



Effect of Liquid Biofertilizers on Growth and Yield of *Vigna mungo* L.

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ABSTRACT

Liquid biofertilizer is increasingly available in the market as one of the alternatives to chemical fertilizer and pesticide. The present paper deals with the effectiveness of the growth of *Vigna mungo* L. using liquid biofertilizers, such as *Azospirillum*, *Rhizobium* *Azotobacter* with control. The soil sample was subjected to Tamil Nadu Government Agriculture Department, Mobile Test Centre, Thiruvarur. The physicochemical parameters such as pH 6.4, carbon (0.60), Potassium (39), Phosphorus (84.25), were tested before and after treatment. Shelf life of liquid biofertilizer is maintained. The seedlings of *Vigna mungo* were transplanted in 8 pots of equal size, which were noted as Treatment (T1-T8). The seedlings of pot were treated with liquid biofertilizers. The un-inoculated pots were denoted as control. Liquid biofertilizer was sprayed on plants at 10 days intervals. Then morphological parameters such as leaves number, height, shoot length, root length, and biochemical constituents such as chlorophyll, carbohydrate, protein and carotenoids content were analyzed at different time intervals (30th, 45th, 60th days) respectively. Compare to all pots the combined inoculation of liquid biofertilizers (T7) *Azospirillum* + *Rhizobium* + *Azotobacter* in 60th day showed better response in all the parameters was tested. To prevent the environment pollution from extensive application of chemical fertilizers the liquid biofertilizer could be recommended to farmers to insure the public health and a sustainable agriculture.

Keywords: *Azospirillum*, *Azotobacter*, *Rhizobium*, *Vigna mungo* Liquid biofertilizers.

INTRODUCTION

India is an agricultural based country. In order to feed the ever growing populations, India has to increase the per unit area productivity. According to united Nations Food and Agriculture Organization (FAO) estimations, the average demand for the agricultural commodities will be 60% higher in 2030 than present time and more than 85% of this additional demand will be from developing countries, for over half a century, the world has relied on the concept of increasing crop yields to supply an ever increasing demand of food. Therefore, vertical expansion of food production is necessary.¹ In order to increase the unit area productivity of agricultural land, the role of different crop nutrients in contributing increased crop yields is vital. Among the crop nutrients, nitrogen as well as phosphorus plays an important role in increasing the productivity.

Biofertilizers are one of the best modern tools for agriculture. It is a gift of our modern agricultural science. Biofertilizers are applied in the agricultural field as a replacement of our conventional fertilizers contains compost, household wastes and green manure. Those are not as effective as chemical fertilizers, so farmers often try to use chemical fertilizers in the agricultural field for crop development. But obviously the chemical fertilizers are not environment friendly. Biofertilizers contains microorganisms which promote the adequate supply of nutrients to the host plants to ensure their proper development of growth and regulation in their physiology. Living microorganisms are used in the preparation of biofertilizers.²

Liquid biofertilizer formulation is the promising and updated technology which inspite of many advantages over the agrochemicals left a considerable dispute among the farmer community in terms of several reasons major being the viability of the organisms. Shelf life is the first and foremost problem of the carrier based biofertilizer which is up to 3 months and it does not retain throughout the crop cycle. LBF on the other hand facilities the long survival of the organisms by providing the suitable medium which is sufficient for the entire crop cycle.³

Liquid biofertilizer is increasingly available in the market as one of the alternatives to chemical fertilizer and pesticide. One of the benefits from biofertilizer is a contribution from population of microorganisms available. Traditionally liquid biofertilizer produced from fermentation of effective microorganisms (EM) was recommended to be used within three months. Nowadays, the production of ready to use liquid biofertilizer from EM is becoming available in market. A preparation comprising requirements to preserve organisms and deliver them to the target regions to improve their biological activity. The black gram combined the treatment of *Azospirillum* to perform significant improvement in plant productivity and quality.⁴ The maximum germination percentage, fresh and dry weight, number of pods per plant, seed yield per plant were increased with the biofertilizer inoculants. Therefore an experiment was planned to study the influence of liquid biofertilizer on growth, yield and associated protein profiling changes in black gram.⁵



MATERIALS AND METHODS

Sample collection

Soil sample were collected from Vaduvur in Thiruvarur District, Tamil Nadu, South India, using sterile polythene bag. Liquid biofertilizers were purchased from Tari Biotech Thanjavur.

