

MICROBIAL INOCULATION INDUCED ENHANCEMENT OF PLANT GROWTH, NODULATION, ENZYME ACTIVITIES, NUTRIENT ACQUISITION, AND PRODUCTIVITY OF BLACKGRAM (*VIGNA MUNGO* L. VBN 8): A MULTIVARIATE PERSPECTIVE

¹Jayapratha C, ²Kumutha K*, ²Gnanachitra M, ³Amutha R, ⁴Gurusamy A, ⁵Kannan P, ⁶Poulomi Mukherjee, ⁷Vijayapriya P

¹Department of Agricultural Microbiology, Agriculture College and Research Institute - Madurai 625103, India

²Department of Agricultural Microbiology, Tamil Nadu Agricultural University, Coimbatore - 641 003, India

³Department of Seed Science and Technology, Agriculture College & Research Institute, Madurai - 625 104, India

⁴Department of Agronomy, Agricultural College and Research Institute, Keezhvelur, 611105, India

⁵Department of Soil Science and Agricultural Chemistry, Agricultural College and Research Institute, Chettinad - 630 102, India

⁶Department of Environmental Biotechnology, Division of Nuclear Agriculture and Biotechnology, BARC, Mumbai

⁷Department of Agricultural Microbiology, Agricultural College and Research Institute, Chettinad - 630 102, India

*Corresponding author: Kumutha K, Email: kkumuthatnau@gmail.com

ABSTRACT

Pulses are integral to Indian agriculture and human nutrition, particularly for the predominantly vegetarian population, owing to their high protein content and capacity for biological nitrogen fixation. Among pulse crops, blackgram (*Vigna mungo* L.) is widely cultivated across diverse agro-climatic regions; however, its productivity remains relatively low due to poor soil fertility, marginal growing conditions, and inadequate nutrient management. The excessive and indiscriminate use of chemical fertilizers has further contributed to soil degradation, nutrient imbalance, and declining nutrient-use efficiency, emphasizing the need for sustainable nutrient management strategies. Biofertilizers offer an eco-friendly and cost-effective approach for improving nutrient availability, soil fertility, and crop productivity while reducing dependence on chemical fertilizers. The present study evaluated twelve elite microbial inoculants for their effects on the growth, nodulation, biochemical and enzyme activities, nutrient uptake, yield, and nutrient-use efficiency of blackgram under field conditions. Field evaluation demonstrated the consistent superiority of *Azospirillum lipoferum* AZ204, *Azospirillum brasilense* CW903, and *Rhizobium leguminosarum* BMBS P47, which markedly improved plant growth, nodulation, grain yield, nutrient uptake, and nutrient-use efficiency compared with uninoculated controls. AZ204 exhibited superior overall performance, particularly in grain yield, nitrogen and phosphorus uptake, and nutrient-use efficiency, whereas CW903 showed pronounced effects on phosphorus related traits and yield attributes. BMBS P47 performed strongly in nodulation, nitrogen uptake, and nitrogen-use efficiency. The cumulative effectiveness of these microbial inoculants was further confirmed by multivariate analysis, which showed that growth, yield, and nutrient-related characteristics significantly contributed to treatment discrimination. These outcomes demonstrate the potential of specific microbial inoculants as viable biofertilizers for blackgram production, particularly AZ204, CW903, BMBS P47, PSB 1, KRB 9 and ZSB 15.

KEYWORDS: Blackgram; microbial inoculants; nutrient use efficiency; nutrient uptake; enzyme activity; sustainable agriculture.

1. INTRODUCTION

Pulses, which comprise the dry, edible seeds of leguminous plants, are highly valued for their culinary versatility and significant nutritional benefits, including high protein content, essential amino acids, and minerals (Zhao *et al.*, 2024). Legumes play a pivotal role in sustainable agriculture by improving soil fertility and health through symbiotic associations with nitrogen-fixing bacteria. Biological nitrogen fixation not only enriches the soil but also reduces dependency on chemical fertilizers. However, intensive cultivation practices, particularly in rice-wheat and rice-rice cropping systems, have led to the depletion of natural resources and declining productivity, threatening long-term sustainability (Majee *et al.*, 2023).

India is one of the world's major producers, consumers, and importers of pulses, contributing approximately 25% of global pulse production, 27% of global consumption, and 14% of global imports. Pulses occupy nearly 20% of the country's food-grain area and contribute about 7-10% of total food-grain production (Kaur *et al.*, 2018). Among the major pulse crops, blackgram (*Vigna mungo* L.), commonly known as urdbean, urd, or mash, is an important food legume cultivated predominantly in South and Southeast Asia. During 2020 -21, blackgram occupied approximately 46.7 lakh hectares in India, with a production of about 23.4 lakh tonnes and an average productivity of 501 kg ha⁻¹, accounting for approximately 15.7% of the total pulse acreage and 9.1% of total pulse production in the country (Agricultural Statistics, 2021). India remains the principal producer and

consumer of blackgram, while the crop is also cultivated in other South Asian countries, including Bangladesh, Nepal, Pakistan and Afghanistan (Kaushik et al., 2023). In view of the increasing demand for pulses and the need to strengthen domestic production, the Government of India launched the Mission for Aatmanirbharta in Pulses for the period 2025-26 to 2030-31, with a target of increasing pulse production to 350 lakh tonnes and expanding the area under pulses to 310 lakh hectares by 2030-31. The mission specifically emphasizes major pulses such as tur, urad, and masoor, along with improved seed availability and assured procurement, highlighting the continued importance of blackgram in achieving India's nutritional and agricultural sustainability goals (Government of India, 2025).

