur District, Tamilnadu, India which consists of Nedumbalam (Ndb) soil series, one of the soil series of Thiruvarur District (Soil Atlas, 2012). Five spots were fixed in a plot for taking one composite mixture of the soil. The surface of the field was scrapped away to obtain a uniformly thick slice of soil from the plough depth from each place. A V-shaped cut was made with a spade to remove 1 to 2 cm slice of soil. The sample collected on the blade of the spade was put in a clean bucket. In the same way the samples were collected from all the spots selected for one sampling unit. Thus the samples were poured on the clean paper and mixed thoroughly. Then the samples were spread evenly and divided into four equal parts. The two opposite quarters were rejected and the remaining samples were mixed. The same process was repeated until the reach of half kg of soil. The sample was collected in a clean bag and marked properly. The mouth of the bag was tied carefully. The same soil was also collected for pot culturing of the plants of *Vigna radiata* for testing the efficiency of EM on the growth parameters and yield of the plants.

Analysis of soil samples

Ndb soil series samples thus collected were first air dried at room temperature, then crushed using a porcelain mortar and pestle and then sieved for further analysis (Kalaivani and Sukumaran, 2013). The physicochemical parameters such as the pH (Ghosh *et al.*, 2004), temperature, EC, Moisture content, Organic carbon (Walkley and Black, 1934), available nitrogen (Subbiah and Asija, 1956), available phosphate (Olsen

et al., 1954), available potassium (Toth and Prince, 1949), available sulphur (Bhargava and Ragupathi, 1993), calcium and magnesium (Cheng and Bray, 1951) were tested before and after the pot cultivation of *Vigna radiata* using EM and waste water treated with EM.

Studies on the growth promoting efficiency of Effective microorganisms in pot culture experiment

A pot culture experiment was conducted using the Klt series soil of Thiruvavur district as the culture medium. Three treatments (T1, T2 and T3) and control (C) pots were maintained. Seeds of *Vigna radiata* were surface sterilized with 1% Sodium hypochlorite for 3 min followed by several washings with sterilized water. The randomized experimental design was set up, T1 - EM alone, T2 - Treated Waste water alone, T3 - EM+ EM treated Waste water, C - Control. Seeds were sown in the pots. Each treatment was replicated 3 times.

Activation (EMa) of EM Stock Solution and application schedule (APNAN, 1995)

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 (Ahmed John et al., 2007). 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. After activation the EM solution was diluted 1:1000 by adding sterilized water. The respective EM treated plots received dilute EM solution (1:1000) at 2 L /m² at fortnight interval throughout the experimental period.

Analysis of plant growth parameters of cultivated plants

Plants were harvested after 35 and 60 days after sowing at flowering and maturity stages, respectively. At flowering stage, alternative plants were harvested from each pot so that more space would be available to the remaining growing plants. At each harvest, plants were uprooted along with the rhizospheric soil and the following parameters were studied. Height of the plant (in cm), Shoot length (in cm), Root Length (in cm), Number of leaves (per plant), Leaf fresh weight (in mg), Leaf dry weight (in mg), Number of nodules (per plant), carbohydrate, protein and chlorophyll contents were recorded at both the harvesting and flowering stage. Number of flowers was recorded in flowering stage only. Root fresh weight (in mg), Root dry weight (in mg), Number of pods (per plant), Length of the pods (in cm) and Yield (in gms) were recorded in maturation. All the data were analyzed statistically by applying Duncan's Multiple Range 't' test (Steel and Torrie, 1980) to separate the treatment means using SPSS.

RESULTS

Analysis of Physico- Chemical Properties Before and After the Treatment of Waste Water

The collected waste water sample was analysed for the physico-chemical properties such as pH, BOD, COD, TS, TDS, TSS, Nitrate and Phosphate contents were recorded (Fig.1).

Analysis of Physico-Chemical Properties of Ndb Soil Series

Physico-chemical parameters of the Ndb soil series were analysed using the standard methods for soil analysis. The soil's physical parameters, primary, secondary and micro nutrients were analysed and the results were recorded (Table-1).

