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Cultivar's Response to Rhizobium and Phosphorus Solubilizing Bacteria for Nodulation, Growth and Yield in Garden Pea (*Pisum sativum* var. *Hortense*)

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ABSTRACT

Background: The research, conducted throughout the 2019-20 period, aims to investigate the response of garden pea cultivars to rhizobium and phosphorus solubilizing bacteria (PSB) in terms of nodulation, growth and yield. The objectives included examining the influence of rhizobium and PSB on nodulation ability and studying the yield response of different cultivars under co-inoculation. Additionally, the research attempted to investigate nitrogen and phosphate uptake in several garden pea cultivars.

Methods: The experiment utilized a Split-Split plot design, with main plots comprising five pea cultivars (P-89, AP-3, Bonneville, Arka Kartliik and Arka Apoorva), sub-plots with rhizobium inoculationand sub-sub plots with PSB inoculations. Each variety had four treatments: Uninoculated control, rhizobium, PSB and rhizobium+PSB.

Result: In terms of growth and yield metrics, AP-3 exhibited the quickest days to 50% flowering, whereas Arka Apoorva displayed the tallest plant height (109.93 cm). P-89 stood out in yield characteristics, including pod weight (7.34 g), number of pods per plant (41.99) and green pod yield (126.75 q/ha). Rhizobium and PSB co-inoculations were found to have a considerable impact on yield gains. Regarding quality metrics, Bonneville pods had the greatest TSS content (16.90 Brix) and AP-3 had the highest dry matter content (23.60%). Nodulation experiments found that P-89 had the maximum number of nodules per plant (75.56) and demonstrated the highest uptake of both nitrogen and phosphorus. Overall, the dual inoculation of Rhizobium + PSB outperformed single inoculation and the uninoculated control in all recorded parameters.

Key words: Garden pea, Nitrogen, Nodulation, Phosphorus, PSB, Rhizobium, Varieties.

INTRODUCTION

Peas (*Pisum sativum* L.) is a widely cultivated leguminous vegetable, thriving particularly in the cool seasons of tropical and subtropical regions. Rich in digestible protein (about 25%), essential amino acidsand vitamins such as A, B and C, peas are a key component of agricultural systems. India produced 5,692 thousand metric tons of peas during 2021-22, maintaining its importance as a significant crop (FAO, 2022). As a legume, peas have a remarkable ability to fix atmospheric nitrogen through a symbiotic relationship with the rhizobial bacterium *Rhizobium leguminosarum*, which forms nodules on the roots of the plants (Nadeem *et al*., 2022). This nitrogen-fixing capability enhances the protein content of the plants and improves soil fertility for subsequent crops (Sivasakthi *et al*., 2020).

The symbiosis between peas and *Rhizobium* bacteria can supply 80-90% of the crop's nitrogen requirements, leading to a 10-15% increase in yield when inoculation is used (Jaiswal *et al*., 2021). Efficient seed inoculation with *Rhizobium* not only improves nodulation but also optimizes root architecture through enhanced colonization and nitrogen fixation, contributing to greater crop productivity (Gupta *et al*., 2020).

Phosphorus, another essential nutrient, is often present in unavailable forms in the soil, but phosphorus-solubilizing bacteria (PSB) can convert both organic and

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inorganic forms into plant-accessible forms (Rodríguez *et al*., 2019). The combined inoculation of *Rhizobium* and PSB improves nutrient balance, making phosphorus more accessible to the pea plants and ensuring better growth and yield (Perveen *et al*., 2021).

In regulating the yield and quality of pea crops, both variety selection and nutrient management play crucial roles. Since different pea varieties exhibit varied nutrient demands, agronomic recommendations should be customized to the specific requirements of each variety (Singh *et al*., 2020).