Blackgram is an integral part of cropping systems due to its short duration and its ability to enrich the soil. Nevertheless, productivity is constrained by poor cultural practices, low soil fertility, inadequate fertilization, weed infestation, and susceptibility to pests and diseases, with weeds being particularly limiting. Prolonged use of chemical fertilizers has also contributed to soil degradation and reduced nutrient use efficiency (Saket et al., 2014; Sahu et al., 2017). Integrated nutrient management (INM), which combines organic, inorganic, and biofertilizer inputs, has emerged as an ecologically sound strategy to maintain and improve soil fertility while sustaining long-term crop productivity (Jote, 2023). In this angle use of microbial inoculants very specific to plant nutrient supply are the efficient tools contributing through fixation of atmospheric nitrogen, solubilisation or mineralization of phosphorous and potassium and hormone production for enhancing growth. Integrated use of these inoculants altogether contribute for enhancing plant growth, nutrient acquisition as well as yield and quality of the crops. In light of these challenges and opportunities, the present study evaluated the effect of elite microbial inoculants on growth, nutrient uptake, enzyme activity, and yield of blackgram under field conditions.

2.0 MATERIALS AND METHODS

The study was conducted under field conditions to evaluate the effects of selected microbial inoculants on the growth, yield, nutrient uptake, and enzyme activities of blackgram (*Vigna mungo* L.).

2.1 Microbial Cultures

A total of twelve elite microbial cultures were used in this study: *Methylobacterium thiocyanatum* VRI7-A4, *Azospirillum brasilense* CW903, *Bacillus altitudinis* SSB4, *Bacillus aryabhattai* KSB N2K7, *Peribacillus endoradicis* CE10, *Priestia endophytica* NE14, *Pseudomonas chlororaphis* ZSB15, *Azospirillum lipoferum* AZ204, *Azospirillum brasilense* SP7, *Rhizobium leguminosarum* BMBS P47, *Bacillus megaterium* PSB1, and *Paenibacillus mucilaginosus* KRB9. These cultures were obtained from the Department of Agricultural Microbiology, Tamil Nadu Agricultural University, Coimbatore, and the Agricultural College and Research Institute, Madurai, Tamil Nadu, India. These cultures were given as seed treatment.

2.3 Experimental setup

The experiment was laid out with fourteen treatments i.e., T1 -*Methylobacterium thiocyanatum* VRI7-A4, T2-*Azospirillum brasilense* CW903, T3-*Bacillus altitudinis* SSB4, T4- *Bacillus aryabhattai* KSB N2K7, T5-*Peribacillus endoradicis* CE10, T6-*Priestia endophytica* NE14, T7-*Pseudomonas chlororaphis* ZSB15, T8-*Azospirillum lipoferum* AZ204, T9- *Azospirillum brasilense* SP7, T10-*Rhizobium leguminosarum*, T11-*Bacillus megaterium* (PSB1), T12 -*Paenibacillus mucilaginosus* (KRB9), T13- Uninoculated control T14 - Absolute Control (No fertilizer and no inoculation). Treatments 1-13 received the 100 % RDF.

2.3.1 Experimental details

The field experiment was conducted at the Experimental Research Farm, Department of Agronomy, Agricultural College & Research Institute, Madurai (latitude 9.963°N, longitude 78.199°E, elevation 101 m MSL). The soil was sandy loam, pH 7.61, EC 0.47 dS m⁻¹, available N 224 kg ha⁻¹, P 16.38 kg ha⁻¹, and K 200 kg ha⁻¹. The climate during the cropping period recorded a maximum temperature of 40.2°C, a minimum of 20.9°C, and total rainfall of 89.4 mm. The experimental design was a Randomized Block Design (RBD) with the above 14 treatments and three replications. Black gram variety VBN-8 was sown at 30 × 10 cm spacing, with a seed rate of 20 kg ha⁻¹. Seeds were inoculated with microbial broth cultures (10⁷ cells mL⁻¹) for 1 h and fertilizers were applied (100% RDF) @ 25:50:25 kg ha⁻¹, shade-dried for 30 min, and sown.

2.3.2 Soil physicochemical properties

Soil physicochemical properties were analyzed in the initial soil using standard procedures. Soil pH and electrical conductivity (EC) were determined in a 1:2.5 soil-to-water suspension as per Jackson, 1973. Organic carbon content was estimated following the Walkley and Black (1934) wet oxidation method, in which soil organic matter was oxidized with potassium dichromate (K₂Cr₂O₇) and concentrated sulfuric acid, and the unreacted dichromate was titrated against ferrous ammonium sulfate to determine percent organic carbon. Available nitrogen was estimated by the alkaline KMnO₄ method (Subbiah and Asija, 1956), where 20 g of soil was treated with alkaline KMnO₄ and distilled; the released ammonia was absorbed in boric acid and titrated against standard H₂SO₄. Available phosphorus was determined by the Olsen method (Olsen et al., 1954). 2.5 g of soil was extracted with 50 mL of 0.5 M sodium bicarbonate (NaHCO₃) solution adjusted to pH 8.5. The suspension was shaken and filtered, and the phosphorus concentration in the extract was determined colorimetrically. The intensity of the developed blue colour was measured at 660 nm using a spectrophotometer. Available potassium was estimated using the ammonium acetate extraction method (Stanford and English, 1949), where 5 g of soil was extracted with neutral 1N ammonium acetate solution, filtered, and the

concentration of K⁺ in the extract was determined using a flame photometer. The results were expressed as % organic C and kg ha⁻¹ of available N, P, and K.

2.4 Sampling and observations

Plant and rhizosphere soil samples were collected at 45 DAS. Plant height (cm), dry weight (g plant⁻¹), and the number of nodules per plant were recorded from three randomly selected plants in each replication. Chlorophyll content was measured using a SPAD meter. Plant enzyme activities, including nitrate reductase and plant phosphatase, as well as soil enzyme activities, including soil phosphatase and urease, were analysed from the collected plant and soil samples.