Effect of EM and Treated Waste Water on the Growth Parameters and Yield of *Vigna radiata* Linn

Height of the Plants

All the treatments were observed for the increase in the height of the plants. The treatments showed significant differences ($p \leq 0.05$) for the tested parameter at flowering and maturity stages. The pots inoculated with EM and treated waste water showed the increased height followed by EM alone, untreated waste water and control. The total height comprises of both the shoot and root length of the plant. Shoot growth occurred in all the treatments over the experimental period. There were significant differences in shoot length among the treatments. The treatment containing effective microorganisms and treated waste water showed the highest shoot length at both the flowering and the maturity stages when compared to all other treatments and uninoculated control. Length of the roots was increased highly at the maturity stage when compared to flowering stage in all the treatments. The root length of the plants was insignificant ($P \leq 0.05$) for all the treatments and control at flowering stage whereas in maturity stage T2 and control showed significance in their results (Table-3 and 4).

Number of leaves, Leaf Fresh and Dry Weight

Application of EM enhanced the leaf production in the treatments. The treatments T1 and T3 not showed significant differences ($P \leq 0.05$) in the production of leaves. T2 and control showed significance in their results for the production of leaves at both the flowering and maturity stage. The increase in the leaf number increases the synthesis of chlorophyll pigment which reflects in the production of more number of seeds i.e., the yield is increased. Plants inoculated with EM and treated waste water recorded the highest leaf fresh weight followed by the EM, untreated waste water and control at both the flowering and maturity stage. In flowering stage, T1 and T3 showed significance in their results and T2 and C showed significant results i.e., the leaf fresh weight of EM amended treatments had slightly similar results. T1 and T3 showed significance in their results of increasing the leaf fresh weight when compared to control at maturity stage. At both flowering and maturity stage, treatments inoculated with EM (T1 and T3) showed significant ($P \leq 0.05$) results and uninoculated treatments, T2 and control showed significance in their results (Table- 3 and 4).

Table 1. 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

Values are mean

Table 2. Analysis of physico-chemical parameters of NDB soil

S.No	Physico-chemical parameters	Before cultivation	After cultivation
1	Soil colour	Dark Brown	Dark Brown
2	Soil texture	Clay loam	Clay loam
3	Soil pH	7.8	7.3
4	Electrical Conductivity (dsm ⁻¹)	1.0	0.08
5	Salinity	NS	NS
Primary Nutrients			
6	Organic Carbon (%)	0.59	0.70
7	Nitrogen (Kg/ac)	121.1	132.5
8	Phosphorus (Kg/ac)	4.56	6.23
9	Potassium (Kg/ac)	284	295
Secondary Nutrients			
10	Calcium(ppm)	9.4	10.2
11	Magnesium (ppm)	9.8	10.4
12	Sulphur (mg/l)	33	34
Micronutrients			
13	Iron (%)	7.72	8.12
14	Zinc (%)	1.11	1.98
15	Manganese (%)	3.23	4.15
16	Copper (%)	2.63	2.77
17	Molybdenum (%)	3.82	3.97

Table 3. Analysis of growth parameters of *Vigna radiata* Linn. at flowering stage of NDB Series

S.No	Treatments	Growth parameters							
		Height (in cm)	Shoot length (in cm)	No. of leaves (per plant)	No. of flowers (per plant)	Root length (in cm)	Leaf fresh weight (mg/plant)	Leaf dry weight (mg/plant)	No. of nodules (per plant)
1	T1	31.86a	25.82a	26.00a	16.66a	6.04a	14.67a	4.36a	24.33a
2	T2	28.72b	23.87b	23.66b	14.67b	4.86b	13.65b	4.13b	21.33b
3	T3	31.95c	25.95c	26.66a	17.00a	6.06c	15.36a	4.56a	25.66a
4	C	26.11d	23.98b	22.00c	13.00c	4.81d	13.33b	3.60b	20.66b

Values are mean having the same letters does not show significant difference by Duncan's Multiple Range 't' Test

Table 4. Analysis of growth parameters of *Vigna radiata* Linn.at maturity stage of NDB Series

