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Harnessing multifunctional *Bacillus* sp. as bioinoculant for sustainable brinjal (*Solanum melongena*) production

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Abstract

The study evaluated the effect of an indigenous *Bacillus* sp. NITR-3, isolated from the aerobic rice ecosystem of ICAR-IARI, New Delhi and was initially characterized for nitrous oxide reducing capacity and later on explored with multiple plant growth promoting traits, on growth, yield, fruit quality and rhizosphere biological properties of two contrasting brinjal (*Solanum melongena*) varieties, Pusa Uttam and Pusa Shyamla, under pot conditions. The plants received recommended dose of fertilizers (RDF) supplemented with *Bacillus* sp. (NITR-3) inoculation significantly increased individual fruit weight and total fruit number in both varieties compared with RDF alone. Fruit quality response was variety dependent, with Pusa Shyamla exhibiting significantly higher total phenolic contents following bacterial inoculation, whereas these parameters remained statistically unchanged in Pusa Uttam. The simultaneous improvement in crop productivity, antioxidant quality and soil biological health demonstrates the multifunctional nature of the bacterial isolate. Given its previously established nutrient solubilizing ability, phytohormone production, siderophore production and nitrous oxide reduction capacity, *Bacillus* sp. NITR-3 represents a promising bioinoculant for improving yield in brinjal. The study highlights the importance of variety specific responses to PGPR and supports the use of indigenous beneficial microorganisms to enhance vegetable productivity while promoting sustainable soil health.

Key words: *Bacillus* sp.; Brinjal; PGPR; Phenolics; Phosphate solubilizing bacteria; Potassium solubilizing bacteria; Soil health

Brinjal (*Solanum melongena* L.) is one of the most important Solanaceous vegetable crops cultivated throughout tropical and subtropical regions. Due to its wide adaptive nature, year-round cultivation and high economic value, it plays an important role in food and nutritional security (Tomar and Saha, 2018). Brinjal fruits are rich in dietary fibre, vitamins, minerals, phenolic compounds, flavonoids, anthocyanins and other antioxidant that contribute to human health (Daunay and Hazra, 2012; Nandi *et al.,* 2021; Lyngdoh *et al.,* 2025).

Continuous dependence on mineral fertilizers has increased crop productivity but also declined nutrient use efficiency, nutrient imbalance, reduced soil biological activity and environmental concerns associated with nutrient losses (Ladha *et al.,* 2022). Consequently, integrating beneficial microorganisms with recommended dose of fertilizers (RDF) has emerged as an effective strategy for improving crop productivity while maintaining soil health.

Among plant growth-promoting rhizobacteria (PGPR), *Bacillus* spp. are considered as one of the most promising microbial inoculants due to their ability to form stress-resistant endospores and survive under diverse environmental conditions. These bacteria promote plant growth through multiple mechanisms, including phosphate and potassium solubilization, phytohormone production, siderophore secretion, biological nitrogen fixation, biofilm formation and suppression of phytopathogens, thereby enhancing nutrient acquisition, plant growth and stress tolerance (Backer *et al.,* 2018; Aloo *et al.,* 2019). Integration of *Bacillus*-based bioinoculants with mineral fertilizers has also been reported to improve fertilizer use efficiency, soil microbial activity, and crop productivity (Adesemoye *et al.,* 2009). Therefore, study evaluated an innovative multifunctional *Bacillus* sp. based bioinoculant technology integrated with RDF for improving the growth, yield, fruit quality and soil health of two contrasting brinjal varieties under pot conditions.

MATERIALS AND METHODS

A pot experiment was conducted during the Kharif season of 2021 at ICAR–Indian Agricultural Research Institute (IARI), New Delhi, India (28°38′ N, 77°10′ E; 228 m amsl). The experimental soil (0-15 cm) was sandy loam with pH 7.3, EC 0.28 dS m^{-1}, organic carbon 0.48%, available N 235 kg ha^{-1}, available P 18.6 kg ha^{-1}, and available K 265 kg ha^{-1}. Soil analyses were performed following standard methods (Jackson, 1973; Subbiah and Asija, 1956).

