



Effect of Water-Soluble Fertilizers and PGPR on Soil Microbial Population, Nodule Count and Economics of Black Gram

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Abstract: Field experiment was conducted on effect of water-soluble fertilizers and PGPR application on soil microbial population, nodule count and economics were studied under rainfed condition during *Kharif*-2019 at the College of Agriculture, Shivamogga. The field experiment was laid out in randomized complete block design and replicated thrice with thirteen treatments consisting of different combinations of 19:19:19 and monopotassium phosphate with or without PGPR application. Treatments significantly differ through foliar application of water-soluble fertilizers and PGPR application. The foliar application of 19:19:19 and monopotassium phosphate (0:52:34) @ 1% each at 30 and 45 days after sowing + PGPR along with a package of practice recorded significantly higher number of nodule count (16 and 31 at 30 and 45 DAS). The higher microbial count such as N fixers (53.73, 57.31 and 47.02), P-solubilizers (35.25, 40.32 and 35.58) and K-solubilizers (25.03, 31.18 and 26.30) $\times 10^5$ colony forming units (CFU) g^{-1} soil at 30, 60 DAS and at harvest respectively over the package of practice.

Keywords: Blackgram, Nodules, PGPR, Rhizosphere, Water soluble fertilizers

Among the grain legumes, blackgram (*Vigna mungo* L.) commonly called urdbean is an ancient and well-known leguminous crop of Asia. Though it is cultivated in every part of Asia, Africa and the West Indies, it is not much important in other countries as compared to India. Besides, it is adapted to a wide range of agro-climatic conditions because of its morphological parameters perfectly suiting for intercropping and sole cropping systems. It is extensively grown as a grain legume and guaranteed considerable importance from the point of food and nutritional security as it contains 24% protein and 60% carbohydrates with calorific value of 347 Kcal per 100 g. In India, black gram contributes about 13% of total pulse area and 10 % of their total production and is cultivated over an area of about 4.6 M ha with a production of 3.56 Mt. with a productivity of 654 kg/ha. In Karnataka, it is cultivated over an area of 1.38 lakh ha with a production of 0.47 lakh tonnes (Anonymous 2018). Nowadays soil application of mineral fertilizers is not enough to meet the nutrient demand of the crop due to various losses like leaching, volatilization, fixing on the soil and also due to soil physical and chemical properties. This result in various deficiency symptoms which effect on the plant vegetative growth and a nutrient deficiency during reproductive stages increase the flower drop and reduce the fruit set. This

evidence creates preconditions to increase the importance of foliar feeding as an alternative to meet plant nutrient demand during the growing season. Foliar feeding is the most effective and economical way to overcome plant nutrient deficiency (Dixit and Elamathi 2007). Besides foliar nutrition application of PGPR to soil increases the nutrient retentive capacity, mobilizes the fixed nutrients and increases the soil biological activity by nodule formation through a symbiotic association with plant roots thus increasing the availability of nutrients. Under rainfed conditions, when the availability of moisture becomes scarce, the application of fertilizers as a foliar spray along with PGPR to soil results in efficient absorption and usage.

Soil microorganisms play an important role in nutrient cycling and are important factors of soil health and fertility. Plant growth-promoting rhizobacteria are a group of helpful soil bacteria that colonise roots and their environs (rhizosphere) (PGPR). They have a substantial impact on crop growth and development by forming symbiotic, associative or neutral relationships with plants. Plant development is stimulated by nutrient mobilisation, solubilization and transformation and plants are protected against pathogenic infections by PGPRs. At several stages of the plant life cycle, PGPR boost growth through direct or

indirect methods. Nitrogen fixation, phosphate solubilization, phytohormone synthesis and mineral availability increase are examples of direct processes. The nitrogen fixation process is aided by an oxygen-sensitive enzyme complex called nitrogenase, which converts atmospheric free nitrogen into an ammonical form (biologically fixed nitrogen) that can be used by plants or released into the soil. Phosphate solubilizers convert fixed forms of phosphorus in the soil into a form that the plant can use. Plant hormones like auxins, gibberellins and cytokinin have a role in plant growth as well. The above field experiment is carried out to determine the effect of water-soluble fertilizers and PGPR on soil microbial activity, nodule count and to work out the monetary viability of different treatments.