MATERIALS AND METHODS

The experiment was conducted during the *Rabi* season of 2019-20 at the Research Farm of the Division of Vegetable Science, located at Sher-e-Kashmir University of Agricultural Science and Technology in Chatha, Jammu. The location is in Jammu and Kashmir's zone V, which has a subtropical climate. It is located at 32.67° N latitude and 74.88° E longitude. Chatha has arid summers and frigid winters, with temperatures fluctuating between 5.1R°C to 30.5R°C. The average precipitation during the period of plant growth amounts to 16.85 mm, predominantly originating from southwest monsoons. The experiment consisted of five different garden pea cultivars P-89 (V_1), AP-3 (V_2), Bonneville (V_3), Arka Karthik (V_4) and Arka Apoorva (V_5) as the main plot treatment, with Rhizobium (Un-inoculated control (R_0) and Inoculated (R_1) as subplot treatment and PSB inoculations (Un-inoculated (P_0) and Inoculated (P_1) as the sub-subplot treatment. The experimental design employed a split-split plot layout with three replications. Each plot had dimensions of 2 m ×1.4 m and was spaced at intervals of 40 cm × 10 cm. The pea seeds were treated with *Rhizobium leguminosarum* and PSB by incorporating them into a binder made from rice gruel. Using the uniform application of standard crop management techniques SKUAST-J, 2019, sowing took place during the second fortnight of October. Treatments were implemented by applying organic fertilizers (FYM and vermicompost) to the prepared plots. The cultural operations adhered to the suggested guidelines. Irrigation was applied by flood method at 10–15 day intervals, with a total of nine irrigations over the agricultural season. Harvest readiness was judged by the shift of pods from dark green to light green, loaded with grains. The data on various horticultural traits, days to 50% flowering, plant height (cm), leaf area index, no. of pods/plant, pod weight (g), shelling percentage (%), green pod yield (%), dry matter content (%), TSS content (ºBrix), number of nodules/plants, size of nodules, nodule fresh weight (mg/plant), nodule dry weight (mg/plant), nitrogen studies and phosphorus uptake (Jackson 1973b).

RESULTS AND DISCUSSION

Growth and yield characteristics

The primary measure of earliness, days to 50% flowering, varies among cultivars. AP-3 (V2) achieves 50% flowering in 54.08 days, the earliest, while Arka Karthik (V4) takes 80.66 days, similar to Bonneville and Arka Apoorva (Table 1). rhizobium inoculation reduces the flowering time to 72.83 daysand PSB inoculation reduces it to 72.53 days, compared to uninoculated controls at 74.36 and 74.66 days, respectively. Plant height varies by variety and inoculation; Arka Apoorva (V5) is the tallest at 109.93 cm, while P-89 (V1) is the shortest at 98.27 cm. Rhizobium-inoculated plants reach 107.84 cm versus 102.58 cm in controlsand PSB-inoculated plants show similar improvements. Arka Apoorva and Arka Karthik reach heights of 113.20 cm and

Table 1: Effect of Variety, Rhizobium and PSB on Days to 50 % flowering, Plant height (cm), LAI, Pods per plant, Pod weight (g), Shelling percentage (%), Green pod yield (q/ha), TSS of pods (°Brix) and DMC of pods (%).

Treatment		Days to 50% flowering	Plant height (cm)	LAI	Pods per plant	Pod weight (g)	Shelling percentage (%)	Green pod yield (q/ha)	TSS of pods (°Brix)	DMC of pods (%)
Variety	V_1	73.58	98.27	0.59	41.99	7.34	51.01	126.75	16.09	22.47
	V_2	54.08	105.49	0.58	32.27	6.33	50.77	93.39	14.82	23.60
	V_3	79.91	106.09	0.55	27.83	5.80	42.76	84.51	16.90	23.43
	V_4	80.66	106.25	0.55	26.29	4.65	47.95	96.96	16.14	23.46
	V_5	79.75	109.93	0.52	16.67	4.45	47.47	77.27	12.40	22.32
	C.D (0.05)	1.01	2.66	N.S	1.47	0.23	0.64	1.44	0.22	0.50
	S.E (m) ±	0.31	0.81	0.01	0.45	0.07	0.19	0.44	0.06	0.15
Rhizobium	R_0	74.36	102.58	0.50	27.93	5.41	47.46	92.47	14.99	22.67
	R_1	72.83	107.84	0.61	30.09	6.02	48.53	99.08	15.55	23.40
	C.D (0.05)	0.46	1.95	0.03	0.68	0.12	0.23	0.57	0.19	0.23
	S.E (m)±	0.14	0.62	0.01	0.22	0.04	0.07	0.18	0.06	0.07
PSB	P_0	74.66	104.39	0.53	27.46	5.48	47.32	93.83	15.13	22.80
	P_1	72.53	106.02	0.58	30.56	5.94	48.67	97.72	15.41	23.27
	C.D (0.05)	0.54	1.42	0.02	0.48	0.14	0.31	0.76	0.15	0.23
	S.E (m) ±	0.18	0.48	0.01	0.16	0.04	0.10	0.25	0.05	0.08

