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Standardization of Different Pre-Sowing Seed Treatment of *Phosphate Solubilizing Bacteria, Rhizobium, Azospirillum* and *Trichoderma* on growth, yield and yield attributing traits of Frenchbean (*Phaseolus vulgaris* L.) variety Ellora Number 11

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ABSTRACT: The present study was laid out in randomized block design along with three replications in the Field Experimentation Center and Notified State Seed Testing Laboratory, Department of Genetics and Plant Breeding during 2022-23. The study was designed to determine the effect of different doses of *Phosphate Solubilizing Bacteria* (8%, 10% and 12%), *Rhizobium* (8%, 10% and 12%), *Azospirillum* (8%, 10% and 12%) and *Trichoderma* (8%, 10% and 12%) for 6hrs on growth, yield, yield traits and seedling parameters of Frenchbean variety Ellora 11. Morphological data were recorded for field emergence, plant height @30DAS, 60DAS, number of primary branches, days to 50% flowering, number of pods/plant, number seeds/ pod, seed yield/ plot, seed index, biological yield and harvest index. It was found that among all the pre sowing seed treatment, *rhizobium* 12% for 6hrs showed better performance. This study can helpful to reach new horizons of research in the field of seed technology to overcome the germination problem, improving crop stand and yield ultimately.

Keywords: French bean, *Azospirillum, PSB, Rhizobium, Trichoderma* and Morphological parameters

INTRODUCTION

Quality seeds play a major role, along with improved package of practices leading to enhanced productivity. The low productivity under rainfed condition is due to use of poor quality seeds, soil moisture deficit, low and erratic rainfall and improper crop management. Hence, the production of high quality seed is necessary and important to the agricultural industry. Seed treatments are followed to increase seed quality. The concept of seed treatment is use and application of biological and chemical agents that control or contain primary soil and seed borne infestation of insects and diseases which pose devastating consequences to crop production and improving crop safety leading to good establishment of healthy and vigorous plants resulting in better yields. Even with the availability of different types of seed treatments, seed hardening, seed inoculation, seed pelleting and seed priming are mostly used seed treatments. Leguminous species have hard seed coat that not does allow easy germination. Since the seed coat is seen as a hindrance to uniform and rapid germination, there is a need for more radical pretreatments to enhance germination [1].

Bio-fertilizers are microbial inoculants of selective microorganisms like bacteria, algae and fungi already existing in nature. They help in improving soil fertility by the way of biological nitrogen fixation from atmosphere, solubilization of insoluble nutrients already present in the soil, decomposing soil residues stimulating plant growth and production. The process is slow, consumes less energy and provides cheap nutrients to plants without polluting the nature. Bio-fertilizers are low cost, renewable and eco-friendly sources of plant nutrients which supplement chemical fertilizer. They can be used either in seed treatment or soil application. Bio-fertilizers are also ideal inputs for reducing the cost of cultivation and for practicing organic farming [2].

Azospirillum is a great plant growth-promoting rhizobacteria (PGPR). *Azospirillum* inoculation may well obviously increment the development in terms of height; number of leaf and plant; length and breadth of leaf; and new and dry weight/plant of rice plant. Azospirillum is well-known to fix a significant amount of atmospheric nitrogen and arrangements to the crop, improve the fertilizer utilized great organization, soil fertility assessment and ensures fragmentary saving of nitrogenous fertilizer [3].

Trichoderma spp. to produce antibiotics that inhibit growth of pathogenic organisms and used as a bio-control agent [4]. *Trichoderma species* are common soil-borne filamentous fungi, with some strains capable of establishing beneficial relationships with plants [5]. Direct plant growth promotion [6] is one of many mechanisms in which *Trichoderma spp.* enhance plant health, however, the molecular mechanism that underlies this is still unclear, *Trichoderma spp*. induce growth promotion by a fungal auxin -dependent mechanism. Using in vitro bioassay, they showed that *Trichoderma virens* Gv29.8 and *T. atroviride* IMI206040 can synthesize IAA (and some of its derivatives) and have been demonstrated to increase the production of Arabidopsis lateral roots. However, the correlation between IAA synthesis and plant growth promotion of plants in soil-based systems is less convincing, many *Trichoderma* strains are capable of synthesizing IAA but only a few can promote plant growth.