MATERIAL AND METHODS

Field experiment was conducted during *Kharif* season of 2019 at University of Agricultural and Horticultural Sciences, Shivamogga which is come under Southern Transition Zone (Zone-7) of Karnataka. The geographical reference point of the experimental site was 13° 58' to 14° 1' North latitude and 75° 34' to 75° 42' East longitude and at an altitude of 650 m above the mean sea level. The soil was sandy loam in texture, slightly acidic pH (6.19) and normal in electrical conductivity (0.70 dS/m), low organic carbon 4.56 g/kg and low in available nitrogen (242.22 kg/ha), high in phosphorus (P₂O₅) (75.08 kg/ha) and medium in potassium (K₂O) status (135.63 kg/ha) and Nitrogen fixers (20×10⁵ CFU/g of soil), P-

solubilizing bacteria (PSB) (16×10⁵ CFU/g of soil), K-mobilizing bacteria (9×10⁵ CFU/g of soil). During the cropping period, the total actual rainfall received was 1088.8mm. Randomized complete block design was used for this experiment with thirteen treatments and three replications. Treatments consisting of different combinations of water soluble fertilizers viz., 19:19:19 (N: P: K) and mono potassium phosphate (0:52:34) sprayed at 30 and 45 days after sowing with or without liquid plant growth promoting rhizomicrobial consortia application along with the package of practice (Table 1). Variety used was Rashmi (LBG-625) which matures in 85 to 90 days with average yield ranging from 8 to 9 q ha⁻¹. The plots of 3.6 m width and 3.0 m length were prepared by making bunds of 50 cm width and 30 cm height. The gross plot size was 3.60m×3.0 m and net plot size was 2.40m×2.20m. Recommended dose of fertilizers and farm yard manure @ 7.5 t/ha was applied at the time of sowing common to all the treatments. Liquid plant growth promoting rhizomicrobial consortia (*Rhizobium leguminosarum*, *Pseudomonas* sp. and *Bacillus* sp.) mixed with farm yard manure at the rate of 750 ml/ha incorporated to soil as per treatments at the time of sowing. The 1% solution of water-soluble fertilizers 19:19:19 and mono potassium phosphate was used for foliar spraying at 30 and 45 days after sowing as per the treatments. The number of nodules per plant was counted manually by uprooting roots of five plants at optimum soil moisture and soaking with water to wash adhered soil. Cost of cultivation and returns of crop is

Table 1. Effect of water-soluble fertilizers and PGPR on nitrogen fixers, phosphorous solubilizers and potassium solubilizer

| Treatments details | N fixers (×10 ⁵ CFU/g of soil) | | | PSB (×10 ⁵ CFU/g of soil) | | | KSB (×10 ⁵ CFU/g of soil) | | |
|---|---|--------|------------|--------------------------------------|--------|------------|--------------------------------------|--------|------------|
| | 30 DAS | 60 DAS | At harvest | 30 DAS | 60 DAS | At harvest | 30 DAS | 60 DAS | At harvest |
| T ₁ : Package of practices | 35.83 | 38.59 | 29.36 | 24.56 | 27.95 | 23.02 | 13.65 | 17.68 | 15.35 |
| T ₂ : T ₁ +19:19:19 @1% at 30 DAS | 37.24 | 40.54 | 31.15 | 25.89 | 28.81 | 25.35 | 15.67 | 19.96 | 17.32 |
| T ₃ : T ₂ + PGPR | 48.40 | 52.59 | 42.32 | 31.15 | 36.51 | 33.36 | 22.68 | 27.65 | 23.35 |
| T ₄ : T ₁ +19:19:19 @1% at 30 and 45 DAS | 38.58 | 41.54 | 34.54 | 25.73 | 29.65 | 26.64 | 16.65 | 20.82 | 18.62 |
| T ₅ : T ₄ + PGPR | 48.58 | 52.79 | 42.17 | 31.81 | 36.81 | 33.65 | 23.02 | 28.27 | 23.32 |
| T ₆ : T ₁ +MPP @1% at 30 DAS | 39.21 | 42.54 | 32.79 | 26.24 | 31.55 | 28.39 | 16.86 | 21.69 | 18.25 |
| T ₇ : T ₆ + PGPR | 48.62 | 53.54 | 42.36 | 32.12 | 37.25 | 33.88 | 23.26 | 27.32 | 24.43 |
| T ₈ : T ₁ +MPP @1% at 30 and 45 DAS | 40.54 | 43.35 | 34.47 | 27.56 | 32.75 | 29.08 | 17.21 | 22.13 | 18.52 |
| T ₉ : T ₈ + PGPR | 48.79 | 54.19 | 43.17 | 33.20 | 37.02 | 34.65 | 23.65 | 30.52 | 24.86 |
| T ₁₀ : T ₁ +19:19:19@1%+ MPP @1% at 30 DAS | 40.99 | 43.62 | 34.58 | 28.62 | 33.18 | 30.82 | 17.00 | 22.65 | 19.35 |
| T ₁₁ : T ₁₀ + PGPR | 51.18 | 55.41 | 44.15 | 34.68 | 38.21 | 34.82 | 23.93 | 30.21 | 25.28 |
| T ₁₂ : T ₁ +19:19:19 @1% + MPP @1% at 30 and 45 DAS | 41.92 | 44.56 | 35.31 | 29.72 | 33.99 | 30.91 | 17.50 | 23.05 | 20.65 |
| T ₁₃ : T ₁₂ + PGPR | 53.73 | 57.31 | 47.02 | 35.25 | 40.32 | 35.58 | 25.03 | 31.18 | 26.30 |
| CD @5% | 9.46 | 5.31 | 8.12 | 4.94 | 5.58 | 3.43 | 4.19 | 4.32 | 3.48 |

*Package of practice (6.5 t ha⁻¹ FYM, 13:25:25 kg NPK ha⁻¹ + 4 kg ZnSO₄ as basal dose)

calculated based on price of inputs during experiment and market price during prevailing year.

