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Evaluation of liquid bio-fertilizers in enhancing seed yield and quality in soybean variety JS 20-116

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Abstract

Soybean (*Glycine max* L.) is a globally important leguminous crop valued for its protein and oil content. However, its productivity is often limited by soil nutrient deficiencies and unsustainable reliance on chemical fertilizers. This study was conducted during the 2024 *kharif* season at VNMKV, Parbhani, under the All India Coordinated Research Project on National Seed Project (AICRP-NSP), to evaluate the efficacy of different liquid biofertilizers in enhancing seed yield and quality of soybean variety JS 20-116. The experiment comprised eight treatments including Rhizobium, Azospirillum, phosphate-solubilizing bacteria (PSB), potassium-solubilizing bacteria (KSB), *Pseudomonas fluorescens*, effective microorganisms (EM), state-recommended practices, and an untreated control, arranged in a randomized block design with three replications. Significant variation was observed among treatments for emergence percentage, nodulation, plant height, pod number, grain yield, seed quality parameters, and economic returns. Rhizobium (T₃) showed the highest emergence (83.6%) and nodulation (74.4 nodules/plant), while KSB (T₆) recorded the tallest plants at harvest (50.2 cm), highest pod number (51.7 pods/plant), and highest grain yield per plant (13.2 g). The maximum seed yield per hectare was observed in the state-recommended package (T₂: 25.5 q/ha), followed by KSB (25.0 q/ha) and Rhizobium (23.9 q/ha). Germination percentage and seedling vigor indices were also enhanced in Rhizobium and KSB treatments. The highest net monetary returns and benefit-cost ratios were recorded in T₂, followed closely by KSB and Rhizobium. ACC-reductase activity, a marker for stress tolerance, was highest in T₂ and T₃. These results highlight the potential of liquid biofertilizers, particularly KSB and Rhizobium, as effective alternatives or complements to chemical inputs for sustainable soybean cultivation, offering improved yield, seed quality, and profitability under rainfed conditions.

Keywords: Soybean (*Glycine max*), liquid biofertilizers, rhizobium, EM culture, seed yield and quality

Introduction

Soybean (*Glycine max* L.) is one of the most economically important leguminous crops worldwide, known for its high protein (approximately 40%) and oil (about 20%) content. It is widely grown in tropical and subtropical regions and serves as a vital raw material for food, animal feed, and industrial products such as biodiesel, lecithin, and protein isolates (Ali *et al.*, 2010) ^[1]. In India, it occupies a significant share in *kharif* cropping systems and contributes substantially to farmers' income. Despite its potential, soybean productivity is often constrained by imbalanced fertilization, low soil organic carbon, and deteriorating soil health due to continuous use of chemical inputs (Saxena & Tilak, 1998) ^[9].

The use of biofertilizers—especially liquid formulations—has emerged as a sustainable strategy for improving crop productivity and seed quality. Liquid biofertilizers are biologically active preparations containing beneficial microorganisms that can fix atmospheric nitrogen, solubilize phosphorus and potassium, and enhance nutrient uptake by plants (Glick, 2012; Vessey, 2003) ^[4, 11]. Compared to carrier-based formulations, liquid biofertilizers offer improved shelf life, consistency in microbial load, and better adaptability under field conditions (Bashan *et al.*, 2014) ^[3]. Microbial strains such as *Rhizobium*, *Azospirillum*, phosphate-solubilizing bacteria (PSB), potassium-solubilizing bacteria (KSB), *Pseudomonas fluorescens*, and effective microorganisms (EM) have shown promising results in promoting plant growth and improving soil fertility (Mahdi *et al.*, 2010; Meena *et al.*, 2018) ^[6, 7].

In soybean, *Rhizobium* inoculation enhances nodulation and biological nitrogen fixation, which

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is vital for protein biosynthesis. PSB and KSB mobilize phosphorus and potassium, respectively, facilitating better root growth, flowering, and grain development. *Azospirillum* and *Pseudomonas* are also known for producing phytohormones such as indole acetic acid (IAA) and gibberellins that regulate plant growth and stress tolerance (Spaepen *et al.*, 2007; Glick, 2014)^[10, 5].