2.4.1 Enzyme assays from plant and soil samples

Enzyme activities in both plant and soil samples were estimated to evaluate the effects of microbial inoculation. Nitrate reductase (NR) activity in plant leaves was determined following Wany et al. (2020). Fresh plant tissues were cut, weighed, and crushed using a micro-pestle or mortar, then homogenized in 2 mL of NR extraction buffer. The homogenate was centrifuged at 14,000 rpm for 30 min at 4°C, and the clear supernatant was collected. To 1 mL of supernatant, 125 µL of 0.5 M zinc acetate and Griess reagent were added, incubated in the dark for 10 min, and absorbance was measured at 546 nm; a magenta colour indicated positive NR activity. Acid phosphatase activity in plant tissues was assayed as per Morton (1955), where in 10 g of tissue was ground with acid-washed sand in 0.2 M acetate buffer (pH 4.5), filtered, and centrifuged at 3000 rpm for 5 min. The supernatant was incubated with p-nitrophenyl phosphate (pNPP), and the released p-nitrophenol was measured at 725 nm and expressed as µg pNP g⁻¹ plant h⁻¹. Total phosphatase activity in soil was estimated following Tabatabai and Bremner (1969), with 1 g soil incubated with 3 mL pNPP and 3.5 mL 0.1 M phosphate buffer for 4 h; reactions were terminated with CaCl₂ and NaOH, filtered, and absorbance recorded at 420 nm. Soil urease activity was determined according to McGarity and Myers (1967) and Kandeler and Gerber (1988). 1 g of soil was incubated with 10% urea solution and citrate buffer (pH 6.7) after the addition of toluene, and incubated at 37°C for 3 h. Following incubation, the reaction mixture was filtered, and the filtrate was treated with phenolate and sodium hypochlorite reagent. The absorbance was measured at 630 nm using a UV-Visible spectrophotometer. The amount of ammonium released was determined from an NH₄⁺-N, and urease activity was expressed as µg NH₄⁺-N released g⁻¹ soil h⁻¹ using the formula: Urease (µg g⁻¹ dwt h⁻¹) = (C × 25) / (dwt × t × 10), where C is NH₄⁺-N concentration, dwt is dry weight, and t is incubation time in hours.

2.5 Yield attributes and nutrient analysis

Blackgram (*Vigna mungo* L.) variety VBN 8 plants were harvested 75 th day after sowing, and plants from the net plot area of each treatment were harvested separately. The harvested plants were sun-dried, and pods were collected, dried, threshed, and winnowed to obtain seed yield. Grain and haulm yields were recorded separately and expressed as kg ha⁻¹.

2.5.1 Plant nutrient analysis

Plant samples were oven-dried, finely ground, and analysed for total nitrogen (N), phosphorus (P), and potassium (K) contents. Total nitrogen was estimated by the micro-Kjeldahl method as described by Humphries (1956). Approximately 10 mL of the plant digest was transferred to a micro-Kjeldahl apparatus, and the released ammonia was distilled into a suitable receiving solution and quantified by titration against standard acid sulfuric acid. Total phosphorus was estimated following Jackson (1973) using the vanadomolybdate yellow colour method after digestion with a triple-acid mixture, and the absorbance was measured using a UV-Visible spectrophotometer. Potassium content was estimated by the flame photometric method following Stanford and English (1949) after triple-acid digestion. The nutrient concentrations were calculated using the respective standard curves or titration values and expressed as percentage (%).

2.5.2 Computation of nutrient uptake and use efficiency

The nutrient uptake (Piper, 1966) and nutrient use efficiency (Moll et al., 1982) were calculated as below

$$\text{Nutrient uptake (kg/ha)} = \frac{\text{Yield} \times \text{Nutrient concentration}}{100}$$

$$\text{Nutrient use efficiency (\%)} = \frac{\text{Nutrient uptake in fertilized plot} - \text{Nutrient uptake in control plot}}{\text{Amount of nutrient applied}} \times 100$$

3. Results

3.1 Effect of bacterial cultures on plant growth and nodulation of blackgram (*Vigna mungo* L. VBN 8) at 45 DAS

Microbial inoculation significantly improved plant growth and nodulation at 45 DAS. *Azospirillum lipoferum* AZ204 recorded the highest plant height (33.40 cm plant), followed by *A. brasilense* CW903 (32.7 cm plant⁻¹) and *Rhizobium leguminosarum* BMBS P47 (32.0 cm plant⁻¹), compared with 19.8 and 18.3 cm plant⁻¹ in the RDF and absolute controls, respectively. Similarly, AZ204 & CW903 recorded the highest biomass (20.2 and 20.1 g plant⁻¹, respectively), followed by BMBS P47 (19.9 g plant⁻¹), compared with 6.3 and 5.9 g plant⁻¹ in their respective controls. Nodulation was highest with AZ204 (18.6 nodules plant⁻¹), followed by BMBS P47 (17.3) and CW903 (15.3), compared with 8.1 and 6.3 nodules plant⁻¹ in the uninoculated and absolute controls, respectively. AZ204, CW903, and BMBS P47 are the overall best treatments in triggering plant growth and nodulation (Figure 1).

3.2 Effect of bacterial cultures on plant and soil enzyme activities at 45 DAS

Microbial inoculation increased the activity of soil and plant enzymes. Nitrate reductase activity was highest in AZ204 inoculation ($59.80 \mu\text{g NO}_2^- \text{ h}^{-1} \text{ g}^{-1}$), followed by CW903 and PSB1 ($57.20 \mu\text{g NO}_2^- \text{ h}^{-1} \text{ g}^{-1}$) and the uninoculated and absolute controls (51.30 and $48.32 \mu\text{g NO}_2^- \text{ h}^{-1} \text{ g}^{-1}$, respectively). CW903 had the highest plant phosphatase activity ($0.41 \mu\text{mol PNP ml}^{-1}$), followed by AZ204 (0.39) and PSB1 (0.38), whereas CW903 had the highest soil phosphatase activity ($2.55 \mu\text{mol PNP ml}^{-1}$), followed by AZ204 (2.51) and BMBS P47 (2.50). AZ204 had the highest soil urease activity ($9.65 \mu\text{g NH}_4\text{-N g dry soil h}^{-1}$), followed by BMBS P47 ($8.77 \mu\text{g NH}_4\text{-N g dry soil h}^{-1}$). In terms of increasing plant and soil enzyme activities, AZ204, CW903, and BMBS P47 performed better (Figures 2-5).