Enumeration of microorganism in rhizosphere: The microbial population in the soil at different growth stages and after harvest was determined by serial dilution pour plate method. Soil samples from different treatments were collected separately from each replication and analysed separately. Ten grams of pooled soil (treatment wise) weighed and mixed in 90 ml sterilized water blank to give 10^{-1} dilutions. Subsequent dilutions up to 10^{-6} were made by transferring serially 1 ml of each dilution to 9 ml sterilized water blanks. The population of N-fixers, P-solubilizer's and K- solubilizer's were estimated by pour plate serial dilution method and by taking 1ml from selected dilution of 10^{-5} were transferred aseptically to petri plates and the desired agar media was prepared by using appropriate ingredients and melted by using hot air oven then added to their respective dilutions. Plating on appropriate media viz., Congo red yeast extract mannitol agar (CRYEMA) media, Pikovskaya's media and Alexandrov's agar media, respectively. The inoculated plates were kept for incubation at $30^{\circ}\text{C} \pm 10^{\circ}\text{C}$ for a week time and emerged colonies were enumerated by digital colony counter. Total CFU was calculated.

$$\text{CFU/g of soil} = \frac{\text{Average number of colonies} \times \text{Dilution factor}}{\text{Weight of soil sample (g)}}$$

RESULTS AND DISCUSSION

N-fixers, P-Solubilizers and K-solubilizers: The significantly higher N-fixers population (53.73, 57.31 and 47.02), P-solubilizing bacteria population (35.25, 40.32 and 35.58) and K-solubilizing bacteria population (25.03, 31.18 and 26.30) $\times 10^6$ CFU/g of soil at 30, 60 DAS and at harvest stage, respectively was recorded with the application of $T_1+19:19:19 @1\% + \text{mono potassium phosphate @1\%}$ at 30 and 45 DAS + PGPR which was on par with all the treatments having PGPR application (Table 1). N-fixer's, PSB and K-solubilizing microorganisms population increased from initial observation, 30 and 60 DAS but decrease at the time of crop harvest when compared to peak period of crop growth stage. Mean population of N-fixer's, PSB and K-solubilizers was observed more in PGPR treatments when compared to without PGPR at different growth stages. This increased microbial population might be due to application of microbial consortia along with farmyard manure. Liquid consortia encourage multiplication of N-fixers, PSB and K-solubilizers in the soil up to 60 DAS later at harvest stage of the crop the population decreases due to non-availability of organic matter for their multiplication, reduced root activity and production of root exudates. Luxuriant growth of crop at vegetative stage as reflected by higher dry matter production

resulted in higher microbial population. Application of liquid PGPR may increase the soil microbial population in the rhizosphere by interacting synergistically with native microorganisms (Gupta et al., 2014). Application of monopotassium phosphate and 19:19:19 each @ 1% at 30 and 45 DAS recorded higher microbial population (Bhavya et al 2020).

Number of nodules per plant: The maximum number of nodules per plant at 30 DAS (16.27) and 45 DAS (31.73) was in $T_1+19:19:19 @ 1\% + \text{mono potassium phosphate @ 1\%}$ at 30 and 45 DAS + PGPR (Table 2). More number of nodules per plant was observed in the treatments that received PGPR than other treatments. Increase in the number of nodules might be due to enhanced biological nitrogen fixation due to application of plant growth promoting rhizomicrobial consortia might result in rapid multiplication of *Rhizobium sp.* in the soil. This increased population bacteria which enhance the infection to pulse roots by rhizobium bacteria might led to increased nodulation process and continuous supply of nitrogen through basal dose during initial stage along with farmyard manure and through the foliar spray at later stages of crop growth. Basim and Raghu (2015) observed that application of Farm Yard Manure and PGPR (*Bacillus sp.*, *Pseudomonas sp.* and *Rhizobium sp.*) recorded significantly higher nodule count (72.25). Similar trend was recorded by Nithukumari et al (2019), Yadav and Choudhary (2012) and Gupta et al (2014). Application of PGPR along with foliar

