

Original Research Article

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Interactive Effect of Bio-regulators and Plant Growth Promoting Bacteria on Yield attributes and Economics of Indian bean (*Lablab purpureus* L. var. *typicus*)

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ABSTRACT

Keywords

Brassinoid, Growth, Indian bean, *Pseudomonas fluorescens*, Quality, *Rhizobium phaseoli*, Salicylic acid and Yield.

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The field experiment was conducted at Horticulture Farm, S.K.N. College of Agriculture, Jobner (Jaipur) during *kharif* season 2016-2017. The experiment consisted of twenty treatment combinations including five bio-regulators (control, brassinoids 0.5 ppm, brassinoids 1.0 ppm, salicylic acid 100 ppm and salicylic acid 150 ppm) and four plant growth promoting bacteria (control, *Rhizobium*, *Pseudomonas* and *Rhizobium* + *Pseudomonas*). They were under taken in randomized block design with three replications. Combined application of brassinoids 1.0 ppm along with *Rhizobium* + *Pseudomonas* inoculation to the seeds of Indian bean significantly increased number of green pods per plant, green pod length (cm), green pod yield per plant, green pod yield per plot, pod yield (88.20 q/ha), net returns (₹88767/ha) and B:C ratio (3.04) as compared to control.

Introduction

Indian bean or Dolichos bean (*Lablab purpureus* L. var. *typicus*) belongs to the family fabaceae (2n=22). It is a multipurpose crop grown for pulse, vegetable and forage. There are two type of cultivated species of Indian bean viz, *Lablab purpureus* var. *typicus* which is vegetable type, cultivated for its soft and edible pods and *Lablab purpureus* var. *lignosus* is the field bean, cultivated for

dry seeds as pulse. The pods of Indian bean are important source of protein, minerals and dietary fibre. Its mature dark coloured seeds contains trypsin inhibitor, which break down into water soluble cyanogenic.

During cooking the purple coloured pods have a strong flavour, which disappears after cooking. The nutritional composition of edible green pods contain 86 percent moisture, 2 percent fibre, 4 percent protein,

7.10 percent carbohydrate, 48 Kcal energy, 68mg phosphorus, 1mg iron, 210mg Ca, 668 IU vitamin-A, 0.08mg thiamine, 0.11mg riboflavin, 0.75mg niacin and 9.3mg vitamin C (Gopalan *et al.*, 2004).

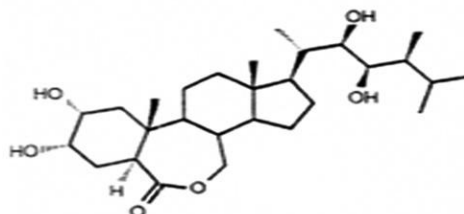
In present study, bio-regulators and PGPB were included as key factors to increase the fertilizer use efficiency as well as to promote/modify the physiological responses in the plants.

The bio-regulators not only regulate the growth of plant species, which play an important role in root induction and growth of plants but also play important roles in DNA replication, cell division, controlling of microgenesis, senescence and resistant to environmental stresses (Kaur-Sawhney *et al.*, 2003).

Among the various bio-regulators, brassinolide is an important steroidal component obtained from pollen grains of *Brassica napus*. It is known to be essential for plant growth and development and is regarded as a new class of plant hormone with a generic name of 'brassinosteroids'. Brassinosteroids are considered as plant hormones with pleiotropic effects as they influence wide array of developmental processes such as seed germination, rhizogenesis, flowering and maturation (Sasse, 1999). Brassinosteroids improve the resistance power in the plants against environmental stresses *viz.*, water stress, salinity stress, low and high temperature

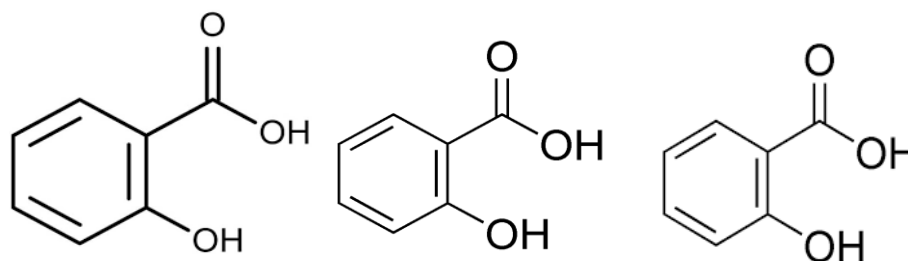
stress (Rao *et al.*, 2002) and crop productivity (Vardhini *et al.*, 2006).