S. No	Treatments	Growth parameters											
		Height (in cm)	Shoot length (in cm)	No. of leaves (per plant)	Root length (in cm)	Leaf fresh weight (mg/plant)	Leaf Dry weight (mg/plant)	No. of nodules (per plant)	Root fresh weight (mg/plant)	Root dry weight (mg/plant)	Pod length (in cm)	No. of pods (per plant)	Yield (in gms)
1	T1	50.02a	43.56a	31.00a	7.32a	16.77a	3.92a	18.33a	5.47a	1.38a	9.93a	15.67a	4.87a
2	T2	48.72b	42.21b	28.66b	6.38b	15.65b	3.67b	15.33b	4.51b	1.14b	9.12b	13.67b	4.55b
3	T3	52.81c	44.82c	31.66a	7.02c	17.76a	3.66a	18.66a	5.27c	1.33c	10.00c	16.33a	4.99c
4	C	40.45d	33.91b	27.00c	6.33b	15.13c	3.58b	14.66b	4.36d	0.95d	8.11d	12.00b	3.97d

Values are mean having the same letters does not show significant difference by Duncan's Multiple Range 't' Test

Table 5. Analysis of Total Chlorophyll, Carbohydrate and Protein content of *Vigna radiata* at flowering and Maturity stage of Ndb Series

S.No.	Treatments	Flowering stage					Maturity stage				
		Chlorophyll					Chlorophyll				
		Chl-a	Chl-b	Total Chl	Carbohydrate	Protein	Chl-a	Chl-b	Total Chl	Carbohydrate	Protein
1	T1	0.187	0.160	0.347	19.51a	80.33a	0.146	0.126	0.272	15.42a	40.88a
2	T2	0.178	0.148	0.326	18.13b	77.47b	0.134	0.107	0.241	14.12b	36.70b
3	T3	0.188	0.165	0.353	19.54c	80.66c	0.148	0.128	0.276	15.53c	41.23c
4	C	0.170	0.144	0.314	17.99d	76.15d	0.130	0.104	0.234	13.90d	36.41d

Values are mean having the same letters does not show significant difference by Duncan's Multiple Range 't' Test for carbohydrate and protein analysis.

Root Fresh and Dry weight

Maximum root fresh weight was recorded in the treatments inoculated with EM. There were significant differences among the treatments for this parameter. At maturity, the treatments with EM showed significance and unamended showed significance at ($P \leq 0.05$) i.e., T1 and T3, T2 and Control respectively. Maximum root dry weight was recorded in the treatments inoculated with EM. There were significant differences among the treatments for this parameter at maturity ($P \leq 0.05$) i.e., T1, T3, T2 and Control respectively. Root dry weight showed quarter the weight of root fresh weight (Table - 4).

Number of flowers and Pods and pod length

The parameter, number of flowers was studied at the flowering stage. The maximum the number of flowers reflects the maximum the number of pods and the yield. Treatments, T1 and T3 showed significance in their results and T2 and C showed significance which represents that the number of flowers in the EM amended soils showed significance and the treatment with unamended soils showed the results as such of control (Table-3). The plant *Vigna radiata* showed an increased number of pods in pots treated with EM. T1 and T3 showed a significant ($P \leq 0.05$) result when compared to other treatment and control. In T3 the number of pods was significantly high showing the best result of the parameter, and then came the T1, T2 and Control (Table-4). Length of the pods was studied for analyzing the improvement of seed yield per plant. Increased pod length was observed in the treatment T3 followed by T1. T1 and T3 showed the significant ($P \leq 0.05$) results and the treatments, T2 and C showed the significant results i.e., the length of the pods was similar in significant results (Table -4).

Number of Nodules and Yield (in gms)'

Number of nodules was studied at both the flowering and maturity stage. The parameter showed the reduction in the number at maturity stage when compared to the flowering stage due to the decomposition of the nodules. The increase in the nodule numbers was observed in the EM amended treatments than the others. When the nodule number increases, it increases the fixation of nitrogen in the soil improving the plant growth and yield. T3 showed highest nodule number followed by T1, T2 and Control. The results were significant ($P \leq 0.05$) for the EM inoculated treatments than the others (Table -3 and 4). Yield of the plant results due to the increase in all the parameters studied above. There were significant differences ($P \leq 0.05$) in the yield of the plants. Treatment T3 showed the highest yield when compared to other treatments. Thus, the results revealed that yield of the plant were significantly increased due to the amendment of EM to the soil (Table- 4).

