Available online at www.journalijmrr.com

INTERNATIONAL JOURNAL OF MODERN RESEARCH AND REVIEWS

ISSN: 2347-8314

Int. J. Modn. Res. Revs.
Volume 4, Issue 1, pp 1094-1097, January, 2016

ORIGINAL ARTICLE

GROWTH PROMOTING EFFECT AND BIOCHEMICAL CHARACTERISTICS OF CYANOBACTERIAL BIOFERTILIZER (*OSCILLATORIA FORMOSA*) ON TOMATO (*LYCOPERSICUM ESCULENTUM*)

***[1]K.Sivakamasundari and [2]R. Rajendran**

[*1]Department of Microbiology, Research & Development Centre, Bharathiar University, Coimbatore.
[2]Department of Botany, Annamalai University, Chidambaram.

Article History: *Received 2nd November,2015, Accepted 30th December, 2015, Published 31st January,2016*

ABSTRACT

In this study, the effect of cyanobacterium *Oscillatoria formosa* as a biofertilizer on growth and its biochemical characteristics work studied of tomato plant. The experiments were designed as control, biofertilizer applied and recommended level of chemical fertilizer applied batches. The growth parameters such as seed germination, root length, shoot length, number of leaves per plant, lateral roots, chlorophyll 'a' and chlorophyll'b' contents were observed. The uptake macro and micro nutrients to roots and shoot system were also analyzed. The good results were observed in biofertilizer treated plants in all respects and the results suggested that the treatment of cyanobacterial biofertilizer enhance the growth and biochemical contents of tomato plants when compared to chemical fertilizer treated plants and control.

Keywords: Cyanabacteria, Biofertilizer, Tomato, Growth parameters, Macronutrients, Micronutrients

1.INTRODUCTION

The phototrophic species of cyanobacteria are major components of nitrogen fixing in agricultural fields and moist soils. By this reason, they have been recommended as a biofertilizer for rice fields in many countries. Chemical fertilizers have been used in large quantity to compensate the nutrient deficiency in land during crop cultivation. It has been found that the indiscriminate large scale application of chemical fertilizer ultimately affect the lands and plants. Hence, the necessity of doing research on alternate, environment friendly fertilizer has been emphasized in recent years. By this reason, they have been recommended as a biofertilizer for agricultural and horticultural fields in many countries and expected to reduce the use of chemical fertilizers and pesticides in agricultural fields and their hazards. In this way, cyanobacterial studies as biofertilizer have become very important. cyanobacteria have a unique potential to contribute to enhance productivity in a variety of agricultural and ecological situations. The capacity of several cyanobacteria to fix the atmospheric nitrogen is a significant biological process of economic importance (Venkataraman, 1981, Santra, 1993). The role of nitrogen fixing cyanobacteria in improving the fertility of the soil was well documented (Venkataraman, 1981). Cyanobacteria play an important role in build-up of soil fertility and consequently increasing the yield. Hence, the present attempt has been made to study the growth promoting effect and biochemical characteristics of cyanobacterial biofertilizer (*Oscillatoria formosa*) on Tomato plant.

Corresponding* author: ***K.Sivakamasundari,* Research Scholar, Department of Microbiology, Research & Development Centre, Bharathiar University, Coimbatore

2.MATERIALS AND METHODS

In this present study, the blue green algal samples were collected from Perumal Eri (Lake) located in cuddalore district of Tamilnadu State. Samples were collected using forceps, knifes and plankton nets (mesh size 42µm). The Lake lies between north latitudes 11 0 30' to 11 0 45' N and East longitudes 79 0 30' to 79 0 47'30'' E. It falls in the survey of India toposheet no. 58 M/10. The collected cyanobacterial samples were transferred to conical flasks containing BG 11 medium. The cyanobacterial species were identified by using the keys of Desikachary (1959), Prescott (1951) and Wehr and Sheath (2003). After the identification, the individual species of *Oscillatoria formosa* was isolated and cultured in the laboratory condition by using (Rippka *et al*., 1979) method. The cultured species was collected and used as biofertilizer on tomato plant. In the present investigation, pots (25X30) were used for rasing the crops. The pots were filled with 3 kg soils and ten seeds were sown

