

Role of effective microorganism in unfertile soil

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Abstract

The present study was conducted to evaluate the effect of Effective microorganisms (EM). The EM isolation is very important for agricultural fields. For this study used the different kinds of natural ingredients such as banana, papaya, pumpkin, egg, cane molasses and neem powder to added and mixed and wait for the fermentation. After 45 days the samples were collected. The collected sample were identified using plating technique, microscopic studies and Biochemical test. The identified effective organism was *Bacillus megaterium*. These Effective organisms acting against the pathogen. The results concluded maximum zone of inhibition against the pathogen Such as *E.coli* (16mm), *P.aeruginosa* (18mm), *K.pneumoniae* (19mm), *S.aureus* (17mm), *S.epidermis* (16mm). The microbial population also counted in control and EM treated soil. The observation revealed that the EM treatment increased the microbial population. The present study suggests that the increased microbial population may be responsible for the increased growth and yield of green gram.

Keywords: Effective microorganisms, Pathogens, Antibacterial activity, MIC

1. Introduction

The effective microorganism is a commercial biofertilizer that contains a mixture of co-existing beneficial microorganism collected from natural environment. Effective microorganism was developed at the University of the Ryukyus, Japan, in the early 1980 by Pro. Dr. Terou Higa. Approximately 80 different microorganisms is capable to positively influencing decomposing organic substance such that it reverts into a "Life promoting" process.

EM is a fermented mixed culture of naturally occurring species of co-existing microorganisms in acidic medium (PH below 3.5). Among the main microorganisms in EM culture are the species of photosynthetic bacteria (*Rhodospseudomonas plastris* and *Rhodobacter sphaeroides*), lactobacilli (*L.plantarum*, *L.casei*, and *streptococcus lactis*), yeast (*saccharomyces spp*) and actinomycetes (*streptomyces spp*). Microorganisms in EM improve crop health and yield by increasing photosynthesis, producing bioactive substance such as hormones and enzymes, accelerating decomposition of organic materials and controlling soil borne disease^{7, 8}. Now days, Arbuscular mycorrhizal fungi (AMF) and plant growth-promoting rhizobacteria (PGPR) are used to improve soil fertility and plant growth^{1, 12, 13}.

Effective microorganisms can be used as an herbal insecticide to control insects and pathogenic microorganisms and also be used as plant growth inducer. Soil microorganisms have an important influence on soil fertility and plant health⁴. The present study deals the Effective organism used as an antibiotic to acting against the pathogen. EM treatment increases more chlorophyll content and which increases the protein synthesis⁶.

2. Materials and methods

2.1 Preparation of Effective microorganisms: Papaya(1kg),pumpkin(1kg),banana(1kg) was collected as the equal volume .The ingredients were chopped and mixing well and this content mixed to the cane molasses (1/2kg),egg(1)and neem powder (1/2kg). These mixer transferring 5 liters of distilled water, mixed well and tightly sealed. The cane mixed well twice a day by rotating the container. The process is continuing for 45 days. After 45 days the EM solution is filtered and transferred to the sterile bottles.

2.2 Unfertile soil collection: The unfertile soil was collected from Thanjavur (Dk), Tamilnadu, India.

2.3 Source materials: The green gram seeds, effective microorganism's solution, unfertile soil.

2.4 Preparation of seeds: The seeds were soaked in EM solution for overnight and planted on the pots on the day after.

2.5 Collection of EM solution: EM solution was collected from the various time duration 45, 50, 55, 60 and 65 days. And this solution is mixing with the unfertile soil. The green gram seeds are also mixing with EM solution. The seeds are sowing to the EM solution mixing infertile soil pot.

2.6 Isolation and identification of effective microorganisms: Effective microorganisms were isolated from the EM solution by using spread plate technique.

The colonies were identified by using of Gram staining and Biochemical test.

2.7 Anti bacterial activity: The antibacterial activity of selected bacterial strains such as *Escherichia coli*, *pseudomonas aeruginosa*, *Klebsiella pneumonia*, *Staphylococcus epidermidis*, *Staphylococcus aureus* were carried out using standard disc diffusion method.

2.7.1 Determination of Minimum Inhibitory Concentration (MIC): Each bacterial strain was diluted 1:10 with fresh sterile Muller Hinton broth, streaked on the agar plates in a radial fashion and incubated at 37°C aerobically for 24-48hrs. Complete suppression of growth by specific concentration of an extract was regarded active. Each extract was examined at a concentration of 0.1, 0.5, 1.0, 5.0 & 10.0 mg/ml.

2.7.2 Chi-square test (χ^2): In this study chi-square test (χ^2) was applied. The purpose of chi-square test (χ^2) was to decide whether the set of observed data (antibiogram of microorganisms) agrees with the standard antimicrobial discs susceptibility test (NCCLS, SS2002).

3. Result and Discussion

The present study Effective microorganisms were prepared using different kinds of natural source like banana, papaya, pumpkin, egg, cane molasses and neem powder (Fig.1). After fermentation, the EM solution was collected and identified the Effective organisms, through Biochemical test (Table-1). The present study EM solution was treated green gram seeds after treatment analysis the microbial population (Bacteria, fungi, Actinomycetes). The microbial population was increased in EM treated plant compared with control.