Carbohydrate and Protein Content

Increased carbohydrate content was observed in the treatments inoculated with EM. All the treatments showed significant differences ($P \leq 0.05$) in the carbohydrate content. Among the treatments, T3 showed significant increase in the carbohydrate content followed by T1, T2 and Control respectively. Protein content of the plants was increased in the EM amended soils when compared to unamended treatments. All the treatments

showed significant differences ($P \leq 0.05$) in the protein content. Among the treatments, T3 showed significant increase in the protein content followed by T1, T2 and Control respectively (Table-5).

Chlorophyll content

The chlorophyll content of the plants was increased in the treatments inoculated with EM. If the chlorophyll content was increased then the synthesis of carbohydrate and protein content were also increased which increases the yield of the plants. The total chlorophyll was calculated by analyzing the chlorophyll- a and chlorophyll- b contents of the plants. T3 showed the highest chlorophyll content followed by T1, T2 and Control (Table-5). Thus, EM along with EM treated waste water showed an increased level in all the treatments followed by EM alone.

DISCUSSION

Treatment disposal and recycling of sewage sludge is the major problem of municipalities throughout the world. Municipal wastes contain biodegradable organic materials and a small amount of organic matter (Elliot, 1986). The physical, chemical and biological properties of sludge show greater variations (Collin *et al.*, 1988, Bruce, 1990). Many methods are used to dispose sewage sludge nowadays from landfill to land application, but the presence of heavy metals, pathogens and toxic substances are the major issues, so proper and selective method should be used for efficient and environmentally safe disposal. Following the strict environmental regulations, newer technologies are used to help in the treatment and safe disposal of sewage sludge. One of the new technologies proposed is the use of Effective microorganisms (Sangakkara, 2002). This study was aimed to treat the waste water from the Women's hostel consisting of organic and inorganic wastes using Effective Microorganisms technology. Effective microorganisms decompose the organic matter by converting it into carbon dioxide (CO_2) and methane (CH_4) and makes the unavailable forms of nutrients to available forms for their utilization in growth and reproduction which occurs in both waste water treatment plants and septic tanks. Freitag (2000) suggested that the introduction of EM in anaerobic treatment plants reduce the unpleasant byproducts and also the production of residual sludge. Thus, EM plays an important role in the treatment of waste water and reduce the amount of sewage sludge. Karthick Raja Namasivayam *et al.* (2011) tested all the parameters under the tested incubation period. He found that the parameters showed reduction in their values except dissolved oxygen. 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 (Karthickrajanamasivayam *et al.*, 2011). 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). 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 (Table-1). 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. Malarvizhi *et al.* (2009) conducted experiments on seven different soil series of Coimbatore (Sandy clay loam, Typic Haplustalf), Dindigul district (SCL, Typic Rodustalf). Analysis of the soils indicated that Igr, Tlk, Pvd and Pld soil series showed an alkaline pH and non-saline in nature. The level of organic Carbon, Nitrogen, phosphorus and zinc were very low in most of the soil series. Likewise, this study also focuses on the Nedumbalam (Ndb) soil series of Thiruvavur district. The initial soil analysis reports showed the reduced level of organic carbon, nitrogen, and phosphorus and they were increased after treatment with EM.

Micronutrient plays a vital role in maintaining soil health and also productivity of crops. These are needed in very small amounts. The soil must supply micronutrients for desired growth of plants and synthesis of human food. Increased removal of micronutrients as a consequence of adaption of HYVs and intensive cropping together with shift towards high analysis NPK fertilizers has caused decline in the level of micronutrients in the soil to below normal at which productivity of crops cannot be sustained. The deficiencies of micronutrients have become major constraints to productivity, stability and sustainability of soils. Soils with finer particles and with higher organic matter can generally provide a greater reserve of these elements whereas, coarse textured soils such as, sand have fewer reserves and tend to get depleted rather quickly. The soil reaction (pH) of these soils ranged from 4.7 to 9.2 *i.e.*, slightly acidic to alkaline reaction. Among the soil series, the average soil pH of 7.76 was recorded in the Somayyanur series followed by 8.03 in the Palaviduthi series and 7.98 in Irugur series. Among the three major soil series, 73% of the soil samples from Irugur followed by 60% and 59% from Somayyanur and Palaviduthi series, respectively exhibited moderate alkalinity. With the increase of pH, the availability of micronutrients decreased, which probably might be due to alkaline nature of soils (Rakesh Kumar *et al.*, 2009). Perveen *et al.* (1993) studied micronutrient status of some agriculturally important soil series of the Northwest Frontier Province, Pakistan and their relationship with various physical chemical properties for 30 soil series. Most silty soils (coarse texture) are deficient micronutrients. Clay soils (fine texture) are not comparatively to low plant available micronutrients. Chhabra *et al.* (1996) studied that available Mg and I decreased with soil pH and available Cu increased with clay and organic carbon content.