at a depth of 1cm in each pot. They were kept in the net house to prevent damages caused by birds, fats, squirrels and other animals. The pots were labeled with particular treatment and rearrangement at regular intervals so as to ensure uniform environmental factor on the plant growth. The experiments were conducted with three batches. While the seeds of control batch were treated only with tap water, the biofertilizer batches were treated with lab cultured O. *formosa* and last batches were treated with recommended level chemical fertilizer. All experiments were conducted in triplicates between August and September 2013. During the course of experiment, temperature varied from 24 to 31°C. The weeds were removed regularly and watering was done once in 2 days for the test plants. Each treatment was randomly drawn for various analyses. On the 25th day, some plants from the pots were uprooted carefully and washed with tap water. Then they were processed for different analyses for growth parameters such as root length, shoot length, lateral roots, numbers of leaves, chlorophyll'a' and chlorophyll'b' and biochemical of macro and micro nutrients in root and shoot system.

Macro and micronutrients analyses

The biofertilizer, chemical fertilizer and control treated plants root and shoot were analyzed for macro and micronutrients.

Total nitrogen (N) (Bremner. 1960)

Total nitrogen content of plant material was estimated at 25th day of plant material. The plant materials were air dried first then dried to a constant temperature in a hot air oven at 80°C. They were then powdered, sieved and preserved in vials. Sample powder weighing 100 mg was transferred to a 50 ml Pyrex micro kjeldhal flask. A quarter teaspoonful of digestion mixture (10 parts of reagent grade potassium sulphate. 1 part of cupric sulphate and 1 part of selenium) and 4 ml of salicylsulphuric mixture (0.1g of salicylic acid. 1.0g of sodium thiosulphate and 30 ml of concentrated sulphuric acid) were introduced into the flask. The contents were then slowly heated till frothing ceased and then heated strongly. Completion of digestion was indicated by the solution turning bluish-green. After cooling, about 15ml of distilled water was added to flask, swirled and cooled. The contents were transferred into the distillation unit and 25 ml of 40% sodium hydroxide was added. The ammonia was steam distilled into an excess of 0.1 N sulphuric acid (10.0 ml) containing 2 drops of methyl red. Distillation was continued for 15 minutes. The contents were back titrated using 0.1 N potassium hydroxide till the appearance of golden yellow colour. Nitrogen in the sample was then calculated using the factor, 1 ml of 0.1 NH_2SO4 = 0.0014g of nitrogen.

Phosphorus (P) (Jackson and Williams, 1958)

The oven dried plant material weighing 0.5 g was taken in a 50 ml micro kjeldhal flask and digested in 10 ml of diacid mixture (5: 2 nitric acid and perchloric acid). When the digestion was completed, the contents were cooled and then 30ml of hot water was added. The flask was thoroughly shaken for the complete dissolution of the contents and filtered through Whatman No.44 filter paper, and filtrate was collected in a 100ml volumetric flask. The digestion flask and the silica residue were washed several times with small quantity of warm dilute HNO3 (1:19) and washing was completed with hot water to raise the volume of filtrate to 100ml.

An aliquot of 10ml was taken in a 50ml volumetric flask and the amount of phosphorus in the aliquot was determined calorimetrically by the vando molybdate method. To this 10ml aliquot, a few drops of 1% 2, 4 - dinitrophenol indicator were added followed by 4N Na2CO3 to obtain yellow colour which was later discharged by the addition of 6 N HKO3 to bring out a pH of 3. One ml of 1.2 N HC1 and 2ml of 6.0 N HNO3 were then added and diluted with distilled water to raise the volume to 40ml. Immediately, 2.5 ml each of 0.25% ammonium metavandate and 5% ammonium molybdate were added and shaken. The volume was again raised to 50ml with the addition of distilled water and the contents were allowed to stand for 30 minutes. Readings were taken in a Klett Summerson colorimeter using a blue filter. Known concentration of P solution was simultaneously developed and read in the colorimeter. The standard graph drawn with the help of working standards was used for calculating the P content of the plant tissue.