The experiment was laid out in a completely randomized design (CRD) with two brinjal varieties, Pusa Uttam (purple round fruited) and Pusa Shyamla (purple long fruited), (Figure 1) two treatments and three replications. Plastic pots (30 cm diameter) containing

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10 kg of soil were used. Treatments comprised T_1: Recommended dose of fertilizers (RDF) and T_2: RDF + *Bacillus* sp. NITR-3. Nitrogen was applied in split doses, while phosphorus and potassium were applied at basal through urea, single superphosphate and muriate of potash, respectively.

The plant growth-promoting isolate NITR-3 was obtained from the rhizosphere of aerobic rice at ICAR-IARI and identified as *Bacillus* sp. by 16S rRNA gene sequencing (27F/1492R primers) followed by BLAST analysis against the NCBI GenBank database). The inoculum was prepared in nutrient broth to approximately 10^8 CFU mL^{-1} ($OD_{600} \approx 0.8–1.0$). Seedlings were root-dip inoculated for 30 min before transplanting, followed by application of 100 mL bacterial suspension as a soil drench around the root zone. Control plants received sterile distilled water.

Bacillus sp. NITR-3 was evaluated for IAA production, phosphate, potassium and zinc solubilization, siderophore production, ammonia production, HCN production, ACC deaminase activity and nitrous oxide (N_2O) reduction potential using standard microbiological protocols (Pikovskaya, 1948; Schwyn and Neilands, 1987; Penrose and Glick, 2003).

Standard crop management practices were followed throughout the experiment. Growth parameters (plant height, plant spread, leaf length and primary branches), yield attributes (fruit length, diameter, fruit index, fruits $plant^{-1}$, single fruit weight, yield $plant^{-1}$ and yield ha^{-1}), fruit quality traits (total phenolics and anthocyanins), and post-harvest rhizosphere biological properties [phosphate-solubilizing bacteria (PSB) and potassium-solubilizing bacteria (KSB)] were determined using standard procedures.

Completely Randomized Design (CRD) was followed and the experimental data were analyzed for variance (ANOVA). Treatment means were compared using the least significant difference (LSD) test at the 5% probability level. Statistical analyses were performed using R.

RESULTS AND DISCUSSION

Application of *Bacillus* sp. NITR-3 in combination with the recommended dose of fertilizers (RDF) positively influenced vegetative growth, fruit characteristics and yield of both brinjal varieties (Tables 1 and 2). Pusa Uttam exhibited greater vegetative growth than Pusa Shyamla, recording higher plant height, plant spread and larger leaves under both treatments. Bacillus inoculation significantly increased plant spread and the number of primary branches in both varieties, whereas improvements in plant height and leaf length were not significant. Fruit characteristics also improved following microbial inoculation.

In Pusa Uttam, single fruit weight increased from 173.67 to 186.33 g, fruit length from 10.67 to 13.67 cm, and fruit diameter from 8.33 to 11.00 cm. Similarly, in Pusa Shyamla, single fruit weight increased from 90.67 to 98.33 g, fruit length from 17.67 to 20.00 cm and fruit diameter from 4.67 to 6.67 cm. Fruit index remained statistically unchanged, although a slight reduction in Pusa Shyamla indicated proportionally greater fruit thickening after inoculation. *Bacillus* application also significantly enhanced reproductive performance, increasing the number of fruits per plant from 9.33 to 11.33 in Pusa Uttam and from 16.00 to 23.00 in Pusa Shyamla, resulting in corresponding increases in yield per plant from 1.49 to 1.68 kg and 1.80 to 2.07 kg, respectively.

Consequently, marketable fruit yield increased significantly in both varieties, from 31.22 to 35.35 t ha^{-1} (13.23%) in Pusa Uttam and from 37.80 to 43.40 t ha^{-1} (14.81%) in Pusa Shyamla. Among all treatments, Pusa Shyamla inoculated with *Bacillus* sp. + RDF recorded the highest yield (43.40 t ha^{-1}). Although Pusa Shyamla was superior in overall productivity, Pusa Uttam maintained its characteristic larger, round fruits, indicating that both cultivars responded positively to Bacillus inoculation while retaining their distinct market preferred fruit types (Fig. 1).

Comprehensive radar plot illustrating the comparative performance of the two contrasting brinjal varieties under RDF alone (T_1) and RDF + Bacillus sp. NITR-3 (T_2) across multiple vegetative and yield-related traits (Fig. 2). In both varieties, inoculation with Bacillus sp. NITR-3 expanded the radar profile, indicating an

Table 1: Effect of *Bacillus* sp. NITR-3 inoculation on vegetative growth parameters of two brinjal varieties. [Values are presented as mean ± standard error of the mean (SeM) based on three replications].