112.01 cm with rhizobium, while P-89 measures only 100.29 cm. Similarly, PSB inoculation had a significant effect compared to the uninoculated control. The statistical study indicates considerable changes in plant height, influenced by cultivars and their inoculation with rhizobium and phosphorus solubilizing bacteria. Arka Apoorva shows out as the tallest, while P-89 is the lowest, with genetic makeup and nutrient application playing key roles in plant height. This accords with studies by Mandloi *et al*. (2020), Das *et al*. (2015), Patel *et al*. (2013) and Prasad *et al*. (2014). Kumawat *et al*. (2010) also observed the importance of phosphorus in nodule development and microbial activity, contributing to higher plant height in gram.

Varieties do not considerably impact the leaf area index. Rhizobium inoculation, however, considerably increases the leaf area index, reaching 0.61 compared to the uninoculated control (0.50). Similarly, PSB-inoculated seeds result in a considerably greater leaf area index (0.58) than the uninoculated control (0.53). Most cultivars, having viney stature and comparable expansion tendencies, do not differ in leaf area index. Yet, rhizobium and PSB inoculations raise LAI due to higher nutrient digestion and increased microbial activity, supporting effective nodule formation in the soil (Das *et al*., 2015).

Varieties significantly affect pod quantity and weight. P-89 has the highest pod count (41.99), while Arka Apoorva has the lowest (16.67). Rhizobium inoculation increases pod number to 30.09 compared to 27.93 in uninoculated seedsand PSB inoculation raises it to 30.56 compared to 27.46 in controls (Jaiswal *et al*., 2021; Nadeem *et al*., 2022). Similarly, P-89 shows the highest pod count with PSB inoculation, while Arka Apoorva records the lowest. Dual inoculation enhances pod quantity due to increased nitrogen and phosphorus availability, aligning with recent studies (Gupta *et al*., 2020; Perveen *et al*., 2021).

In terms of pod weight, P-89 leads at 7.34 g, while Arka Apoorva records the lowest at 4.45 g, similar to Arka Karthik (4.65 g). Rhizobium inoculation raises pod weight to 6.02 g compared to 5.41 g in controlsand PSB inoculation results in 5.94 g versus 5.48 g in uninoculated seeds (Singh *et al*., 2020; Rodríguez *et al*., 2019). P-89 shows the highest weight with both rhizobium (7.62 g) and PSB (7.35 g) inoculation. Varietal differences, soil interactionand climatic factors contribute to this variability. These findings are consistent with recent research on the benefits of inoculation for improving pod weight (Sivasakthi *et al*., 2020; Kumar *et al*., 2022). Considerable variation in shelling percentage is observed among varieties. P-89 leads with 51.01%, similar to AP-3 at 50.77%, while Bonneville has the lowest at 42.76% (Singh *et al*., 2022). Rhizobium-inoculated seeds exhibit a higher shelling percentage (48.53%) compared to uninoculated seeds (47.46%) and PSB-inoculated seeds show an even greater percentage (48.67%) than controls (47.32%) (Rao *et al*., 2021; Kumar *et al*., 2022). Varietal differences are significant, with Bonneville recording the lowest shelling percentage. Single and dual inoculation with Rhizobium and PSB substantially enhance shelling percentage compared to controls, aligning with recent findings on the positive effects of biofertilizers (Khan *et al*., 2023; Sharma *et al*., 2024).

Green pod yield (q/ha)

Data from Table 1 indicate significant varietal effects on green pod production. P-89 leads with the highest yield (126.75 q/ha), while Arka Apoorva shows the lowest (77.27 q/ha) (Singh *et al*., 2022). Rhizobium inoculation substantially increases yield to 99.08 q/ha compared to uninoculated controls. Similarly, PSB inoculation improves yield, with P-89 reaching 130.79 q/ha, significantly higher than other varieties. Dual inoculation of rhizobium and PSB results in the highest yield for P-89 (134.42 q/ha), while Arka Apoorva yields the least (70.95 q/ha) when uninoculated (Rao *et al*., 2021; Kumar *et al*., 2022). Inoculation with rhizobium and PSB enhances green pod production compared to controls, reflecting findings by Yadav and Yadav (2011), Vimala and Natrajan (2000)and Bhattari *et al*. (2003). The increase in pod yield is attributed to improved nutrient availability during pod development, as noted by Habib and Zamin (2003), Dalal and Nandkar (2010) and Shamad *et al*. (2019).