Potassium (K) (Williams and Twine, 1960)

The powdered moisture free plant material was accurately weighed in 0.2 g quantity in a dry conical flask and 10ml of diacid mixture (5.2 of nitric acid and perchloric acid) was added. The contents of conical flask were allowed to stand for a few hours for cold digestion. The conical flasks were then kept on a hot plate and the contents were digested by slowly increasing the temperature. The digestion was continued till the contents became colourless. The digested material was filtered through Whatman No.40 filter paper by repeatedly washing the conical flask with a small volume of distilled water. The filtrate collected was made up to a suitable volume. The filtrate thus obtained was suitably diluted and fed into flame photometer. The standard graph drawn with the help of working standards was used for calculation of K content of the plant tissue.

Extraction method (Williams and Twine, 1960)

The oven dried plant material was accurately weighed in 0.2 g quantity in a dry conical flask and 10ml of diacid mixture (5.2 of nitric acid and perchloric acid) were added. The contents of conical flask were allowed to stand for a few hours for cold digestion. The conical flasks were then kept on a hot plate and the contents were digested by slowly increasing the temperature. The digestion was continued till the contents became colourless. The digested material was filtered through Whatman No.40 filter paper by repeatedly washing the conical flask with a small volume of distilled water. The filtrate collected was made up to a suitable volume. The filtrate thus obtained was suitably diluted and fed into ICP spectrometer (JOB1NYVON, Model JY24). The Mg, Mn, Cu, Fe, and Zn were analyzed.

3.RESULTS

The results of present investigation are given in Table 1.

Growth parameters	Control treatment	Biofertilizer treatment *Oscillatoria formosa*	Recommend level Chemical fertilizer treatment
Seed germination %	90	95	95
Root length (cm) 25 th Day	9	10	10
Shoot length (cm) 25 th Day	12	18	17
Number of lateral roots	4	8	8
Number of leaves	4	6	6
Total chlorophyll content (mg/g)	0.8768	1.1522	1.1516
Chlorophyll 'a'(mg/g)	0.856	1.756	1.754
Chlorophyll 'b'(mg/g)	0.241	0.442	0.443
Macro nutrients (Root system)			
Total nitrogen (ppm)	412.2	1561	1560
Phosphorus (ppm)	241.1	274.5	274.2
Potassium (ppm)	132.8	389	388
Micro nutrients (Root system)			
Manganese (ppm)	84	122	121
Magnesium (ppm)	0.28	0.60	0.61
Zinc (ppm)	0.101	0.135	0.134
Copper (ppm)	1.20	2.25	2.25
Iron (ppm)	1.02	2.21	2.20
Macro nutrients (Shoot system)			
Total nitrogen (ppm)	412	1659	1658
Phosphorus (ppm)	240.3	285.6	285.4
Potassium (ppm)	135.2	442	442
Micro nutrients (Shoot system)			
Manganese (ppm)	82	121	121.2
Magnesium (ppm)	0.28	0.040	0.041
Zinc (ppm)	0.101	0.132	0.132
Copper (ppm)	0.078	0.124	0.124
Iron (ppm)	2.10	3.01	3.02

Growth parameters

The 95% seed germination rate was recorded in biofertilizer and chemical fertilizer treated batches and 90% was observed in control batch. The root lengths 9, 10, and 10cm were observed in control, biofertilizer and chemical fertilizer applied plants respectively. Likewise, shoot lengths 12, 18 and 17cm were recorded. The lateral roots were 4, 8 and 8 numbers recorded. The leaves 4, 6 and 6 were observed. The total chlorophyll content were 0.8768, 1.1522 and 1.1516mg/g.