Soil Analysis

Collected soil sample was subjected to Tamil Nadu Government Agriculture Department, Mobile Test Centre, Thiruvarur. The physicochemical parameters such as pH 6.4, carbon (0.60), Potassium (39), Phosphorus (84.25), were tested for before and after treatment.

Test of crop plant

The test plant was selected for the present study i.e. *Vigna mungo*, third important pulse crop in India. It is annual pulse crop and native to central Asia.

Seeds

Viable seeds were obtained from Tamil Nadu Rice Research Institute, Aduthurai, care was taken in selecting the seeds of uniform size and they were stored in polythene bags, containing sterile soil samples. The seeds were mixed with liquid biofertilizers to 12hrs. Then the sowing seeds were soaked in particular doses. Then the seeds were sowed and observed for germination and early growth.

Pot culture

Vigna mungo seeds were sown in 8 pots (8seeds/pots). The seedlings of *Vigna mungo* were transplanted in eight pots of equal size, 30cm, in height and 6cm in dm. 5kg of soil was used, the pots were provided with water facilities, control was also maintained without biofertilizer. All the experiments were conducted in triplicates.

Physical features of liquid biofertilizer

Liquid biofertilizer are dull white in colour, pH 6.8- 7.5 no bad smell, and no foam formation. All liquid biofertilizer have shelf life for 6 months, microbial load was tested for every 10 days of interval at the time of foliar spray.

Treatment

There were eight pots used for the treatment. The pots were maintained in the open shade at the temperature of 27°C- 30°C. The crop plant *Vigna mungo* was treated with liquid biofertilizers.

T1 - *Azospirillum*, T2- *Rhizobium*, T3- *Azotobacter*, T4- *Rhizobium*+ *Azospirillum*, T5- *Azospirillum*+ *Azotobacter*, T6- *Rhizobium*+ *Azotobacter*, T7- *Rhizobium*+ *Azospirillum*+ *Azotobacter*, C- Control.

Morphological parameters

After 30th, 45th, 60th days of growth, the plants per pot were removed from all samples, and studied for the

following, morphological parameters, They were, height of the plant (in cm), Number of leaves (per plant), Number of roots (per plant), Shoot length (in cm), Root length (in cm), Root nodules (per plant).

Biochemical constituents

Carbohydrate, Protein, Chlorophyll, Total Chlorophyll, Carotenoids were also estimated.⁶⁻⁹

Statistical Analysis

All the experiments were repeated as triplicates. The results obtained in the present investigation were subjected to statistical analysis like mean and standard deviation.¹⁰

RESULTS

The effect of liquid biofertilizer treatment on vegetative growth of blackgram was significantly higher in single or combined inoculation than control plants. A significant variation in plant height and number of leaves due to application of biofertilizers was found. Statistically significant increase in plant height, number of leaves, Shoot length, Root length, number of roots, number of nodules. The soil sample was subjected to Tamil Nadu Government Agriculture Department, Mobile Test Centre, Thiruvarur. The physicochemical parameters such as pH 6.4, carbon (0.60), Potassium (39), Phosphorus (84.25), were tested before inoculation and the above said parameters were increased after liquid biofertilizer inoculation treatments.

Table 1: Soil analysis before and after treatment

Carbon (%)	Available Potassium (ppm)	Available Phosphorus (ppm)	pH
0.60	39	84.25	6.4
0.87	56	87.5	6.8

Impact of liquid biofertilizers on the growth of *Vigna mungo L.* was studied compared with control. The seed inoculation with liquid biofertilizer was planned as (T1) - *Azospirillum*, (T2) *Rhizobium* (T3) *Azotobacter*, (T4) *Rhizobium* + *Azospirillum*, (T5) *Azospirillum* + *Azotobacter*, (T6) *Rhizobium*+ *Azotobacter* (T7) *Rhizobium* + *Azospirillum* + *Azotobacter*, and (T8)-Control. The treatments were significantly inoculated in plant growth and yield of *Vigna mungo*. The number of leaves, height of plant, leaf area (length and breadth), shoot length, root length, number of roots, number of nodules, was measured at 60th days, after sowing, among the overall treatments, (T7) was showed better response for combined inoculations than the other treatments and control. The biochemical parameters such as chlorophyll, protein, carbohydrate, carotenoids were also increased in treatment with combined inoculation of liquid biofertilizers (*Rhizobium*+*Azospirillum*+*Azotobacter*) in the treatment T7 was showed better response than the other treatments and control.