Variety	Treatment	Plant height (cm)	Plant spread (cm)	Leaf length (cm)	Primary branches ($plant^{-1}$)
Pusa Uttam	RDF (T_1)	93.67 ± 2.75	142.33 ± 3.75	13.33 ± 0.76	7.33 ± 0.29
Pusa Uttam	RDF + *Bacillus* (T_2)	96.67 ± 3.82*ns*	148.33 ± 5.20*	14.00 ± 0.50*ns*	9.33 ± 0.29*
Pusa Shyamla	RDF (T_1)	63.00 ± 4.09	89.00 ± 1.80	13.67 ± 0.29	9.67 ± 0.76
Pusa Shyamla	RDF + *Bacillus* (T_2)	65.67 ± 2.57*ns*	93.33 ± 3.82*	14.33 ± 0.29*ns*	12.33 ± 0.29*
LSD(P≤0.05)		12.71	1.88	1.8	1.72
CV (%)		8.46	6.5	7.23	9.4

Table 2: Effect of *Bacillus* inoculation on growth, yield attributes and fruit yield of two brinjal varieties. [Values are presented as mean ± standard error of the mean (SeM) based on three replications].

Variety	Treatment	Fruit length (cm)	Fruit diameter (cm)	Fruit index	Fruits plant^{-1}	Single fruit weight (g)	Yield plant^{-1} (kg)	Yield (t ha^{-1})
Pusa Uttam	RDF (T_1)	10.67 ± 0.58	8.33 ± 0.29	1.29 ± 0.10	9.33 ± 0.58	173.67 ± 4.04	1.49 ± 0.04	31.22 ± 0.85
Pusa Uttam	RDF + *Bacillus* (T_2)	13.67 ± 0.58*	11.00 ± 0.87*	1.26 ± 0.09*ns*	11.33 ± 0.76*	186.33 ± 4.65*	1.68 ± 0.04*	35.35 ± 0.80*
Pusa Shyamla	RDF (T_1)	17.67 ± 1.26	4.67 ± 0.29	3.87 ± 0.51ns	16.00 ± 0.50	90.67 ± 2.02	1.80 ± 0.05	37.80 ± 1.05
Pusa Shyamla	RDF + *Bacillus* (T_2)	20.00 ± 1.00*	6.67 ± 0.29*	3.03 ± 0.28*ns*	23.00 ± 1.50*	98.33 ± 1.76*	2.07 ± 0.03*	43.40 ± 0.61*
LSD($P \leq 0.05$)		3.39	1.88	1.13	3.48	12.64	0.151	3.17
CV (%)		11.63	13.04	25.48	12.39	4.9	4.56	4.56

overall improvement in plant growth and productivity compared with RDF alone. The enhancement was particularly evident for plant height, plant spread, number of primary branches, average fruit weight, fruit length, number of fruits per plant, yield per plant and yield per hectare. Pusa Uttam exhibited superior plant stature and larger fruit size, whereas Pusa Shyamla produced a greater number of fruits and higher overall yield. The radar plot further demonstrates that Bacillus sp. NITR-3 positively influenced multiple agronomic traits simultaneously without adversely affecting variety specific fruit characteristics, highlighting its potential as an effective plant growth promoting inoculant for sustainable brinjal production.

Fig. 1: Fruits harvested from *Bacillus* sp. applied along with RDF for both the variety

The total phenolic content of brinjal fruits was significantly influenced by both cultivar and *Bacillus* sp. NITR-3 inoculation (Fig. 3). Pusa Shyamla consistently recorded higher phenolic content than Pusa Uttam, indicating inherent genotypic differences in antioxidant metabolite accumulation. In Pusa Uttam, total phenolic content remained statistically at par between RDF (T_1) and RDF + *Bacillus* (T_2) ($P > 0.05$), whereas Pusa Shyamla exhibited a significant increase from 2.38 to 2.70 mg GAE g^{-1} fresh weight following bacterial inoculation ($P \leq 0.05$). The enhanced phenolic accumulation in Pusa Shyamla may be attributed to improved nutrient acquisition and phytohormone mediated activation of the phenylpropanoid pathway, resulting in greater synthesis of antioxidant compounds (Bhattacharyya and Jha, 2012; Backer *et al.*, 2018). The absence of a significant response in Pusa Uttam suggests that PGPR-