3.3 Effect of bacterial cultures on yield, nutrient uptake and nutrient use efficiency

Yield, nutrient absorption, and nutrient-use efficiency were all enhanced by microbial inoculation. AZ204 had the highest grain yield (952.7 kg ha^{-1}), closely followed by CW903 (952.3 kg ha^{-1}) and BMBS P47 (915.0 kg ha^{-1}), whereas the uninoculated and absolute controls had grain yields of 800 and 723 kg ha^{-1} , respectively. SSB4 had the largest haulm yield ($2210.55 \text{ kg ha}^{-1}$), followed by SP7 ($2198.31 \text{ kg ha}^{-1}$) and KSB N2K7 ($2209.23 \text{ kg ha}^{-1}$). BMBS P47 had the highest total N uptake (27.1 kg ha^{-1}), followed by AZ204 (27.0) and CW903 (26.9). AZ204 had the highest P uptake (16.2 kg ha^{-1}), followed by BMBS P47 (15.6) and PSB1 (15.5) (Table 1). AZ204 had the highest PUE (17.2%), followed by BMBS P47 (16.0%) and PSB1 (15.8%), while BMBS P47 had the highest NUE (30.4%), followed by AZ204 (30.0%) and CW903 (29.6%). Overall, the best inoculants for increasing grain output, nutrient uptake, and nutrient-use efficiency were AZ204, CW903, and BMBS P47 (Table 1 & 2) and (Figure 6).

3.4 Principal component analysis of growth, biochemical and nutrient use traits

PCA was performed to determine the relationships among microbial treatments and growth, biochemical, yield, and nutrient-use traits. The first two principal components explained 83.2% of the total variation, with Dim1 and Dim2 accounting for 71.0 and 12.2% , respectively (Figures 7 & 8). Plant height, biomass, nodulation, grain yield, nitrate reductase, plant phosphatase, soil phosphatase, soil urease, N uptake, P uptake, N use efficiency (NUE), and P use efficiency (PUE) were positively associated with Dim1, whereas haulm yield showed a contrasting negative association. Among the variables, PUE, NUE, P uptake, nodulation, and N uptake showed relatively higher contributions to the PCA. The treatment score plot showed clear separation among inoculants, with AZ204 and BMBS P47 positioned distinctly toward the positive Dim1 region, while CW903 also showed a favourable association with major performance traits. The remaining inoculants occupied intermediate positions, whereas the uninoculated and absolute controls were clearly separated from most inoculated treatments.

4. DISCUSSION

Microbial inoculation markedly improved growth, nodulation, nutrient acquisition and productivity of blackgram compared with the uninoculated control. Among the tested cultures, AZ204, CW903 and BMBS P47 consistently performed better for plant growth, nodulation, enzyme activities and grain yield. The superior response of these cultures may be associated with complementary plant-growth-promoting mechanisms, including biological N fixation, phytohormone production, improved root development and enhanced nutrient mobilization. Similar improvements in growth and productivity of blackgram following the application of beneficial microorganisms have been reported by Kumpawat (2010), Soni et al., (2017) and Kanwadiya et al., (2023).

The enhanced enzyme activities further indicated improved nutrient transformation under microbial inoculation. AZ204 recorded the highest nitrate reductase and urease activities, whereas CW903 showed greater phosphatase activity. Increased nitrate reductase activity may facilitate N assimilation, while microbial phosphatase activity can enhance P mobilization from less available soil P pools. Microorganisms capable of P solubilization and mineralization can improve P availability through organic-acid production and phosphatase activity (Chen et al., 2016; Khalid et al., 2023). Recent findings in blackgram also demonstrated that phosphate-solubilizing bacterial inoculation enhanced phosphatase activity, nutrient uptake and P-use efficiency (Sushmita et al., 2025).

Microbial inoculation also increased N and P uptake over the uninoculated control. BMBS P47 recorded the highest N uptake (27.1 kg ha^{-1}), representing a 7.1% increase over the uninoculated control (25.3 kg ha^{-1}), followed by AZ204 (27.0 kg ha^{-1} ; 6.7%) and CW903 (26.9 kg ha^{-1} ; 6.3%). Compared with the absolute control (19.5 kg ha^{-1}), N uptake was higher by 39.0 , 38.5 and 38.0% under BMBS P47, AZ204 and CW903, respectively. The greater N uptake under these cultures may be related to enhanced N fixation, root proliferation and improved N assimilation. Previous studies have similarly reported increased N uptake in blackgram following *Rhizobium* and biofertilizer application (Tanwar et al., 2003; Kanwadiya et al., 2023).

The response of P uptake was more pronounced than N uptake. AZ204 recorded the highest P uptake (16.2 kg ha^{-1}), which was 19.1% higher than the uninoculated control (13.6 kg ha^{-1}), followed by BMBS P47 (15.6 kg ha^{-1} ; 14.7%) and PSB1 (15.5 kg ha^{-1} ; 14.0%). Relative to the absolute control (7.6 kg ha^{-1}), P uptake increased by 113.2% with AZ204, 105.3% with BMBS P47 and 103.9% with PSB1. This improvement could be attributed to microbial-mediated P solubilization, phosphatase activity and enhanced root acquisition of available P. The positive effect of *Rhizobium* and phosphate-solubilizing microorganisms on P uptake and productivity of blackgram has also been reported previously (Tanwar et al., 2003; Surya Kant et al., 2017; Sushmita et al., 2025).

The improvement in nutrient uptake was reflected in N-use efficiency (NUE) and P-use efficiency (PUE). NUE increased from 23.2% in the uninoculated control to 30.4% with BMBS P47, representing a 31.0% increase, followed by AZ204 (30.0%; 29.3%) and CW903 (29.6%; 27.6%). Thus, BMBS P47, AZ204 and CW903 were the most effective cultures for improving NUE. Similarly, PUE increased from 12.0% in the uninoculated control to 17.2% with AZ204, representing a 43.3% increase, followed by BMBS P47 (16.0%; 33.3%) and PSB1 (15.8%; 31.7%). Therefore, AZ204 was the most effective culture for PUE, followed by BMBS P47 and PSB1. The improvement in NUE and PUE suggests that these cultures enhanced both nutrient acquisition and the efficiency with which absorbed nutrients were utilized for plant production. Similar improvements in nutrient uptake and nutrient-use efficiency following biofertilizer and PSB application have been reported in blackgram (Surya Kant et al., 2017; Swain et al., 2025).