Vaxevanidou *et al.* (2015) suggested that chemical fertilizers, pesticides, herbicides and other agricultural inputs derived from fossil fuels have increased agricultural production, yet the growing awareness and concern over their adverse effects on soil productivity and environmental quality cannot be ignored. The high cost of these products, the difficulties of meeting the demand for them, and their harmful environmental legacy have encouraged scientists to develop alternative strategies to raise productivity, with microbes playing a central role in these efforts. This study was also an attempt to prove the efficiency of microbes as inoculants to improve the crop yield and to protect the environment from deterioration. From this study it

was revealed that the EM treated waste water served as the better source for irrigating the cultivated plant. Nutrients in waste water along with the organisms enhanced the growth of the plants and also increased the natural flora of the soil. When the rhizospheric flora was increased, the soil becomes more fertile when compared to the untreated control.

Karthick Raja Namasivayam and Arvind Bharani (2012) studied the distinct differences in all the tested growth parameters of *Vigna mungo* were recorded in compost (prepared using EM) treated plots. There was a significant increase in the shoot length, total height of the plant, leaf number per plant, leaf surface area, total new branch emerged, total foliage count per plant was observed. Increase in leaf area and number should result to higher rates of photosynthesis hence increased plant growth. For plants, a high rate of net carbon assimilation can result in higher biomass accumulation, favouring future growth and reproduction. The position and distribution of leaves along shoot influences the sink strength of the plants. During early stages of leaf growth, synthesis of chlorophyll, proteins and structural compounds is high resulting in high catabolic rates to support energy needs by the plants. Inoculation of effective microorganisms can increase the available nutrition for plant roots and improve photosynthesis (Chrispaul Mataura *et al.*, 2010). The stem diameter of the cucumber, pumpkin, and squash transplants was significantly larger in EM treatment in both the experiments when compared to control (Margit Olle, 2015). The results from the study indicate that inoculation of *Vigna radiata* with effective microorganisms increased the height of the plant (in cm), Internodal length (in cm), Shoot length (in cm), Root Length (in cm), Number of leaves (per plant), Leaf fresh weight (in mg), Leaf dry weight (in mg), Number of flowers (per plant) Number of nodules (per plant), Root fresh weight (in mg), Root dry weight (in mg), Number of pods (per plant), Length of the pods (in cm) and Yield (in gms), carbohydrate, protein and chlorophyll contents. Increase in leaf number, leaf fresh and dry weight increase the photosynthetic activity of the plants.

Beadle (1993) studied the increase in chlorophyll contents of pigweed may contribute to increased photosynthetic activity. The synthesis and degradation of the photosynthetic pigments are normally associated with the photosynthetic efficiency of the plants and their growth adaptability to different environments. Sharma and Namdeo (1999) also explained that the increase in leaf chlorophyll content could in turn lead to increased protein synthesis of the plants and this could have a direct consequence on the plant growth and photosynthesis. In this study, chlorophyll a and b contents of the plants were increased in all the treatments whereas the treatments inoculated with effective microorganisms showed relatively higher rate of chlorophyll synthesis. When the chlorophyll content increases it increases the synthesis of protein and carbohydrate contents thus increasing the growth parameters and yield of the plants. Nitrogen is one of the essential nutrients involved as a constituent of biomolecules such as nucleic acids, coenzymes and proteins (Hendry *et al.*, 1987), any deviation in these constituents would inhibit the growth and yield of plants. Protein concentrations in plants tend to increase with fertility level of the growth medium (Grant and Bailey, 1993). In this study, there was the increase in the number of nodules at the flowering stage increasing the nitrogen fixation thus increasing the nitrogen content of the plants. If the constituents decrease it increases the plant