Rhizobium is a bacteria found in soil that helps fixing nitrogen in leguminous plants. It ensures proper nodulation in root zone which lead to adequate Nitrogen uptake. Rhizobium also exhibits antifungal anti-bacterial activities which allow seeds for healthy seedling rise. Plant growth promoting rhizobacteria (PGPR) with 1-aminocyclopropane-1-carboxylic acid (ACC) deaminase activity has the potential to promote plant growth and development under adverse environmental conditions [7].

Phosphorus (P) is one of the essential elements that are necessary for plant development and growth; it makes up about 0.2% of a plant's dry weight. It is second only to nitrogen among mineral nutrients most commonly limiting the growth of crops. Soil phosphorus deficiency is addressed by the application of phosphorus fertilizers. Apart from chemical fertilization, microbial P-solubilization and mineralization is the only possible way to increase plant available phosphorus. Many species of soil fungi and bacteria are able to solubilize phosphorus in vitro and some of them can mobilize phosphorus in plants [8]. They solubilize insoluble inorganic (mineral) phosphorus and mineralize insoluble organic phosphorus [9]. A lot of research work has already been carried on the effects of different pre sowing treatments on seed germination and seedling growth in vegetable crops. Furthermore, research needs to be conducted to standardize the treatments for different environmental conditions. Hence, the present study was undertaken to standardize the doses of different pre-sowing seed treatment of *PSB, Azosprillum, Rhizobium and Trichoderma* on growth, yield and yield attributing traits of Frenchbean in Vindhyan region of Uttar Pradesh.

MATERIALS AND METHODS

Seeds of Frenchbean variety Ellora Number 11 were treated with different doses of *PSB, Rhizobium, Azospirillum and Trichoderma i.e.* T_1 - PSB 12%, T_2 - PSB 10%, T_3 - PSB 8%, T_4 - Azospirillum 12%, T_5 - Azospirillum 10%, T_6 - Azospirillum 8%, T_7 - Rhizobium 12%, T_8 - Rhizobium 10%, T_{9} - Rhizobium 8%, T_{10} - Trichoderma 12%, T_{11} - Trichoderma 10%, T_{12} - Trichoderma 8% for 6 hours duration in Notified State Seed Testing Laboratory alongwith control (T_0) was laid out in randomized block design along with 3 replications during the rabi season 2022 at the Field Experimentation Centre, Department of Genetics and Plant Breeding, Naini Agricultural Institute, Sam Higginbottom University of Agriculture, Technology and Sciences (SHUATS), Prayagraj, Uttar Pradesh. Observations were recorded for every treatment for five randomly selected plants in each replication of field emergence percentage, plant height (cm), number of leaves per plant, length of leaves (cm), area (cm^2), root length (cm), root diameter (cm), root fresh weight (gm), root yield per plot (kg), chlorophyll content (mg/g) and moisture percent. Statistical analysis of the field data was adopting the procedure described by [10].

RESULTS AND DISCUSSION

The analysis of the experimental data reveals significant insights into the effects of various treatments on different parameters in Table 1, 2a and 2b.

Field emergence (%)

The treatments showed significant effect of pre-sowing seed treatment on field emergence. Mean values of field emergence ranged between 71.17 to 88.87% with the grand mean value of 80.25%. Maximum field emergence was observed 88.87% in seed treatment T_7 rhizobium at 12% for 6hrs followed by 87.35% in T_6 rhizobium at 10% for 6hrs and minimum field emergence was observed 71.17% in T_0 control. The poor germination from untreated seed may be due to their failure to mobilize the resource from the seeds during initial period of germination but inoculated seeds made of the loss by using an improved synthesis of secondary metabolites and the presence of growth promoting substances which migrates into the seeds have brought this positive effect.