The improved nutrient acquisition was accompanied by greater grain yield. AZ204 (952.7 kg ha⁻¹) and CW903 (952.3 kg ha⁻¹) recorded the highest grain yields, followed by BMBS P47 (915.0 kg ha⁻¹), compared with 800.0 kg ha⁻¹ in the uninoculated control and 723.0 kg ha⁻¹ in the absolute control. The higher productivity of these cultures was associated with their superior growth, nodulation, enzyme activities and nutrient-use efficiency, indicating an integrated microbial effect on crop performance. Previous studies have likewise demonstrated that biofertilizer and PSB application can improve growth, nutrient uptake and yield of blackgram (Tanwar et al., 2003; Surya Kant et al., 2017; Swain et al., 2025).

The PCA provided additional evidence for the overall superiority of the inoculated treatments. The first two principal components accounted for 83.2% of the total variation, with Dim1 and Dim2 explaining 71.0 and 12.2%, respectively. NUE, PUE, P uptake, N uptake and nodulation showed strong positive associations with Dim1, indicating that nutrient acquisition and nutrient-use efficiency were major contributors to the variation among the bacterial cultures. AZ204 and BMBS P47 were positioned towards the positive region of Dim1, while CW903 also showed a favourable association with the major growth and nutrient-use-efficiency attributes. This agrees with the PCA study of Ajaykumar et al. (2023) in blackgram, where grain yield, dry matter production and nitrate reductase activity were among the major contributors to the principal components following application of biological inputs. Thus, the PCA supports the individual trait results and identifies AZ204, BMBS P47 and CW903 as the most promising cultures, with AZ204 particularly associated with P uptake and PUE, and BMBS P47 with N uptake and NUE.

1. CONCLUSION

This study demonstrated the complementary value of field experiments in evaluating the impact of microbial inoculants on blackgram growth, yield, and nutrient acquisition under field conditions. Certain microbial cultures, including *Azospirillum lipoferum* AZ204, *Azospirillum brasilense* CW903, *Pseudomonas chlororaphis* ZSB15, *Bacillus megaterium* (PSB1), and *Paenibacillus mucilaginosus* (KRB9), consistently enhanced plant growth, yield, and nutrient uptake relative to the uninoculated control. PCA analysis further highlighted that growth- and nutrient related traits (plant height, biomass, grain yield, nutrient uptake, and nutrient use efficiencies) accounted for most of the observed variation, with superior clustering of AZ204, CW 903 and BMBS P 47 treatments along Dim1, indicating their strong influence on plant performance. Based on these results the bacterial cultures such as *Azospirillum lipoferum* AZ204, *Azospirillum brasilense* CW903, *Rhizobium leguminosarum* BMBS P 47, *Bacillus megaterium* (PSB1), *Paenibacillus mucilaginosus* (KRB9) and *Pseudomonas chlororaphis* ZSB15 can be considered for the development of bacterial consortium for enhancing nutrient use efficiency.

Data Availability Statement: The data supporting the findings of this study are available within the article and its supplementary materials.

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Table 1: Effect of bacterial cultures on yield and nutrient uptake of black gram (VBN-8) under a field experiment

Treatments		Grain yield (kg/ha)	Halum yield (kg/ha)	Total uptake N (kg/ha)	Total uptake P (kg/ha)
T1	VRI7-A4 - <i>Methylobacterium thiocyanatum</i>	842 ^{cd}	2100.09 ^{cd}	25.4 ^b	14.2 ^{cd}
T2	CW903 - <i>Azospirillum brasilense</i>	952.3 ^a	1899.77 ^{fg}	26.9 ^a	14.5 ^c
T3	SSB4 - <i>Bacillus altitudinis</i>	769 ^{ef}	2210.55 ^a	26.0 ^{ab}	14.1 ^{cd}
T4	KSB N2K7- <i>Bacillus aryabhatai</i>	788 ^e	2209.23 ^a	25.5 ^b	13.7 ^d
T5	CE10- <i>Peribacillus endoradicis</i> ;	805 ^{de}	2156.42 ^{abc}	25.4 ^b	13.9 ^d
T6	NE14 - <i>Priestia endophytica</i>	802 ^e	2110.92 ^{bc}	25.5 ^b	13.6 ^d
T7	ZSB15- <i>Pseudomonas chlororaphis</i>	904.6 ^b	1977.43 ^{ef}	25.9 ^{ab}	14.6 ^c
T8	AZ204- <i>Azospirillum lipoferum</i>	952.7 ^a	1873.25 ^{gh}	27.0 ^a	16.2 ^a
T9	SP7- <i>Azospirillum brasilense</i>	741 ^{fg}	2198.31 ^{ab}	25.6 ^b	14.1 ^{cd}
T10	BMBS P47- <i>Rhizobium leguminosarum</i>	915 ^{ab}	1953.77 ^{efg}	27.1 ^a	15.6 ^{bc}
T11	PSB1- <i>Bacillus megaterium</i>	910.7 ^b	1927.92 ^{efg}	26.5 ^{ab}	15.5 ^b
T12	KRB9- <i>Paenibacillus mucilaginosus</i>	860 ^c	1936.73 ^{efg}	26.3 ^{ab}	15.2 ^b
T13	Uninoculated control (100% RDF)	800 ^e	1780.21 ^h	25.3 ^b	13.6 ^d
T14	Absolute control (Without any application)	723 ^g	2012.76 ^{de}	19.5 ^c	7.6 ^e
	CD	37.5	88.434	1.144	0.528
	SEd	18.24	43.023	0.556	0.118