growth, yield and nutrient status of the plants. In general, effective microorganisms produced a direct impact on growth and yield of *Vigna radiata*. Previous studies have demonstrated a consistent positive response with the use of effective microorganisms in crop production and indicate the potential of this technology to reduce fertilizer use and increase the yield and quality of crops (Higa and Widhana, 1991). Thus this study proved the efficiency of waste water treated with EM increased the yield of the crop plant tested. It paves the way for reducing and recycling the wastes in a proper way. the presence of nutrients improved the soil nature and also the yield of the crop in a cost effective manner. Better alternative for waste water treatment and to renew the fertility of the soil.

Conclusion

The results of this study reveal that the inoculation of effective microorganisms for the cultivation of *Vigna radiata* improved all the growth parameters, carbohydrate, protein and chlorophyll content of the plant. EM already having many beneficial potentialities, it produced more effects along with waste water treated with EM. Although waste water having much more nutrients, it showed more effects on plant growth on combination with Effective microorganisms. Once the organisms in EM flourish as normal flora of the soil, there will be reduction in the need of applying any of the fertilizers. Day by day, the application of chemical fertilizers to the soil will make it sterile in the future i.e., making it unfit or unfertile for cultivation of crops. Thus to prevent environmental pollution and to reduce the extensive use of chemical fertilizers, the effective microorganisms can be recommended to the farmers to ensure public health and a sustainable agriculture. Steps have to be taken to introduce organic farming to the agrarians to achieve the goal of protecting the fertility of their cultivable lands.

Acknowledgement

We express our sincere thanks to Dr.V. Dhivaharan, Dean, Department of Lifesciences, STET Women's College, Sundarakkottai, Mannargudi, Thiruvavur Dt, Tamilnadu, India for providing us all the facilities and encouragement for completing this study successfully.

REFERENCES

- Ahmad, W., Z. Shah, F. Khan, S. Ali and W. Malik.2013. Maize yield and soil properties as influenced by integrated use organic, inorganic and bio-fertilizers in low fertility soil. *Soil Environ.*, 32(2): 121-129
- Ahmed John, S., Ahamedjeelanibasha, A., Packialakshmi, N., Lawasheeladevi, A., 2007. Application of Effective Microorganisms for the purification of different kinds of water. *Asian jr. of Microbiology. Env.Sci.*, 9,337-340
- 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.
- Asia-Pacific Natural Agriculture Network. 1995. EM Application manual for APNAN countries, Shintani, M. (ed)1st Asia-Pacific Natural Agriculture Network, Bangkok, Thailand, 34.
- Beadle, C.L., 1993. Growth analysis. In: Hall, D.O., Scurlock J.M.O., Bolh ar-Nordenkampf H.R., Leegood R.C., Log S.P., (eds), Photosynthesis and production in a changing environment- A field and laboratory manual. 36-46. Chapman and Hall, London.
- Bhargava, B.S., Ragupathi, H.B., 1993. Analysis of plant materials for macro and micronutrients. In: methods of analysis of soils, plants, waters and fertilizers (Ed. By H.L.S Tandon) Fertilizer Development and Consultation Organization, New Delhi.
- Cheng, J.L., and Bray, R.N., 1951. Determination of calcium and magnesium in soil and plant materials. *Soil Science*. 72.449-459.
- Chhabra, G., Srivastava, P. C., Ghosh, D and Agnihotri, A. K. 1996- Distribution of available micronutrient cations as related to soil properties in different soil zones of Gola-Kosi interbasin. *Crop Research-Hisar*. 11(3).296-303.
- Chrispaul Mathaura, David M., Musyimi, Joseph A., Ogur Samuel V., Okello. 2010. Effective Microorganisms and their influence on growth and yield of pigweed. *ARPN Journal of Agriculture and Biological Science*, 5(1).17-22.
- 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 Sludge', in *Journal of Soil and Water Conservation*, (41). 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>.
- Freitag, D.G. 2000 (updated 3 May 2000, accessed 27 Aug 2002), 'The use of Effective Microorganisms (EM) in Organic Waste Management' Effective Microorganisms @emtrading.com, http://emtrading.com/em/htmlpapers/em_wasterepfreitag.html#9.
- Freitag, D.G. 2000 (updated 3 May 2000, accessed 27 Aug 2002), 'The use of Effective Microorganisms (EM) in Organic Waste Management' Effective Microorganisms @emtrading.com, http://emtrading.com/em/htmlpapers/em_wasterepfreitag.html#9.
- Ghosh, P.K., Ramesh, P., Bandyopadhyay, K.K., Tripathi, A.K., Hati, K.M., Misra, A.K. 2004. Comparative effectiveness of cattle manure, poultry manure, phosphocompost and fertilizer- NPK on three cropping systems in vertisols of semi arid tropical crop yield and systems in performance. *Bioresource Technology*, 95.77-83.
- Grant, C.A. and Bailey, L.D. 1993. Fertility management in Canola production. *Canadian J. Plant Science*. 73.651-670.
- Harris, B.L., Hoffman, D.W. and Mazac Jr, F.J. 2001. (updated 15 Aug 2001, accessed 27 Aug 2002), 'Reducing Contamination by Improving Household Wastewater Treatment' *Blackland Research Centre Water Sciences Laboratory*. <http://waterhome.brc.tamus.edu/texasyst/household.html>.