Given the spatial variability in soil types, climatic conditions, and crop responses, it is essential to evaluate the performance of various liquid biofertilizers under local agro-ecological settings. This experiment was conducted under the All India Coordinated Research Project on National Seed Project (AICRP-NSP) to evaluate the efficacy of different liquid biofertilizers in enhancing field performance, seed yield, quality, and economic returns in soybean. The ultimate aim is to identify suitable biofertilizer options for organic seed production and promote sustainable intensification of soybean-based cropping systems.

Materials and Methods

Experimental Site and Soil

The experiment was carried out during the 2024 *kharif* season at the Seed Technology Research Unit, Vasantrao Naik Marathwada Krishi Vidyapeeth (VNMKV), Parbhani, Maharashtra, located at 19.15°N latitude and 76.77°E longitude with an elevation of 414 meters above mean sea level. The site experiences a semi-arid climate with mean annual rainfall of 800-900 mm, predominantly during June to September. The soil of the experimental field was medium black clay loam with moderate fertility. Pre-sowing analysis indicated the soil had a slightly alkaline pH (7.6), electrical conductivity (EC) of 0.41 dS/m, organic carbon content of 0.52%, and available nitrogen, phosphorus, and potassium contents of 242, 18.7, and 311 kg/ha, respectively.

Experimental Design and Treatments

The experiment was laid out in a randomized block design (RBD) with eight treatments replicated thrice. The soybean variety JS 20-116, known for its high yield potential and adaptability, was used. The net plot size was 5.0 m × 2.25 m with a spacing of 45 cm between rows and 5 cm between plants. Sowing was undertaken on 10 July 2024.

Treatment details

- T₁: Control (no seed treatment)
- T₂: State recommended package of practices
- T₃: Jawahar *Rhizobium* culture @ 20 ml/kg seed
- T₄: Jawahar EM culture @ 20 ml/kg seed
- T₅: Jawahar PSB @ 20 ml/kg seed
- T₆: Jawahar KSB @ 20 ml/kg seed
- T₇: Jawahar *Azospirillum* @ 20 ml/kg seed
- T₈: Jawahar *Pseudomonas* @ 20 ml/kg seed

Crop Management and Observations

Standard cultural practices recommended for soybean under rainfed conditions were adopted uniformly across all treatments. Seed treatment was carried out by mixing the biofertilizer formulations thoroughly and allowing the seeds to air-dry before sowing. Irrigation, weed control, and pest management operations were performed uniformly. Observations were recorded on seedling emergence percentage, plant population at establishment, number of nodules per plant, SPAD chlorophyll readings, plant height at 30 days after sowing (DAS) and at harvest, number of pods per plant, grain yield per plant, plot and

hectare basis, and 1000-seed weight. Seed quality parameters included laboratory germination percentage, seedling vigor indices (VI-I and VI-II), nitrogen metabolism rate (NMR), benefit-cost ratio (B: C), and ACC-reductase activity (ARA), expressed in µmol ethylene per gram dry weight.

Statistical Analysis

The experimental data were subjected to analysis of variance (ANOVA) for RBD using appropriate statistical software. Treatment means were compared using critical difference (CD) at 5% level of significance. Coefficient of variation (CV), standard error of mean (SEm), and least significant difference (LSD) values were calculated to ascertain the reliability of observed differences among treatments.