Table 2: Effect of bacterial cultures on nitrogen (N) and Phosphorus (P) use efficiency of black gram (VBN-8) under a field experiment

Treatments		NUE(%)	PUE(%)
T1	VRI7-A4 - <i>Methylobacterium thiocyanatum</i>	23.6 ^{de}	13.2 ^e
T2	CW903 - <i>Azospirillum brasilense</i>	29.6 ^a	13.8 ^d
T3	SSB4 - <i>Bacillus altitudinis</i>	26.0 ^c	13.0 ^e
T4	KSB N2K7- <i>Bacillus aryabhatai</i>	24.0 ^{de}	12.2 ^f
T5	CE10- <i>Peribacillus endoradicis</i> ;	23.6 ^{de}	12.6 ^{ef}
T6	NE14 - <i>Priestia endophytica</i>	24.0 ^{de}	12.0 ^f
T7	ZSB15- <i>Pseudomonas chlororaphis</i>	25.6 ^c	14.0 ^d
T8	AZ204- <i>Azospirillum lipoferum</i>	30.0 ^a	17.2 ^a
T9	SP7- <i>Azospirillum brasilense</i>	24.4 ^d	13.0 ^e
T10	BMBS P47- <i>Rhizobium leguminosarum</i>	30.4 ^a	16.0 ^b
T11	PSB1- <i>Bacillus megaterium</i>	28.0 ^b	15.8 ^b
T12	KRB9- <i>Paenibacillus mucilaginosus</i>	27.2 ^b	15.2 ^c
T13	Uninoculated control (100% RDF)	23.2 ^c	12.0 ^f
T14	Absolute control (Without any application)	-	-
	CD	1.011	0.561
	SEd	0.490	0.272

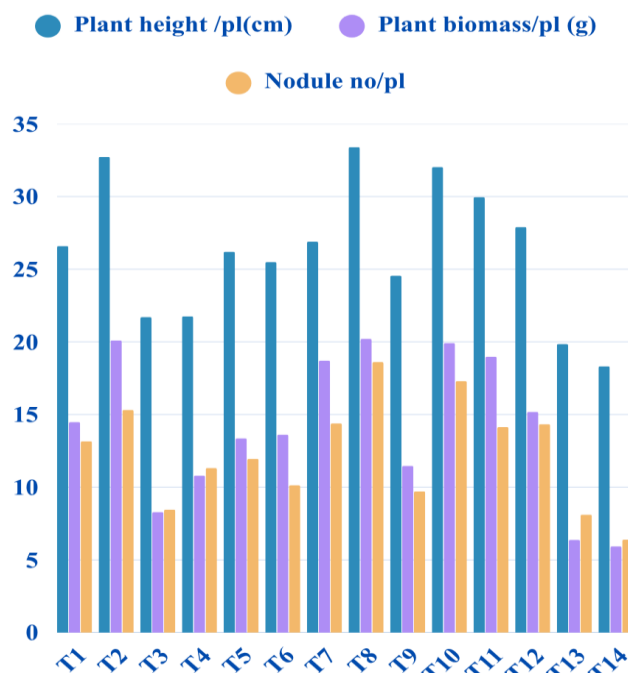


Figure 1: Effect of bacterial cultures on plant growth and nodulation in blackgram at 45 DAS.

*Bars represent treatment means

T1-VRI7-A4 - *Methylobacterium thiocyanatum*; **T2-CW903** - *Azospirillum brasilense*; **T3-SSB4** - *Bacillus altitudinis*; **T4-KSB N2K7** *Bacillus aryabhattai*; **T5-CE10** *Peribacillus endoradicis*; **T6-NE14** *Priestia endophytica*; **T7-ZSB15** *Pseudomonas chlororaphis*; **T8-AZ204** *Azospirillum lipoferum*; **T9-SP7** *Azospirillum brasilense*; **T10-BMBS P47** *Rhizobium leguminosarum*; **T11-PSB1** *Bacillus megaterium*; and **T12-KRB9** *Paenibacillus mucilaginosus*; **T13-** Unioculated control (100% RDF); **T14-** Absolute control (Without any application)

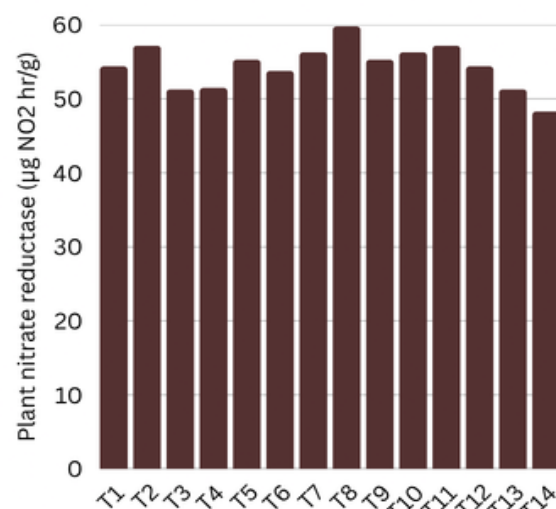


Figure 2: Effect of bacterial cultures on nitrate reductase in blackgram plants at 45 DAS.