- Hendry G.A.F., Houghton J.D., Brown Tansley S.B. 1987. Review No 11: The degradation of chlorophyll-A biological enigma. *New Phytologist*, 107. 255-302
- Higa, T. 1995. Effective microorganisms : their role in Kyusei Nature Farming and Sustainable Agriculture. In: Parr J.F., Hornick 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
- Higa, T. and Chinen, N. 1998. 'EM Treatments of Odor, Waste Water, and Environmental Problems', College of Agriculture, University of Ryukyus, Okinawa, Japan.
- Higa, T. and Widhana, G.N., 1991. Concept and theories of effective microorganisms. In: Proceedings of 1st Kyusei Nature Farming. (Eds. JF Parr, SB Hornick, CE Whitman), USDA, Washington, D.C. USA, October 17-21, 1989, 118-124.
- 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.
- Kalaivani, R. and Sukumaran, V. 2013. Characterization of bacterial diversity in marine sediment at different seasons in Karanadu, Tamilnadu, India. *Int. J.Pure and Applied Zoology*, 1(1). 97-108.
- Karthick Raja Namasivayam, S., Arvind Bharani, R.S. 2012. Effect of compost derived from decomposed fruit wastes by Effective Microorganisms (EM) Technology on plant growth parameters of *Vigna mungo*. *J.Bioremed Biodeg.*, 3.16
- Karthick Raja Namasivayam, S., Narendrakumar,G.,Arvind kumar, J. 2011. Evaluation of Effective microorganisms (EM) for the treatment of domestic sewage. *J.Exp.Sci.*, 2(7).30-32.
- Kurepin. L. V., J. A. Ozga., M. Zaman and R. P.Pharis. 2013. The physiology of plant hormones incereal, oilseed and pulse crops. *Prairie Soils Crops*, 6: 7-23.
- Lévai L., Veres S.Z., Makleit P., Marozsán M., Szabó B. 2006. New trends in plant nutrition. Proceedings of 41st Croatian and 1st International Symposium on Agriculture, ISBN 953-6331-39-X, pp. 435-436.
- Malarvizhi, P., S. Thiyageshwari, M. Paramasivan, R. Geetha, V. Kasthuri Thilagam, T. Nagendra Rao and T. Satyanarayana.2009. Nutrient Management to Improve Maize Productivity in Tamil Nadu. *Better Crops- India*.22-24
- Margit O. and Ingrid Williams, 2015. The influence of Effective Microorganisms on the growth and nitrate content of vegetable transplants. *J.Adv.Agr.Tech.*, 2(1).25-28.
- Nishio, M., 1996. Microbial Fertilizers in Japan. Food and Fertilizer Technology Center. 12.
- Olsen, S.R., Cole, C.V., Watanabe, F.S., Dean, L.A., 1954. Estimation of available phosphorus in soils by extraction with sodium bicarbonate. U.S.Department of agriculture.
- Parr, J. F., Higa, T., 1994. Beneficial and effective microorganisms for a sustainable agriculture and environment. International Nature Farming Research Center, Atami, Japan.
- Perveen, S., M. Tariq, Farmanullah, J. K. Khattak and A. Hamid. 1993. Study of micronutrient status of some important sites of N.W.F.P., Pakistan. *Sarhad Journal of Agriculture*, 9(5): pp467-473.