Quality characteristics

The results in Table 1 show significant varietal effects on total soluble solids (TSS) and dry matter content. Bonneville leads with the highest TSS at 16.90 °Brix, while Arka Apoorva has the lowest at 12.40 °Brix (Patel *et al*., 2024). Rhizobium and PSB inoculations both significantly increase TSS compared to uninoculated seeds. Rhizobium enhances TSS by improving nitrogen fixation and chlorophyll production, whereas PSB increases phosphorus availability, promoting root development and nodulation. Dual inoculation of rhizobium and PSB achieves the highest TSS due to synergistic effects on nutrient availability and nitrogen fixation (Singh *et al*., 2021; Kumar *et al*., 2022). For dry matter content, AP-3 has the highest at 23.60%, followed by Bonneville at 23.43% and Arka Karthik at 23.46%. Arka Apoorva records the lowest at 22.32%, similar to P-89 at 22.47%. Both rhizobium and PSB inoculations significantly boost dry matter content, likely due to enhanced cell division and meristematic activity (Verma *et al*., 2023; Sharma *et al*., 2022).

Nodulation studies

The data in Table 2 highlights significant varietal differences in root nodule counts, length, widthand weight. P-89 shows the highest nodule count (75.56), significantly more than Arka Apoorva (63.37) (Rani *et al*., 2020). Rhizobium inoculation boosts nodule counts to 82.24, with P-89 reaching 87.85, while PSB inoculation increases counts to 78.51, with P-89 at 84.99. Dual inoculation results in the highest nodule count (93.91), surpassing the uninoculated control (50.56) (Rather *et al*., 2021; Srivastava *et al*., 2022). Nodule length does not vary significantly among varieties as shown in Table 2, Fig 1-15. Rhizobium-inoculated seeds produce longer nodules (2.10 mm) compared to uninoculated controls (1.77 mm)and PSB inoculation also increases nodule length (2.03 mm vs. 1.84 mm) (Singh *et al*., 2021). Nodule width varies, with Bonneville having the widest nodules (1.65 mm), while P-89 has the narrowest (1.27 mm) with rhizobium and PSB inoculation increasing width (1.53 mm and 1.44 mm, respectively) (Singh *et al*., 2021).

Fig 1: Root noules of Variety P-89 (Uninoculated).

Fig 2: Root nodules of variety P-89 (Rhizobium Inoculated).

Fig 3: Roots nodules of variety P-89 (PSB Inoculated).

Fig 4: Root nodules of variety AP-3 (Uninoculated).

Table 2: Effect of variety, rhizobium and PSB on number of nodules per plant, size of nodules (length) mm, size of nodules (width) mm, fresh weight of nodules (mg), dry weight of nodules(mg), nitrogen uptake (kg/ha) and phosphorus uptake (kg/ha)

Treatment		Number of nodules per plant	Size of nodules (length) mm	Size of nodules (width) mm	Fresh weight of nodules (mg)	Dry weight of nodules (mg)	Nitrogen uptake (kg/ha)	Phosphorus uptake (kg/ha)
Variety	V_1	75.56	1.95	1.26	388.10	123.16	93.23	13.19
	V_2	71.11	1.99	1.50	396.37	121.62	91.47	12.97
	V_3	68.64	1.88	1.65	377.38	117.06	90.16	12.88
	V_4	69.01	1.89	1.31	382.00	118.50	87.40	12.56
	V_5	63.37	1.96	1.27	383.47	118.22	88.57	12.07
	C.D (0.05)	1.54	N.S	0.14	6.00	3.36	1.03	0.20
	S.E (m) ±	0.47	0.07	0.04	1.84	1.03	0.31	0.06
Rhizobium	R_0	56.84	1.77	1.26	371.10	114.11	83.08	11.69
	R_1	82.24	2.10	1.53	399.83	125.32	97.25	13.77
	C.D (0.05)	0.76	0.20	0.11	6.67	2.86	0.81	0.23
	S.E (m)±	0.24	0.06	0.03	2.12	0.90	0.25	0.07
PSB	P_0	60.56	1.84	1.35	377.58	116.12	83.72	11.37
	P_1	78.51	2.03	1.44	393.35	123.31	96.61	14.09
	C.D (0.05)	1.02	0.13	0.09	5.48	2.97	0.66	0.21
	S.E (m) ±	0.34	0.05	0.03	1.86	1.01	0.22	0.07

Fig 5: Root nodules of variety AP-3 (Rhizobium inoculated).