Table 2. Effect of water-soluble fertilizers and PGPR on Seed yield, haulm yield and number of nodules per plant in blackgram

| Treatments details | Seed yield (kg/ha) | Haulm yield (kg/ha) | No. of nodules per | |
|--------------------|--------------------|---------------------|--------------------|--------|
| | | | 30 DAS | 45 DAS |
| T ₁ | 802 | 1603 | 10.47 | 17.47 |
| T ₂ | 869 | 1640 | 11.07 | 18.13 |
| T ₃ | 876 | 1653 | 14.60 | 27.34 |
| T ₄ | 883 | 1708 | 11.73 | 17.90 |
| T ₅ | 965 | 1767 | 15.13 | 27.40 |
| T ₆ | 868 | 1624 | 11.73 | 17.27 |
| T ₇ | 877 | 1657 | 15.63 | 27.53 |
| T ₈ | 886 | 1715 | 11.33 | 17.73 |
| T ₉ | 967 | 1753 | 15.40 | 28.27 |
| T ₁₀ | 955 | 1733 | 12.33 | 18.20 |
| T ₁₁ | 1085 | 1861 | 15.63 | 28.47 |
| T ₁₂ | 994 | 1706 | 13.30 | 22.53 |
| T ₁₃ | 1167 | 2019 | 16.27 | 31.73 |
| CD (p=0.05) | 130.46 | 210.81 | 2.62 | 4.67 |

*See table 1 for treatment details

Table 3. Economics of black gram as influenced by different treatments of water-soluble fertilizers and PGPR

| Treatments details | COC (₹/ha) | Gross returns (₹/ha) | Net returns (₹/ha) | B:C |
|--------------------|------------|----------------------|--------------------|------|
| T ₁ | 24863 | 47366 | 22502 | 1.91 |
| T ₂ | 25988 | 51204 | 25215 | 1.97 |
| T ₃ | 26138 | 51623 | 25484 | 1.97 |
| T ₄ | 27113 | 52087 | 24973 | 1.92 |
| T ₅ | 27263 | 56808 | 29544 | 2.08 |
| T ₆ | 25708 | 51140 | 25431 | 1.99 |
| T ₇ | 25858 | 51699 | 25840 | 2.00 |
| T ₈ | 26703 | 52257 | 25553 | 1.96 |
| T ₉ | 26853 | 56899 | 30045 | 2.12 |
| T ₁₀ | 26433 | 56216 | 29782 | 2.13 |
| T ₁₁ | 26583 | 63736 | 37152 | 2.40 |
| T ₁₂ | 28153 | 58417 | 30264 | 2.07 |
| T ₁₃ | 28283 | 68573 | 40289 | 2.42 |

*See table 1 for treatment details

nutrition of 19:19:19 @ 1% and mono potassium phosphate @ 1% at 30 and 45 DAS resulted in the formation of active and a greater number of nodules (Hamayun and Chaudhary 2014, Hiwale 2015, Kachlam 2017).

Seed yield and haulm yield : The application of 19:19:19 and Mono potassium phosphate each @ 1 % at 30 and 45 DAS + PGPR along with a package of practice treatment recorded significantly higher seed yield (1167 kg/ha) and haulm yield (2019 kg/ha) when compared to other treatments. The increase in yield due to foliar application of fertilizers at 30 and 45 DAS enhances the absorption a rate of photosynthesis which supplies the photosynthates during critical stages such as flowering and pod formation stages and also PGPR application leads to colonization of microorganisms around the roots, it increases the nutrient availability and nutrient uptake results higher yield.

Comparative economics: Different treatments caused variation in cost of cultivation due to water soluble fertilizers and PGPR. However, foliar application of 19:19:19 and mono potassium phosphate each @ 1% at 30 and 45 DAS+PGPR along with package of practice recorded the higher gross returns (68573 ₹/ha) and net returns (40289 ₹/ha) as compared to other treatments. This is attributed by the greater grain and haulm yield in that treatment. The lower gross returns (47366 ₹/ha) and net returns (22502 ₹/ha) were obtained with Package of practice. The maximum benefit cost ratio (2.42) was with foliar application of 19:19:19 and mono potassium phosphate each @ 1% at 30 and 45 DAS + PGPR along with Package of practice and lower benefit cost

ratio (1.91) was in treatment applied with package of practice (Table 3). Jadhav and Kulkarni (2016) also reported that foliar spray of 19:19:19 recorded significantly higher net return (35838 ₹/ha), similar results are also concluded by Mallesha et al (2014); Mudalagiriappa et al (2016).

CONCLUSION

Foliar application of 19:19:19 and Mono potassium phosphate and soil application of liquid PGPR by mixing with farm yard manure improves the soil microbial population viz., N-fixers, P-solubilizer's and K-solubilizer's in the soil during cropping period which ameliorates the nutrient mobility and availability to the crop. It facilitates better nutrient uptake results proper pod setting enhanced the final economical yield of crop fetches more cost benefit ratio.

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