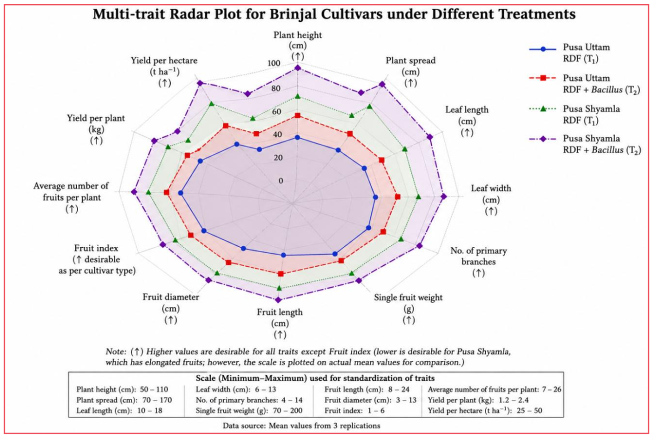

Fig. 2: Integrated visualization of growth responses of brinjal varieties to *Bacillus* based biofertilizer using a multi-trait radar plot.

mediated regulation of secondary metabolism is genotype dependent. Importantly, the increased phenolic content in Pusa Shyamla was accompanied by higher fruit yield, indicating that *Bacillus* sp. NITR-3 simultaneously improved both productivity and nutritional quality without compromising crop performance.

Inoculation with *Bacillus* sp. integrated with the recommended dose of fertilizers (RDF) significantly improved vegetative growth, fruit characteristics and yield of both brinjal varieties. The enhanced plant growth is attributed to the multifunctional plant growth promoting traits of *Bacillus*, including phytohormone production, nutrient solubilization and improved nutrient uptake, which collectively stimulate root development and biomass accumulation (Glick, 2012).

Bacillus inoculation also increased fruit length, diameter, single fruit weight and fruit yield, indicating improved assimilate partitioning and reproductive efficiency. Enhanced phosphorus and potassium availability together with microbial growth regulators likely promoted fruit development and sink strength (Bhattacharyya and Jha, 2012). While Pusa Uttam produced larger fruits, Pusa Shyamla exhibited greater increases in fruit number and overall productivity, suggesting genotype-dependent responses to microbial inoculation (Lucy *et al.*, 2004).

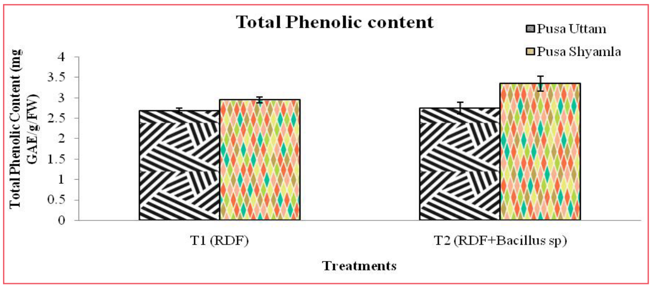

Fig. 3: Influence of *Bacillus* sp. NITR-3 on total phenolic content of fruits in contrasting brinjal varieties.

The significant yield improvement observed under RDF + *Bacillus* resulted from the combined increase in fruit number and fruit weight, reflecting enhanced nutrient use efficiency and rhizosphere functioning. These findings agree with previous reports demonstrating that *Bacillus*-based bioinoculants improve crop productivity by enhancing nutrient acquisition and overall plant performance (Backer *et al.,* 2018). Overall, integration of multifunctional *Bacillus* sp. with RDF represents a promising and sustainable nutrient management strategy for improving brinjal productivity while reducing dependence on chemical fertilizers.

CONCLUSION

The study demonstrated that *Bacillus* sp. NITR-3, when integrated with recommended dose of fertilizers, significantly enhanced brinjal growth, yield, fruit quality, and rhizosphere biological activity under pot conditions. Bacillus inoculation increased fruit number, single fruit weight, and marketable yield in both Pusa Uttam and Pusa Shyamla, while significantly enhancing total phenolic content only in Pusa Shyamla, indicating genotype-dependent improvement in antioxidant quality. The inoculated treatments also recorded higher populations of phosphate- and potassium-solubilizing bacteria, reflecting improved rhizosphere functioning and nutrient cycling.

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