Brassinosteroid is a class of polyhydroxy steroids that have been recognized as a sixth class of plant hormone.



Likewise, Salicylic acid (SA) is also an important substance which is classified as phenolic growth regulator, a non- enzymatic antioxidant, a signalling or messenger molecule to induce responses in the plants to environmental stress. Salicylic acid plays an important role in the regulation of some physiological processes in plants. It has also been found that SA positively affects growth and development, photosynthesis, transpiration, ion uptake, transport, and membrane permeability in the plants (Simaei *et al.*, 2012).

SA is a monohydroxy benzoic acid, a type of phenolic acid and a beta hydroxy acid. It has the formula $C_7H_6O_3$. This colourless crystalline organic acid is widely used in organic synthesis and functions as a plant hormone.



It involves signalling and mediating in plant defence against pathogens by inducing the production of pathogenesis related proteins. It also involved in the systemic acquired resistance (SAR) in which a pathogen attack on one part of the plant induces resistance in other parts. Plant Growth Promoting Bacteria (PGPB) enhance plant growth and productivity. *Rhizobium* is well known biological N fixer and *Pseudomonas* has known for its activity of biological control.

Co-inoculation of *Pseudomonas spp.* and *Rhizobium spp.* have been shown to increase the degree of colonization of the legume rhizobia resulting in enhanced plant nodulation. This tripartite association composed of legume plant and two soil bacteria *i.e.* *Rhizobium spp.* and *Pseudomonas spp.* have been reported to increase root and shoot weight, plant vigour, N fixation and grain yield in legumes. *Pseudomonas fluorescens* is considered most significant phosphate solubilizing bacteria, which not only provide phosphorus to the plants, but also produce siderophore, antibiotic and phytohormones such as indole-acetic acid (Leinhos and Nacek, 1994).

A number of strains of *Pseudomonas fluorescens* suppress plant diseases by protecting the seeds and roots from fungal infection (O'Sullivan and O'Gara, 1992). This effect is the result of production of a number of secondary metabolites including antibiotics, siderophores and hydrogen cyanide. Competitive exclusion of pathogens as the result of rapid colonization of the rhizosphere by *Pseudomonas fluorescens* may also be an important factor in disease control. *Pseudomonas fluorescens* induced accumulation of lignin in pea roots was reported by Benhamou *et al.*, 1996. *Pseudomonas spp.* can form gluconic acid through the oxidative glucose metabolism (Gyaneshwar *et al.*, 2002).

Pseudomonas species have shown to be effective in controlling pathogenic fungi and stimulating plant growth by a variety of mechanisms, including production of siderophores, synthesis of antibiotics, production of phytohormones, enhancement of phosphate uptake by the plant, nitrogen fixation, and synthesis of enzymes that regulate plant ethylene levels (Abdul Jaleel, 2007).

Materials and Methods

The experiment was conducted at Horticulture Farm, S.K.N. College of Agriculture, Jobner (Jaipur) during *Kharif* season 2016-2017. In Rajasthan, this region falls under agro-climatic zone-III A (Semi-Arid Eastern Plains). The experiment consisted of twenty treatment combinations including five bio-regulators (control, brassinoids 0.5 ppm, brassinoids 1.0 ppm, salicylic acid 100 ppm and salicylic acid 150 ppm) and four plant growth promoting bacteria (control, *Rhizobium*, *Pseudomonas* and *Rhizobium + Pseudomonas*).

The experiment was laid out in Randomized Block Design with with three replications. The process of inoculation was preceded by seed treatment with fungicide then seed inoculation with *Rhizobium phaseoli* and *Pseudomonas fluorescens* before sowing by putting seeds in 20 % sucrose solution and then inoculated with @ 10 g/kg of seeds by putting the uniform coating of chalk form powder on seeds and were allowed to air dry in shade. The seeds were sown on the same day after inoculation.

The seeds of control plot treated with sucrose solution only. Brassinoids was sprayed @ 0.5 ppm and 1 ppm at 30 and 45 DAS. Similarly, salicylic acid was also sprayed @ 100 ppm and 150 ppm at 30 and 45 DAS in respective plots. Each plot measured $2.8 \times 1.4 \text{ m}^2$ (4.32

m²) area. The crop geometry was kept at 60 x 30 cm. All the cultural operations were followed which were necessary to raise the good crop. The observations like plant height (cm), number of branches per plant, dry matter accumulation (g/m), CGR at 45-60 and 60-75 (g/m²/day) days after sowing, chlorophyll content in leaves (mg), leaf area (cm²), number of green pods per plant, average pod weight (g), green pod length (cm), number of pickings per plant, green pod yield per plant (g), green pod yield per plot (kg), green pod yield per hectare (q), protein content and crude fibre content taken manually.