- Pham, D.T., 2004. FNCA Biofertilizer Newsletter. Japan Atomic Industrial Forum, Inc. 4.1-8.
- Piqueres, P.A., Hermann, E.V., Alabouvette, C., Steinberg, C., 2005. Response of soil microbial communities to compost amendments. *Soil Biology and Biochemistry* 38. 460-470.
- Poincelot, R.P. 1986. Toward a More Sustainable Agriculture. AVI publishing Company Inc., Conneticut. 14.
- Rakesh Kumar, A., Sarkar, K., Singh, K.P., Agarwal, B.K. and Karmakar, S. 2009. Appraisal of available nutrients status in Santhal Paraganas, Region of Jharkhand. *J. Indian Soc. Soil Sci.*, 57(3): 366-369.
- Rakesh Kumar, A., Sarkar, K., Singh, K.P., Agarwal, B.K. and Karmakar, S. 2009. Appraisal of available nutrients status in Santhal Paraganas, Region of Jharkhand. *J. Indian Soc. Soil Sci.*, 57(3): 366-369.
- Sangakkara, U.R. 2002. The Technology of Effective Microorganisms : case studies of application. Cirencester, UK : Royal Agricultural College. Exp. Sci. 2(7). 30-32.
- Sharma K.N., Namdeo, K.N., 1999. Effect of biofertilizers and phosphorus on NPK contents, uptake and grain quality of soybean (*Glycinemax* L.Merill).*Crop.Res.(Hisar)*, 17.164-169.
- Singh, H. 1995.Nitrogen mineralization, microbial biomass and crop yield as affected by wheat residue placement and fertilizer in a semi-arid tropical soil with minimum tillage. *The Journal of Applied Ecology*, 32 (3). 588-595.
- Soil Atlas of Thiruvarur District, 2012. Soil Survey and Land use Organization, Department of Agriculture, Tamil Nadu.
- Steel, R.G.D., Torrie, J.H., 1980. Principles and procedures of statistics. A Biometrical Approach. 2nd ed. McGraw Hill Book Co. Inc.New York, USA.
- Subbiah, B.V. and Asija, G.L. 1956. Analysis of mineralizable nitrogen from soil.*Curr.Sci.*25.258-260.
- Taylor, C., Yahner, J., Jones, D. and Dunn, A. 1997. 'Wastewater' in *Pipeline*, 8 (4).
- Toth, S.J., Prince, A.L., 1949. Estimation cation exchange capacity and exchangeable Ca, K, and Na contents of soils by Flame photometer. *Soil Science*, 64.439-446.
- Vaxevanidou, K., Christou, C., Kremmydas, G.F., Georgakopoulos, D.G. and Papassiopi, N. 2015. Role of indigenous arsenate and iron (III) Respiring microorganisms in controlling the mobilization of arsenic in a Contaminated soil Sample. *Bull Environ Cont. Toxicol.*, 94(3).282-288
- Walkley, A. and Black, I.A. 1934. Analysis of organic carbon from soil. *Soil Science*, 37. 29-38.
- Zaman, M. and S.X. Chang. 2004. Substrate type, temperature, and moisture content affect gross and net soil N mineralization and nitrification rates in agro forestry systems. *Biol Fertil Soils*, 39(4): 269-279.
- Zaman, M., L.V. Kurepin, W. Catto, R.P. Pharis, 2014. Enhancing crop yield with the use of N-based fertilizers co-applied with plant hormones or growth regulators. *J. Sci. Food and Agri.*, 1002(10): 6938.