Results and Discussion

Significant differences were observed among treatments for field emergence and plant stand establishment (CD@5% = 5.09% and 5.05 plants/m², respectively). The highest emergence percentage was noted in T₃ (*Rhizobium*, 83.6%) followed by T₅ (PSB, 83.0%) and T₂ (State package, 82.4%). This enhanced emergence in biofertilizer treatments is attributed to early microbial colonization of the rhizosphere, which improves seedling vigor and nutrient accessibility (Vessey, 2003)^[11]. *Rhizobium* facilitates symbiotic nitrogen fixation, thereby enhancing early seedling development (Glick, 2012)^[4]. *Rhizobium* (T₃) recorded the highest nodulation (74.4 nodules/plant), followed by KSB (T₆, 73.2) and *Pseudomonas* (T₈, 72.3). This result reinforces the role of *Rhizobium* in enhancing biological nitrogen fixation (Yadav *et al.*, 2011)^[12], while the contributions of KSB and *Pseudomonas* could be due to their phosphate and potassium solubilization traits, indirectly enhancing root development and symbiosis (Mahdi *et al.*, 2010)^[6]. SPAD readings, though statistically non-significant, remained within optimal chlorophyll thresholds (45-46), suggesting that none of the treatments led to chlorophyll degradation, and photosynthetic capacity remained intact.

Plant height at 30 DAS was significantly greater in PSB (T₅: 35.4 cm) and T₂ (33.5 cm), indicating early vegetative growth enhancement due to increased P availability. At harvest, KSB (T₆) produced the tallest plants (50.2 cm), which is consistent with its role in solubilizing potassium that improves turgor, enzyme activation, and translocation of photosynthates (Basak & Biswas, 2010)^[2]. Pod number was significantly influenced by treatments, with KSB (51.7 pods/plant) and PSB (49.7) outperforming others, reflecting increased nutrient use efficiency and better source-sink relationship (Meena *et al.*, 2018)^[7]. The highest grain yield per hectare was obtained with T₂ (25.5 q/ha), followed closely by T₆ (25.0 q/ha) and T₃ (23.9 q/ha). Grain yield per plant was also highest in KSB (13.2 g), suggesting improved nutrient acquisition and mobilization at critical reproductive stages. These results align with Reddy *et al.* (2014)^[8], who reported that biofertilizers like PSB and KSB significantly enhanced pod set and yield through improved soil nutrient bioavailability. Furthermore, T₆ and T₃ recorded superior 1000-seed weights (105.0 and 107.4 g, respectively), indicating better seed development and filling.

T₃ (*Rhizobium*) recorded the highest germination percentage (84.6%), followed by T₂ (82.6%) and T₅ (83.2%). Higher germination in *Rhizobium*-treated seeds may be due to increased protein synthesis and balanced hormonal regulation in seeds (Glick, 2012)^[4]. Vigor indices (VI-I and VI-II) were superior in T₆ and T₂, indicating that biofertilizer applications positively affect seedling vigor. The role of potassium solubilization by

KSB in enhancing enzyme activity and osmotic balance might explain its effect on seedling performance (Basak & Biswas, 2010) [2]. Maximum net monetary return (NMR) and benefit-cost ratio (B: C) were obtained in T₂ (Rs. 1, 08, 791; B:C 1.9), closely followed by KSB (T₆: Rs. 1,05,131; B:C 1.8) and Rhizobium (T₃: Rs. 98,391; B:C 1.7). This implies that the economic benefit of biofertilizer use can be comparable to full chemical packages, promoting sustainable and cost-effective seed production systems. Biofertilizer-treated plots required lower external inputs while still maintaining competitive yields,

enhancing overall profitability (Mahdi *et al.*, 2010) [6]. ACC-reductase activity, which serves as a proxy for ethylene regulation and plant stress mitigation, was highest in T₂ (4.25 µmol ethylene/g dw) and T₃ (4.12). This suggests that both full nutrient package and Rhizobium application help mitigate stress-induced ethylene synthesis, thereby promoting better seedling development under variable field conditions (Glick, 2014) [5]. Enhanced ARA in these treatments indicates improved hormonal regulation and enzymatic resilience, aiding plant performance and seed quality.