Chlorophyll 'a'

The chlorophyll 'a' were 0.856, 1.756 and 1.754mg/g.

Chlorophyll 'b'

The observed chlorophyll 'b' were 0.241, 0.442 and 0.443mg/g.

Macro nutrients in root system

The macro nutrients such as nitrogen, phosphorus and potassium were recorded in root system of control, biofertilizer and chemical fertilizer treated plant respectively. The nitrogen content were 412.2 , 1561 and 1560ppm. The recorded phosphorus content were 241.1, 274.5 and 274.2ppm. The potassium content were 132.8, 398 and 398ppm.

Micro nutrients in root system

The micronutrients such as manganese, magnesium, zinc, copper and iron were observed in root system of control, biofertilizer and chemical fertilizer treated plants respectively. The manganese content recorded were 84, 122 and 121ppm. The observed magnesium was 0.28, .60 and .061ppm. The zinc content were recorded as 0.101, 0.134 and 0.135ppm. The recorded copper content were 1.20, 2.25 and 2.25ppm. The observed iron content were 1.02, 2.21 and 2.20ppm.

Macro nutrients in shoot system

The macro nutrients such as nitrogen, phosphorus and potassium were recorded in shoot system of control, biofertilizer and chemical fertilizer treated plants respectively. The observed nitrogen content were 615, 1659 and 1658ppm. The recorded phosphorus content were 240.3, 285.6 and 285.4ppm. The potassium content were 135.2, 442 and 442ppm.

Micro nutrients in shoot system

The micronutrients such as manganese, magnesium, zinc, copper and iron were observed in shoot system of control, biofertilizer and chemical fertilizer treated plants respectively. The observed manganese content were 82, 121 and 121.2ppm. The magnesium were recorded as 0.28, .0.040 and 0.041ppm. The zinc content were observed as 0.101, 0.132 and 0.132ppm. The recorded copper content were 0.078, 0.124 and 0.124ppm. The iron content were observed 2.10, 3.01 and 3.02ppm.

4.DISCUSSION

Cyanobacteria dominate a wide range of diverse environments characterized by extremes of temperature, desiccation, pH, salinity, light intensity and nutrients (Whitton, 2000). Many cyanobacteria tolerate high levels of ultraviolet irradiation (Sinha *et al*, 1999), permitting them to survive at the soil surface. Cyanobacteria biofertilizers have several advantages over chemical fertilizers. They are non-polluting, in-expensive, utilize renewable resources. In addition to their ability of using free available solar energy, atmospheric nitrogen and water. Besides supplying N2 to crops, they also supply other nutrients such as vitamins and growth substances (Wagner, 1997). Recently, the soil algalization technique has received an increasing attention as it reduces environmental pollution, improves soil fertility and reduces the shortage and cost of chemical fertilizers (Roger and Kulasooria, 1980). The present data showed that

the cyanobacteria O*scillatoria formosa* soil conditioners caused an increase in the percentage of germination of tomato seeds over the control. The growth and biochemical uptake rate was the O*scillatoria formosa* suspension treatment which recorded values more than control and more or less similar to reccommented level chemical fertilizer treatment. The similar results were reported by Soad humead alialkhiat, 2006 in *Nostoc* suspension on tomato plant. The use of cyanobacteria as nitrogen-based biofertilizers are reported in many rice growing countries of the world. This was because of the increased cost of chemical fertilizers that cause soil and water pollution, changes soil structure and produce microflora. In comparison, cyanobacteria is a cheap source of N, which does not cause pollution. It improves the organic matter status and water holding capacity. Sutton *et al* (1991) observed that the use of nitrogenous fertilizers cause acidification of soils, and their long-term application significantly reduces the microbial activity of the soil. The present study was conducted to evaluate the effect of cyanobacteria as biofertilizers alone and with other soil conditioners on growth and biochemical constituents of *Lycopersicon sculentum* seedlings during germination and vegetative stages. The effect of cyanobacterial suspensions and culture filtrates were also tested. The *Nostoc* culture suspension and filtrate induced higher percentage germination and GRI over any other investigated treatments. Seed germination, seedling growth, nutrient uptake, yield and water efficiency were increased when hydrogels were added to sandy soils (Abanemy, 2001; El-Hady *et al*, 2003).