CGR was calculated by Radford, 1967 method. chlorophyll content was determined using the method of Arnon (1949) with slight modifications. Nitrogen content in the green pods was estimated by using Nessler's reagent by spectrophotometer method (Snell and Snell, 1949), protein content in the pods was calculated by multiplying nitrogen concentration (%) by the factor 6.25 (A.O.A.C., 1960). Crude fibre content in pods was determined by the method suggested by A.O.A.C. (1960). The data obtained from the trial were subjected to statistical analysis and the results were documented, analysed and presented in tabular form.

Results and Discussion

It is evident from data (Table 1 and Fig 1,2 and 3) that the combined effect of different bio-regulators and plant growth promoting bacteria was noticed significantly increased the yield and yield attributed. Total number of pods (42.10), pod length (9.50 cm), total green pod yield per plant (158.55 g), total green pod yield per plot (3.810 kg) and pod yield 88.20 q/ha were found maximum under treatment B₂P₃ (brassinoids 1.0 ppm along with *Rhizobium* + *Pseudomonas* inoculation) followed by B₁P₃ i.e. brassinoide 0.5 ppm and

Rhizobium + *Pseudomonas*) and minimum under control. The treatment combination B₂P₃ (brassinoids 1.0 ppm and *Rhizobium* + *Pseudomonas*) remained statistically at par with treatment B₁P₃ (brassinoids 0.5 ppm and *Rhizobium* + *Pseudomonas*).

The foliar application of brassinoid increased yield and yield attributes at all levels on crop productivity and photosynthetic activity (Mona *et al.*, 2011). These bio-regulators in general have to increase number of flowers as well as pods on the plants. The flower and pod drop may be reduced to same extent (Ramesh and Thirumuguran, 2001, Sangupta and Tamang, 2015 and Matwa *et al.*, 2017). The increase in yield and yield attributes under foliar spray of brassinoids was also observed by Gojraj Jat *et al.*, (2012) and Choudhary (2017).

These findings are in accordance with the results of Vardhini *et al.*, (1998) who reported in groundnut that brassinolide application increased the total biomass and then might have resulted in an increase in assimilate transport from source to sink and their ultimate conversion into final reserved food. Similar results were also reported by Matwa *et al.*, (2017) and Choudhary (2017).

The beneficial effects of *Rhizobium* as explained earlier thus might have increased the availability of nitrogen and phosphorus alongwith other nutrients which in term resulted in to higher production of assimilates and their partitioning to different reproductive structures such as yield attributes and ultimately, green pod yield. Co-inoculation of legumes with *Rhizobium* and PGPR *Pseudomonas* strains, were able to alleviate salt stress of plants, grown on salt affected soils and increased plant growth, yield and controlled the plant diseases of leguminous plants is recorded by Egamberdieva *et al.*, (2013).

Table.1 Combined effect of bio-regulators and plant growth promoting bacteria on yield attributes of Indian bean

Total number of green pod				
Treatments	P₀	P₁	P₂	P₃
B₀	27.00	29.50	28.10	30.95
B₁	30.35	31.00	29.00	39.50
B₂	37.70	37.10	37.60	42.10
B₃	35.00	36.50	37.70	35.00
B₄	33.00	37.50	37.00	37.90
SEm±	1.44			
CD (P=0.05)	4.11			
Pod length				
Treatments	P₀	P₁	P₂	P₃
B₀	5.00	5.10	5.45	5.50
B₁	5.75	5.70	6.80	9.20
B₂	8.50	8.60	6.80	9.50
B₃	7.65	8.99	8.45	7.95
B₄	7.82	8.40	8.45	8.65
SEm±	0.14			
CD (P=0.05)	0.40			
Total green pod yield per plant (g)				
Treatments	P₀	P₁	P₂	P₃
B₀	68.50	82.33	76.05	93.70
B₁	94.29	105.94	96.10	146.45
B₂	119.14	128.97	126.75	158.77
B₃	107.49	123.31	123.50	128.27
B₄	103.11	128.89	123.32	141.32
SEm±	4.62			
CD (P=0.05)	13.22			
Total green pod yield per plot (kg)				
Treatments	P₀	P₁	P₂	P₃
B₀	1.644	1.976	1.825	2.249
B₁	2.263	2.543	2.307	3.515
B₂	2.859	3.095	3.042	3.810
B₃	2.580	2.959	2.964	3.079
B₄	2.475	3.093	2.960	3.392
SEm±	0.111			
CD (P=0.05)	0.317			
Pod yield (q/ha)				
Treatments	P₀	P₁	P₂	P₃
B₀	38.06	45.74	42.25	52.06
B₁	52.38	58.86	53.39	81.36
B₂	66.19	71.65	70.41	88.20
B₃	59.72	68.51	68.61	71.26
B₄	57.28	71.61	68.51	78.51