*Bars represent treatment means

T1-VRI7-A4 - *Methylobacterium thiocyanatum*; **T2-CW903** - *Azospirillum brasilense*; **T3-SSB4** - *Bacillus altitudinis*; **T4-KSB N2K7** *Bacillus aryabhattai*; **T5-CE10** *Peribacillus endoradicis*; **T6-NE14** *Priestia endophytica*; **T7-ZSB15** *Pseudomonas chlororaphis*; **T8-AZ204** *Azospirillum lipoferum*; **T9-SP7** *Azospirillum brasilense*; **T10-BMBS P47** *Rhizobium leguminosarum*; **T11-PSB1** *Bacillus megaterium*; and **T12-KRB9** *Paenibacillus mucilaginosus*; **T13-** Unioculated control (100% RDF); **T14-** Absolute control (Without any application)

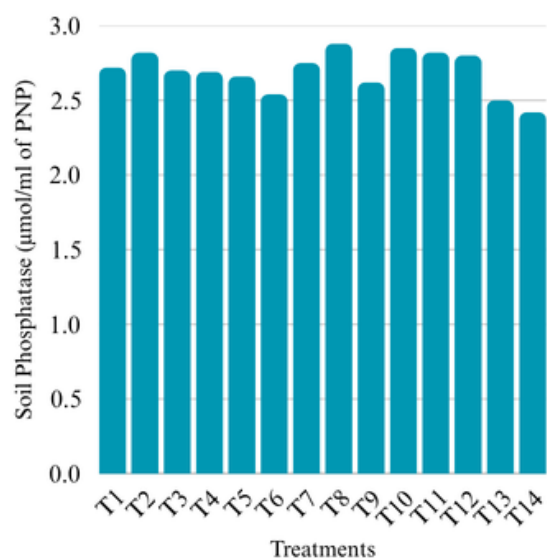
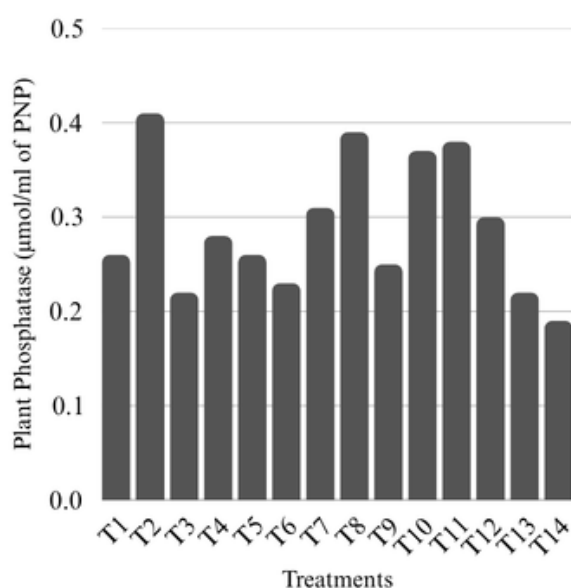


Figure 3: Effect of bacterial cultures on plant phosphatase activity in blackgram at 45 DAS

*Bars represent treatment means

T1-VRI7-A4 - *Methylobacterium thiocyanatum*; **T2-CW903** - *Azospirillum brasilense*; **T3-SSB4** - *Bacillus altitudinis*; **T4-KSB N2K7** *Bacillus aryabhattai*; **T5-CE10** *Peribacillus endoradicis*; **T6-NE14** *Priestia endophytica*; **T7-ZSB15** *Pseudomonas chlororaphis*; **T8-AZ204** *Azospirillum lipoferum*; **T9-SP7** *Azospirillum brasilense*; **T10-BMBS P47** *Rhizobium leguminosarum*; **T11-PSB1** *Bacillus megaterium*; and **T12-KRB9** *Paenibacillus mucilaginosus*; **T13-** Unioculated control (100% RDF); **T14-** Absolute control (Without any application)

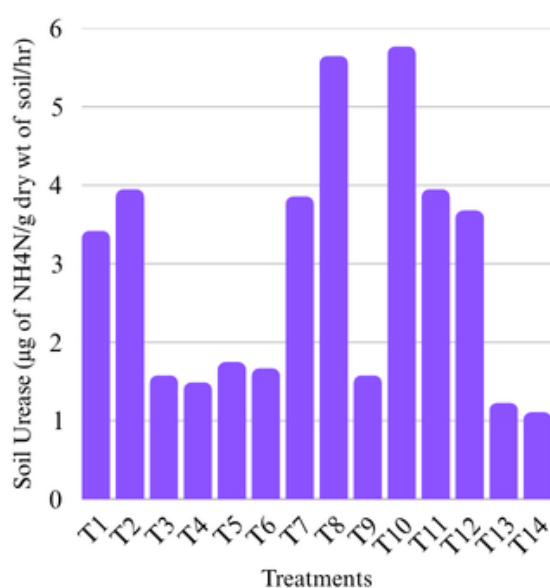


Figure 5: Effect of bacterial cultures on soil urease activity in blackgram at 45 DAS

*Bars represent treatment means

T1-VRI7-A4 - *Methylobacterium thiocyanatum*; **T2-CW903** - *Azospirillum brasilense*; **T3-SSB4** - *Bacillus altitudinis*; **T4-KSB N2K7** *Bacillus aryabhattai*; **T5-CE10** *Peribacillus endoradicis*; **T6-NE14** *Priestia endophytica*; **T7-ZSB15** *Pseudomonas chlororaphis*; **T8-AZ204** *Azospirillum lipoferum*; **T9-SP7** *Azospirillum brasilense*; **T10-BMBS P47** *Rhizobium leguminosarum*; **T11-PSB1** *Bacillus megaterium*; and **T12-KRB9** *Paenibacillus mucilaginosus*; **T13-** Unioculated control (100% RDF); **T14-** Absolute control (Without any application)

Figure 4: Effect of bacterial cultures on soil phosphatase activity in blackgram at 45 DAS

*Bars represent treatment means

T1-VRI7-A4 - *Methylobacterium thiocyanatum*; **T2-CW903** - *Azospirillum brasilense*; **T3-SSB4** - *Bacillus altitudinis*; **T4-KSB N2K7** *Bacillus aryabhattai*; **T5-CE10** *Peribacillus endoradicis*; **T6-NE14** *Priestia endophytica*; **T7-ZSB15** *Pseudomonas chlororaphis*; **T8-AZ204** *Azospirillum lipoferum*; **T9-SP7** *Azospirillum brasilense*; **T10-BMBS P47** *Rhizobium leguminosarum*; **T11-PSB1** *Bacillus megaterium*; and **T12-KRB9** *Paenibacillus mucilaginosus*; **T13-** Unioculated control (100% RDF); **T14-** Absolute control (Without any application)