Table 2: Impact of liquid biofertilizers on morphological parameters in *Vigna mungo* L. (60th day)

Treatment	Height of the plant(cm)	Number of leaves/ plant	Shoot length(cm)	Root length (cm)	Number of roots/ plant	Number of nodules/plant
T1	25.2±2.57	23.7± 6.51	13.6± 2.57	12.8± 0.36	14.8± 0.41	11.9±1.52
T2	24.7±1.52	26.4± 0.2	15.6± 6.74	13.8± 0.71	11.8± 0.66	13.5± 2.15
T3	21.4± 3.39	24±1.52	11.7± 2.02	12.1 ± 0.84	19.4± 1.56	14.8± 0.56
T4	23.7± 2.98	25.7± 6.51	12.6± 2.57	12.8± 1.6	15± 0.49	12.6± 0.49
T5	27.4± 1.82	24.7±0.56	12.1± 2.18	11.3± 2.6	17.3± 1.52	11.7 ± 0.96
T6	26.3± 3.52	22.5 ±1.80	14.9 ± 1.04	13.4± 1.82	18.5± 0.41	13.8±3.12
T7	31.6± 1.52	27.6±3.2	20.5± 03.2	15.6± 0.8	21.8± 0.30	15.4± 0.20
C	19.5± 2.32	20.6±2.64	9.4± 1.1	8.7± 1.2	10.8±0.26	7.5±1.50

Values are triplicates, mean± standard deviation

T1 - *Azospirillum*, T2- *Rhizobium*, T3- *Azotobacter*, T4- *Rhizobium*+ *Azospirillum*, T5- *Azospirillum*+ *Azotobacter*, T6- *Rhizobium*+ *Azotobacter*, T7- *Rhizobium*+ *Azospirillum*+ *Azotobacter*, C- Control.

Table 3: Biochemical constituents in *Vigna mungo* L. (60th day)

Treatment	Chlorophyll a	Chlorophyll b	Total chlorophyll	Carbohydrate (mg)	Carotenoids (mg)	Protein (µg/g)
T1	0.453±0.43	0.0389± 0.86	0.0452 ±012	4.45±0.49	0.389± 0.86	3.354 ±0.041
T2	0.497± 0.034	0.0423± 0.56	0.0490 ±0.089	4.69± 0.034	0.423± 0.56	3.367 ±0.040
T3	0.510± 0.030	0.0484± 0.049	0.0521± 0.423	5.45± 0.030	0.484± 0.049	3.421± 0.087
T4	0.0534± 0.02	0.523± 0.11	0.0578± 0.345	5.34± 0.023	0.523± 0.11	3.478± 0.067
T5	0.0623± 0.30	0.0576± 0.098	0.0549± 0.076	5.623± 0.30	0.576± 0.098	3.548± 0.023
T6	0.598± 0.089	0.0521± 0.078	0.0567± 0.078	6.598± 0.089	0.521± 0.078	4.587± 0.034
T7	0.643± 0.56	0.0598 ±0.067	0.0678± 0.087	6.643± 0.56	0.598 ±0.067	4.620± 0.057
C	0.0321± 0.09	0.0311 ±0.076	0.412± 0.56	0.0321± 0.09	0.311 ±0.076	4.265± 0.051

Values are triplicates, mean± standard deviation

The seed inoculation with biofertilizers like *Rhizobium*, *Azospirillum* and Phosphobacteria at various treatments was significantly increased in plant growth and yield of green gram. The number of leaves, leaf area, shoot length, root length number of nodules were significantly more with their combined treatment. The yield concepts such as number of flowers and number of pods were increased in the combined treatments with *Rhizobium*, *Azospirillum* and Phosphobacteria, inoculated plants of green gram.¹¹ The fruit yield have been affected by the nitrogen fertilizer The maximum fruit yield was observed in combined inoculation *Azotobacter* treated plants had higher fruit yield in compared of *Azospirillum* and control. *Azotobacter* enhanced the available nitrogen in the soil which could enhance the yield in safflower.¹²

Height of the plant

Utilization of liquid biofertilizer in combined inoculation of treatments such as *Rhizobium* + *Azospirillum* + *Azotobacter* (T7) in 60th day was the best response (31.6±1.52cm). Followed by other treatments and control. In 60th day liquid biofertilizers *Azospirillum* + *Azotobacter* (T5) was agreed with above said response in height of plants (27.4 ± 1.82cm) followed by other treatments and control (Table 2).