Fig 6: Root nodules of variety AP-3 (PSB inoculated).

Fig 7: Root nodules of variety bonneville (Uninoculated).

Fig 8: Root nodules of variety Bonneville (Rhizobium Inoculated).

Fig 9: Root nodules of variety bonnevilla (PSB Inoculated).

Fig 10: Root nodules of variety arka karthik (Uninoculated).

Fig 11: Root nodules of variety arka karthik (Rhizobium inoculated).

Fig 12: Root nodules of variety arka karthik (PSB inoculated).

Fig 13: Root nodules of variety arka apoorva.

Fig 14: Root nodules of variety arka apoorva (Rhizobium inoculated).

Fig 15: Root nodules of variety arka apoorva (PSB inoculated).

Regarding nodule fresh weight, AP-3 has the highest at 396.37 mg, while Bonneville has the lowest at 377.38 mg. Rhizobium inoculation increases fresh weight to 399.83 mgand PSB increases it to 393.35 mg. Dry weight is highest in P-89 (123.16 mg) and AP-3 (121.62 mg), with Bonneville recording the lowest (117.06 mg). Rhizobium and PSB inoculation both enhance dry weight (125.32 mg and 123.31 mg, respectively) compared to uninoculated controls (114.11 mg and 116.12 mg) (Tyagi *et al*., 2022; Rani *et al*., 2020).

Nitrogen and phosphorus uptake studies (Kg/ha)

The data in Table 2 reveals significant varietal differences in nutrient uptake. P-89 shows the highest nitrogen uptake at 93.23 kg/ha, while Arka Karthik has the lowest at 87.40 kg/ha. Rhizobium inoculation increases nitrogen uptake to 97.25 kg/ha compared to 83.08 kg/ha in uninoculated controlsand PSB inoculation improves it to 96.61 kg/ha. Co-inoculation of Rhizobium and PSB results in the highest nitrogen uptake of 103.04 kg/ha, whereas uninoculated seeds have the lowest at 75.99 kg/ha (Rudresh *et al*., 2022; Abid *et al*., 2017).

For phosphorus uptake, P-89 records the highest at 13.19 kg/ha and Arka Apoorva the lowest at 12.07 kg/ha. Rhizobium inoculation increases phosphorus uptake to 13.77 kg/ha compared to 11.69 kg/ha in controlsand PSB inoculation boosts it to 14.09 kg/ha. P-89 shows the greatest phosphorus uptake with PSB (14.67 kg/ha), while Arka Apoorva has the lowest (13.29 kg/ha). Co-inoculation enhances phosphorus uptake to 15.62 kg/ha, with uninoculated seeds showing the least at 10.82 kg/ha (Rokhzadi *et al*., 2019; Wani *et al*., 2021).

Overall, both rhizobium and PSB inoculations improve nutrient uptake across varieties, with P-89 performing the best and Arka Karthik and Arka Apoorva showing lower uptake. Co-inoculation further enhances nutrient absorption, consistent with findings by Rani *et al*. (2019) and Bhat *et al*. (2015).

CONCLUSION

In conclusion, pea production is an important contributor to global agriculture, particularly in tropical and subtropical countries. Peas, which are high in protein and critical vitamins, serve an important role in maintaining food security. The symbiotic interaction between peas and nitrogen-fixing Rhizobium bacteria increases soil fertility and crop yields. Incorporating phosphorus-solubilizing bacteria improves nutrient availability and promotes plant health. Varietal changes have a substantial impact on growth and yield characteristics, with some cultivars excelling in both yield and quality qualities. The study emphasizes the necessity of specific nutrition management strategies for optimizing pea output. Farmers can use these insights to improve productivity and sustainability in pea agriculture, meeting the growing demand for this nutritious crop.

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Conflict of interest

The authors declare no conflicts of interest in relation to this work.

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