The earlier study reported that Nitrogen is the most important plant nutrient required for plant growth which is more abundant in the earth atmosphere¹⁰. Many microbes are involving in the process of biological nitrogen fixation and the enrichment of microbes is the alternative source for Nitrogen¹¹. The highest grain yield in soybean when host plant was inoculated with *Bradyrhizobium* in combination with NPK fertilizers. Effective microbes treatment brings better yield in plants³. Application of EM is known to enhance crop growth and yield in many crops both leguminous and non-leguminous^{9, 2}. Bacteria, fungi, and Actinomycetes are known plant growth promoting microorganisms which are survives in and around the root rhizosphere. These microbes enhance the plant growth and yield either directly or indirectly⁵. In the present study, the microbes such as bacteria, fungus and actinomycetes are increased in their population when treated with Effective microorganisms.

In the present study observed the minimum inhibitory concentration determined by the MIC values for *Escherichia coli* (16mm), *Staphylococcus aureus* (17mm), *Pseudomonas aeruginosa* (16mm), *Klebsiella pneumoniae* (19mm), *Staphylococcus epidermidis* (16mm). Antibacterial activity was done at various concentrations such as 128 µg, 64 µg, 32 µg and 16 µg for the test organism (Table 2). The works conclude that 64 µg concentrations get the maximum zone of inhibition for all pathogens (Fig 2).

The chi-square values obtained respectively which was less than the calculated table value. $\chi^2_{(0.05)-3,841}$ at 5% level of significance. Above results lead to the conclusion that the data was consistent with the hypothesis, the diameter of inhibition zone obtained from the observed data showed the similarities with experimental data.

The present study concludes that *Bacillus megaterium* acting against the pathogens. Because it release the antibacterial substance, vitamins, growth promoting hormones. So this work suggested the EM solution applying field gave the maximum yield.

Fig.1. Preparation of Effective microorganisms



Fig 2. Zone of inhibition by *Bacillus megaterium* against pathogens

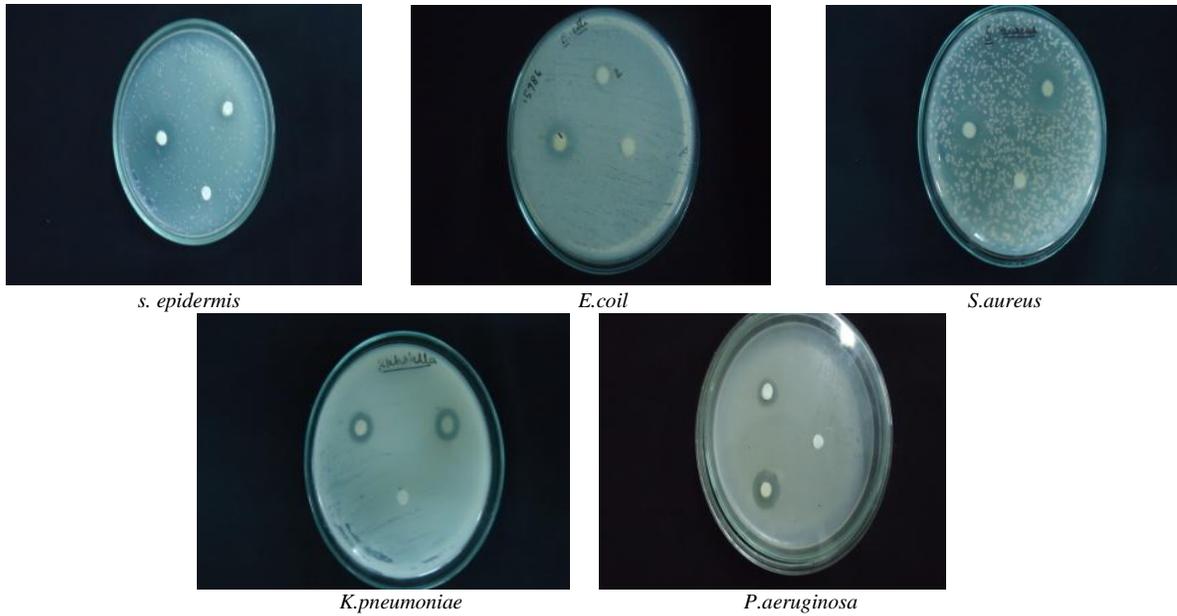


Table 1: Biochemical Test

Biochemical test	<i>Bacillus megaterium</i>
Indole	Negative
MR/VP	Positive/Negative
Citrate	Negative
Nitrate	Negative
Urease	Negative
Catalase	Positive
Oxidase	Positive
TSI	H2S negative/acid slant

Table 2. Zone of inhibition by *Bacillus megaterium* against bacterial strain

S. no	Sample	Bacterial strain	µg	Zone of inhibition in diameter(mm)		$\chi^2 = [\sum(o-E)^2]/E$
				standard value	observed value	
1	Effective microorganism solution	<i>E. coli</i>	128µg	20	16	0.8
2		<i>P. aeruginosa</i>		20	18	0.2
3		<i>K. pneumoniae</i>		20	19	0.05
4		<i>S. aureus</i>		20	17	0.45
5		<i>S. epidermis</i>		20	16	0.8

Table value $\chi^2(0.05) = 3.841$.

Chi-square value significance at 5% level

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