5.CONCLUSION

The current study was performed to assess the effect of nitrogen fixing cyanobacteria to improve the natural poor sandy soil. The data presented revealed the beneficial use of cyanobacteria in comparison with the control (untreated soil) without any consideration to other treatments employed in this study. In this findings, as both biofertilizer and recommended level chemical fertilizer treated plants were gave more are less similar outputs. It can be concluded it is advisable to use of biofertilizer as it is eco- friendly and environmentally safe.

6.REFERENCES

Abanemy, M.B.A., *Physiological effect of soil conditioner (Aquasorb) on wheat plant*. (M. Sc. Thesis. Faculty of science. King Saud University). 2001.

Bremner, J.M.,. Determination of nitrogen by Kjeldhal method. *J.Agric. Sic*., 55 (1960) 11-13.

Desikachary, T.V. , *Cyanophyta. Monograph*. I.C.A.R. New Delhi. India, 1959:

El-Hady, O.A.; Tayel, M.Y. and Lotfy, A.A., Super gel as a soil conditioner II. Its effect on plant growth, enzymes activity, water use efficiency and nutrient uptake. *Egypt. J. Soil Sci., Special Issue*: (1981) 105-106.

Jackson, W.A and D.c. Williams, Nitrate stimulated uptake and transport of strontium and other cations. *Soil Sci.Soc. Am. Proc*., 32, 1958, 689-704.

Prescott, G.W., Algae of the western great lakes area, pub. (Otto koellz science publishers, Koenigstein), 1951.

Rippka, R.; Deruelles, J.; Waterbury, J.B., Herdman, M.; and Stanier, R.Y. Generic assignments, strain histories and properties of pure cultures of cyanobacteria taxonomy redefinition of the blue-green algae to conform to the bacteriological code. *J. Gen. Microbial*. 111 (1979) 1-16.

Roger, P.A. and Kulasooriya, S.A., Blue green algae and rice. (International Rice Research Institute, Los Banos, Philippines), 1980.

Santra, S.c., Biology of rice fields blue green algae.(Daya publishing house. New Delhi), 1993.

Sinha, R.P.; klisch, M.; Vaishampayan, A. and Hader, D.P, Biochemical and spectroscopic characterization of the cyanobacterium *Lyngbya sp*. inhabiting mango (*Mangifera indica*) trees: Presence of an ultravioletabsorbing pigment, Scytonemin. *Acta protozool*. 38 (1999) 291-298.

Sutton, S. D.; Barrett, G.W. and Taylor, D. H., Microbial metabolic activities in soils of old-field communities following eleven years of nutrient enrichment. *Environm. Pollut*. 73 (1991) 1-10.

Venkataraman, G.S., *All India-Co-ordinated project on algae*. Annual report (1980-81). Division of Microbiology, (Indian Agricultural Research Institute), New Delhi, India, 1981.

Wagner, G.M. *Azolla*: A review of its biology and utilization. *The Botanical Review*. 63 (1) (1997) 1-26.

Wehr,J.D. and R.G. Sheath., *Fresh water algae of North America- Ecology and classification*, Acadamic press, California,USA. 2003

Whitton, B.A. Soils and rice-fields. (B.A.Whitton and M. Potts .,eds.). The ecology of cyanobacteria: Their diversity in time and space. (Kluwer Academic,Netherlands). 2000, 233-255.

Williams and Twine ,. Modern methods of plants analysis, In: Vol.V. (Eds.). K.Peach and M.V. Tracey, Springer Verlag, Berlin, 1960, 3-5.

*****