Number of leaves

Number of leaves was increased in combined inoculation of liquid biofertilizer treatments such as *Rhizobium* + *Azospirillum* + *Azotobacter* (T7) in 60th day showed maximum response in leaves of plants (27.6±3.2) followed by other treatments and control. In 60th day biofertilizer alone *Rhizobium* (T2) was effective in leaves of plants (26.4 ± 0.2) followed by *Azospirillum*, *Azotobacter* and control (Table 2).

Shoot length

Combined inoculation of liquid biofertilizer treatments such as *Rhizobium* + *Azospirillum* + *Azotobacter* (T7) in 60th day showed higher response in shoot length of plants (20.5±03.2cm) followed by other treatments and control. In 60th day liquid biofertilizer alone *Rhizobium* (T2) was significant increase response in shoot length of plants (15.6±6.74cm) followed by *Azospirillum*, *Azotobacter* and control (Table 2).

Root length

The maximum root length was recorded in combined inoculation of liquid biofertilizer treatments such as *Rhizobium* + *Azospirillum* + *Azotobacter* (T7) in 60th day was 15.6±0.8cm followed by other treatments and control. Average root length liquid biofertilizer alone



Rhizobium (T2) was observed (13.8±0.71cm) followed by *Azospirillum*, *Azotobacter* and control (Table 2).

Number of roots

The efficacies in combined inoculation of three liquid biofertilizer treatments such as *Rhizobium* + *Azospirillum* + *Azotobacter* (T7) in 60th day was recorded in number of roots in plants (21.8±0.30) followed by other treatments and control. In 60th day liquid biofertilizer alone *Azotobacter* (T3) was exhibited better response in number of roots in plants (19.4±1.56) followed by *Azospirillum*, *Rhizobium* and control (Table 2).

Root nodules

Combined inoculation of liquid biofertilizer treatments such as *Rhizobium* + *Azospirillum* + *Azotobacter* (T7) in 60th day was maximum response in number of root nodules in plants (15.4±0.20) followed by other treatments and control. In 60th day liquid biofertilizer alone *Azotobacter* (T3) was noticed in number of root nodules in plants (14.8±0.56) followed by *Azospirillum*, *Rhizobium* and control (Table 2).

The biochemical contents such as Chlorophyll, Carbohydrate, Protein, Carotenoids were estimated in 60th day treatment for combined inoculation. (Table 3).

The biochemical contents such as chlorophyll and protein content were determined to find out variation in these single and dual inoculations of tomato and control. Results showed that *Azotobacter* and *Azospirillum* treated plants had the highest chlorophyll and protein contents. Similar results were reported by the biofertilizer significantly improved chlorophyll concentration in black gram.¹³

The result showed that *Azotobacter* and *Azospirillum* treated plants had the highest chlorophyll and protein contents. Similarly results were reported by the biofertilizer significantly improved chlorophyll concentration in chilli and black gram.¹⁴ Our findings were coincide with Murugesan (2008), Ramakrishnan (2012), overall utilization of biofertilizers with single and combined treatments in addition to increased yield could be a strategy to achieve sustainable agriculture.¹⁵⁻¹⁶

CONCLUSION

Hence our study was clearly highlighted that combined inoculation of liquid biofertilizers such as *Rhizobium*, *Azospirillum*, *Azotobacter* could enhance the morphological parameters such as height of the plant, number of leaves, Shoot length, Root length, number of roots, root nodules, and biochemical constituents such as Chlorophyll, Carbohydrate, Protein Carotenoids. Compared to individual inoculation and control, could be the collective effect of liquid biofertilizer, and also reduce the use of chemical fertilizers. Inoculation with plant

growth promoting rhizobacteria (PGPR) may enhance crop productivity either by making the other nutrients available or protecting plants from pathogenic microorganisms. To combat the threat of global food crises the alternative technologies in the agriculture like liquid biofertilizers are obligatory. Liquid biofertilizer of course have the capacity to replace the traditional chemical fertilizers and carrier based biofertilizers and play a major role in restoring the soil health, but a lot of measures in terms of technology, government support, subsidies, and constructive awareness by well trained technicians among the agrarians are emphasized.

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