SEm±	2.57		
CD (P=0.05)	7.35		
Bio-regulators		Plant Growth Promoting Bacteria	
B ₀ Control		P ₀ Control	
B ₁ Brassinoids 0.5 ppm		P ₁ Rhizobium	
B ₂ Brassinoids 1.0 ppm		P ₂ Pseudomonas	
B ₃ Salicylic acid 100 ppm		P ₃ Rhizobium + Pseudomonas	
B ₄ Salicylic acid 150 ppm			

Table.2 Combined effect of bio-regulators and plant growth promoting bacteria on economic attributes of Indian bean

Net returns (₹/ha)				
Treatments	P ₀	P ₁	P ₂	P ₃
B ₀	13915	25287	20061	34707
B ₁	35367	44926	36737	78538
B ₂	56034	64074	62132	88767
B ₃	46257	59288	59460	63285
B ₄	42532	63864	59231	74081
SEm±	3738			
CD (P=0.05)	10702			
B:C Ratio				
Treatments	P ₀	P ₁	P ₂	P ₃
B ₀	1.32	1.58	1.46	1.80
B ₁	1.82	2.04	1.85	2.81
B ₂	2.30	2.48	2.43	3.04
B ₃	2.07	2.36	2.37	2.45
B ₄	1.98	2.47	2.36	2.70
SEm±	0.09			
CD (P=0.05)	0.27			

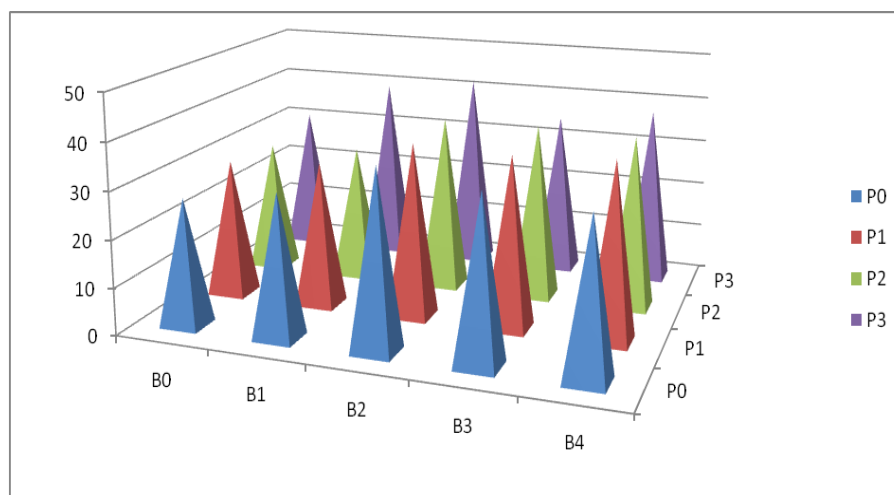


Fig.1 Combined effect of bio-regulators and plant growth promoting bacteria on number of green pods /plant of Indian bean