Table 1: Evaluation of liquid bio-fertilizers in enhancing seed yield and quality

Treatments	Field emergence %	Plant stand establishment/m ²	No of Nodules per plant	SCMR (SPAD)	Plant height @ 30 DAS (cm)	Final height at harvesting (cm)	No of pods per plant	Grain yield/plant (g)	Grain yield/Plot (kg)	Grain yield/ha (q)	1000-seed wt (g)
T ₁	76.1	39.0	67.1	45.6	30.7	44.1	43.7	10.8	2.17	17.3	100.1
T ₂	82.4	46.2	71.4	45.7	33.5	49.6	48.1	12.4	2.87	25.5	106.1
T ₃	83.6	44.2	74.4	45.8	32.8	46.4	47.4	12.7	2.69	23.9	107.4
T ₄	76.5	37.3	67.5	45.9	31.8	45.4	47.2	11.0	2.38	21.2	105.7
T ₅	83.0	48.7	72.4	46.1	35.4	46.1	49.7	12.1	2.54	22.6	106.3
T ₆	80.8	41.7	73.2	45.1	31.1	50.2	51.7	13.2	2.81	25.0	105.0
T ₇	76.1	39.0	64.9	46.4	30.8	43.8	45.0	9.6	2.16	19.2	103.4
T ₈	75.6	36.7	72.3	45.8	31.5	44.0	44.9	10.8	2.01	17.9	107.1
CV	4.15	7.43	4.99	4.85	9.30	5.20	7.42	10.80	10.23	10.20	1.61
SEm	1.71	1.69	2.87	0.75	0.73	1.27	1.82	0.72	0.14	126.22	0.98
CD @5%	5.09	5.05	6.21	NS	2.19	3.80	5.57	2.21	0.44	390.22	2.99

Treatments	GP %	VI-I	VI-II	NMR (Rs)	B:C ratio	ARA umol ethylene/g dry weight
T ₁	76.5	1550	2574.2	68154.5	1.2	0.84
T ₂	82.6	1867.56	3515.5	108791.2	1.9	4.25
T ₃	84.6	1814.12	3018.4	98391.3	1.7	4.12
T ₄	77.5	1720.64	2781.6	80287.7	1.4	3.26
T ₅	83.2	1787.76	3003.0	89532.1	1.6	3.45
T ₆	81.8	1871.44	3030.1	105131.9	1.8	3.55
T ₇	77.0	1675.8	2872.8	67576.8	1.2	4.01
T ₈	76.0	1710.54	2793.0	58910.2	1.0	3.85
CV %	4.243	4.5	8.9	17.281	16.755	3.24
SEm	1.96	46.13	151.28	8281.89	0.14	0.16
CD @5%	6.00	141.27	463.31	25363.92	0.44	0.51

Conclusion

This study highlights the effectiveness of liquid biofertilizers, particularly potassium-solubilizing bacteria (KSB) and *Rhizobium*, in enhancing growth, yield, and seed quality of soybean variety JS 20-116 under rainfed conditions. *Rhizobium* significantly improved emergence, nodulation, and seed germination, while KSB enhanced plant height, pod number, grain yield, and seedling vigor due to improved nutrient availability. Although the state-recommended chemical package recorded the highest yield (25.5 q/ha), KSB (25.0 q/ha) and *Rhizobium* (23.9 q/ha) provided comparable performance with reduced input costs, resulting in high net returns and benefit-cost ratios. Biofertilizer-treated seeds also showed higher ACC-reductase activity, indicating improved stress tolerance and seedling establishment. These findings confirm that liquid biofertilizers can serve as sustainable and cost-effective alternatives or supplements to chemical fertilizers. Their use not only supports higher productivity and seed quality but also promotes environmental sustainability and soil health. Integration of KSB and *Rhizobium* into soybean production can help reduce fertilizer dependency and strengthen organic and low-input farming systems. Overall, liquid biofertilizers offer a practical solution for improving soybean cultivation in rainfed agro-ecosystems, ensuring profitability and resilience in the face of changing climatic conditions.

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