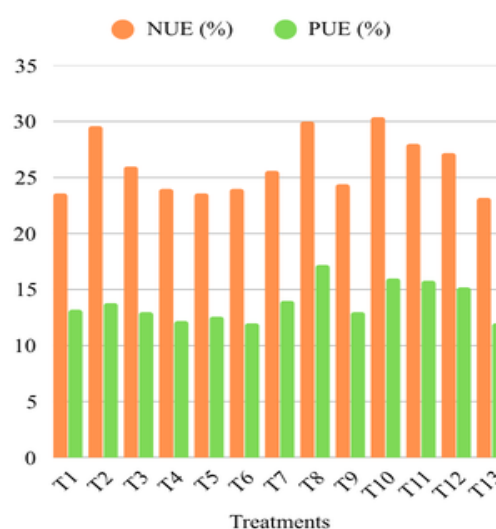


Figure 6: Effect of bacterial cultures on Nitrogen use efficiency and Phosphorus use efficiency in blackgram

*Bars represent treatment means

T1-VRI7-A4 - *Methylobacterium thiocyanatum*; **T2-CW903** - *Azospirillum brasilense*; **T3-SSB4** - *Bacillus altitudinis*; **T4-KSB N2K7** *Bacillus aryabhattai*; **T5-CE10** *Peribacillus endoradicis*; **T6-NE14** *Priestia endophytica*; **T7-ZSB15** *Pseudomonas chlororaphis*; **T8-AZ204** *Azospirillum lipoferum*; **T9-SP7** *Azospirillum brasilense*; **T10-BMBS P47** *Rhizobium leguminosarum*; **T11-PSB1** *Bacillus megaterium*; and **T12-KRB9** *Paenibacillus mucilaginosus*; **T13-** Unioculated control (100% RDF); **T14-** Absolute control (Without any application)

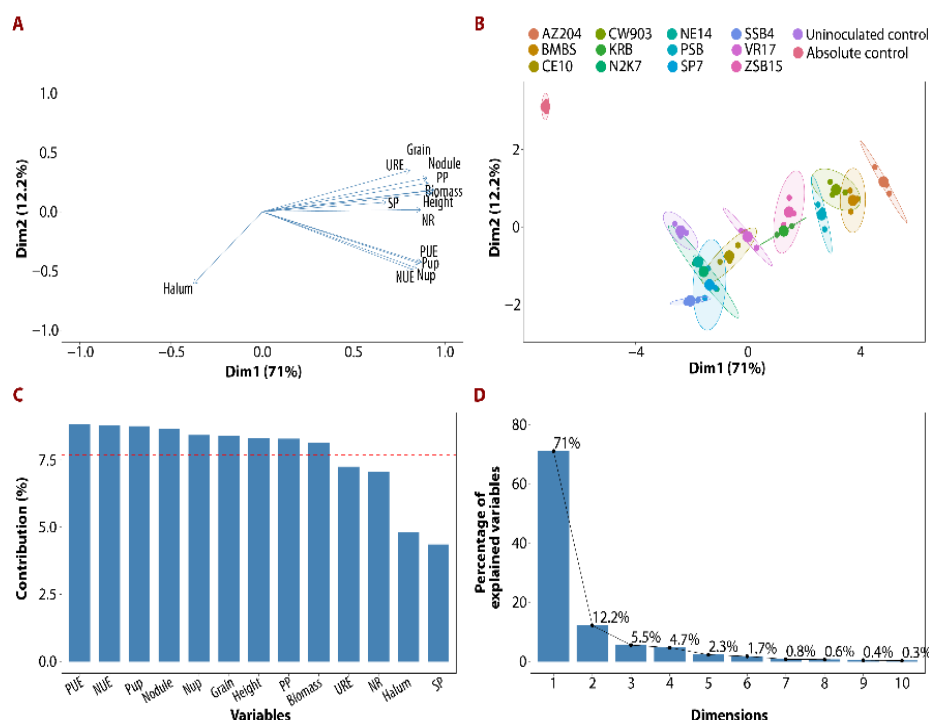


Figure 7: Principal component and correlation analysis of growth, enzyme activities, yield, and nutrient-use traits of blackgram under bacterial inoculation. (A) PCA variable loading plot showing the association of measured traits with Dim1 and Dim2; (B) treatment score plot showing the distribution of microbial inoculants and control treatments; (C) percentage contribution of individual variables to the PCA; and (D) percentage of variance explained by successive principal components. The variables included in the PCA were plant height, plant biomass, nodulation, grain yield, haulm yield, nitrate reductase, plant phosphatase, soil phosphatase, soil urease, N uptake, P uptake, N-use efficiency (NUE) and P-use efficiency (PUE). Dim1 (PC1) and Dim2 (PC2) explained 71.0 and 12.2% of the total variation, respectively, with the first two components cumulatively explaining 83.2% of the total variation.

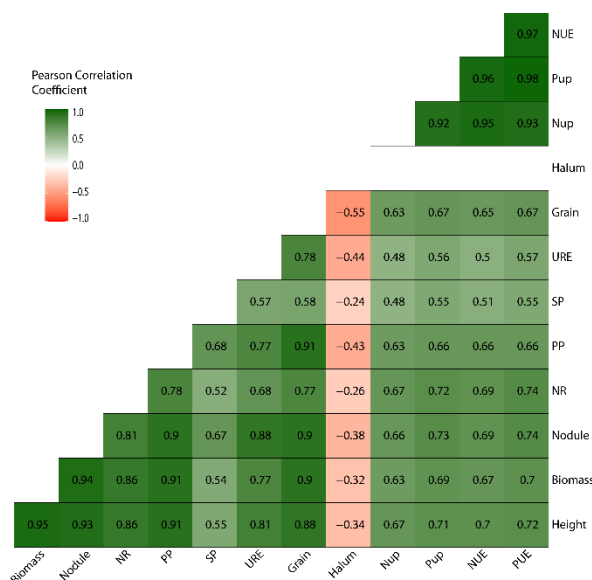


Figure 8: The correlation matrix shows Pearson correlation coefficients among plant growth, biochemical, yield, and nutrient-use traits.