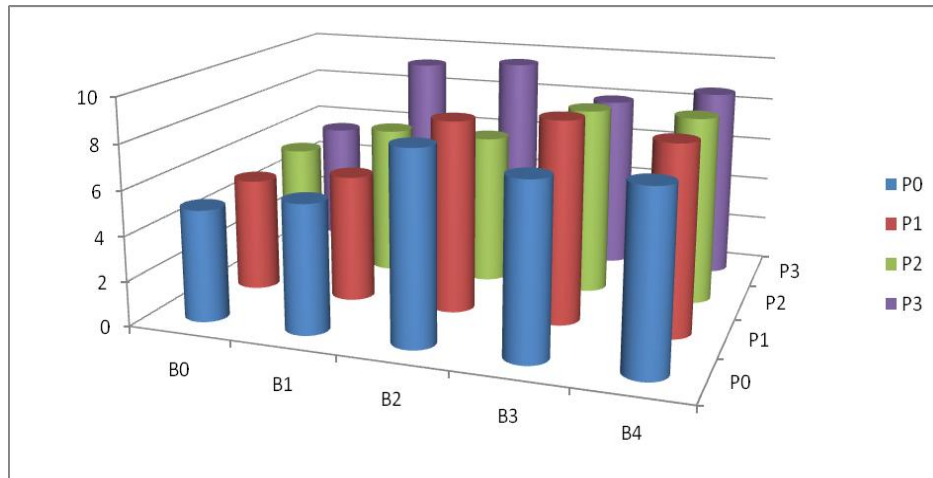


Fig.2 Combined effect of bio-regulators and plant growth promoting bacteria on pod length (cm) of Indian bean

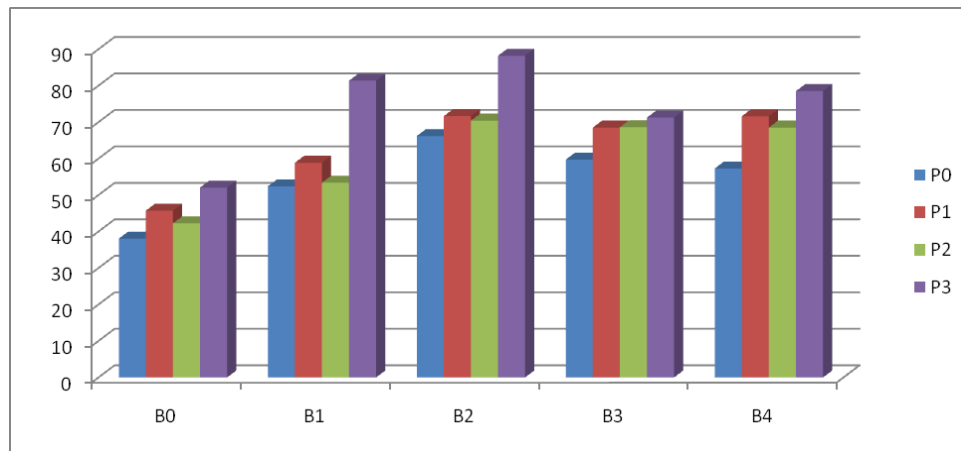


Fig.3 Combined effect of bio-regulators and plant growth promoting bacteria on green pods yield q/ha of Indian bean

Application of phosphate solubilizing microbes *i.e.* *Pseudomonas*, around the roots of plants, in soils and in fertilizers has been shown to release soluble phosphorus, promote plant growth and protect plants from pathogen infection (Biswas *et al.*, 2006, Ouahmane *et al.*, 2007). The production of phosphate enzyme by phosphate solubilize bacteria and microbial phytases activity was reported by Ponmurugan and Gopi, 2006. The plant growth promoting rhizobacteria colonize in roots of plants and promote plant growth and development through activation of phosphate solubilization and promotion of the mineral

nutrient uptake are usually believed to be involved in plant growth promotion and finally in yield (Glick, 1995 and Lalande *et al.*, 1989).

It is also indicated (Table 2) that the higher net returns of green pod (₹88,767/ha) and B:C ratio (3.07) was obtained under the treatment B₂P₃ (brassinoids 1.0 ppm along with *Rhizobium* + *Pseudomonas* inoculation) followed by B₁P₃ *i.e.* brassinoide 0.5 ppm and *Rhizobium* + *Pseudomonas*. The treatment combination B₂P₃ (brassinoid 1.0 ppm and *Rhizobium* + *Pseudomonas*) remained

statistically at par with treatment B₁P₃ (brassinolide 0.5 ppm and *Rhizobium* + *Pseudomonas*).

On the basis of one year experiment results, it may be concluded that the combination of bio-regulators as brassinoids 1.0 ppm and plant growth promoting bacteria as *Rhizobium phaseoli* + *Pseudomonas fluorescens* was found most suitable in terms of comparable green pod yield, net returns and B:C ratio (88.20 q/ha, ₹ 88,767 and 3.07, respectively). Thus, combined application of brassinoids 1.0 ppm and *Rhizobium phaseoli* + *Pseudomonas fluorescens* to Indian bean crop is recommended.

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