Table 1. Analysis of variance for different treatments for pre and post-harvest characters in French Bean

Traits	Sum of squares	DF	Mean square of treatment	F ratio
Field emergence	1196.37	12	99.70*	128.69
Plant height @30DAS	71.81	12	5.99*	15.95
Plant height @60DAS	118.45	12	9.87*	12.13
Number of primary branches	17.49	12	1.46*	9.61
Days to 50% Flowering	50.26	12	4.19*	7.1
Number of pods per plant	103.2	12	8.60*	11.45
Number of seeds per plant	12.87	12	1.07*	3.1
Yield of pod per plant	24274.46	12	2022.87*	2193
Seed yield per plot	1374984.12	12	114582.01*	16.27
Seed yield per h	220001.16	12	18333.43*	16.27
Seed Index	71.65	12	5.97*	6.39
Biological Yield	634.93	12	52.91*	9.58
Harvest Index	20.56	12	1.71*	18.27

*5% level of significance

Table 2a. Mean performances of different treatments for Pre harvest parameters in French Bean

Treatment Symbols	Field emergence (%)	Plant height at @30 DAS (cm)	Plant height at 60 DAS (cm)	Number of primary branches	Days to 50% Flowering	Number of pods per plant	Number of seeds per pod
T_0	71.17	19.15	38.28	2.58	47.66	7.66	3.24
T_1	73.85	19.88	41.42	2.54	46	9.66	4.00
T_2	72.33	21.15	41.21	3.25	46	10.37	4.03
T_3	75.71	21.43	41.03	3.17	45	9.6	4.11
T_4	83.45	20.88	40.71	3.5	46.33	11.46	4.06
T_5	78.6	21.87	42.38	2.94	47.66	10.95	4.12
T_6	87.35	23.5	44.37	4.57	44.33	13.26	5.21
T_7	88.84	24.77	45.37	4.75	44	14.5	5.39
T_8	80.65	21.85	43.32	4.12	45.66	9.93	5.17
T_9	85.43	21.04	40.78	3.37	47.33	11.38	4.04
T_{10}	78.59	21.46	42.81	3.94	47	10.7	4.59
T_{11}	84.27	21.97	42.31	3.12	45.66	10.37	4.3
T_{12}	83.48	21.45	41.31	3.47	46.66	11.14	4.51
Grand Mean	80.25	21.57	41.92	3.47	46.1	10.84	4.37
F Test	S	S	S	S	S	S	S
C.V.	5.65	1.46	1.91	0.75	1.31	1.79	0.75
SE	0.9	0.023	0.3	0.12	0.21	0.28	0.12

Legends : T_0 - control, T_1 - PSB 12%, T_2 – PSB 10%, T_3 - PSB 8%, T_4 - *Azospirillum* 12%, T_5 - Azospirillum 10%, T_6 - Azospirillum 8%, T_7 - Rhizobium 12%, T_8 - Rhizobium 10%, T_9 - Rhizobium 8%, T_{10} - Trichoderma 12%, T_{11} - Trichoderma 10%, T_{12} - Trichoderma 8%.

[11] reported in French bean that Rhizobium + PGPR treatment has been found to improve the field emergence.

Plant height (cm)

The mean performance of plant height at 30 DAS was ranged between 19.15cm to 24.77cm with the mean value of 21.57 cm. It was found that the maximum plant height at 30 DAS was under T_7- Rhizobium at 12% for 6hrs, *i.e.* 24.77 cm followed by T_6 rhizobium at 10% for 6hrs, *i.e.* 23.50 cm and the minimum plant height at 30 DAS was under T_0 control, *i.e.* 19.15 cm. The mean performance of plant height at 60 DAS was ranged between 38.28 cm to 45.37 cm with the mean value of 41.92 cm. It was found that the maximum plant height at 60 DAS was under T_7 rhizobium at 12% for 6hrs, *i.e.* 45.37 cm followed by T_6 rhizobium at 10% for 6hrs, *i.e.* 44.37 cm and the minimum plant height at 60 DAS was under T_0 control, *i.e.* 38.28 cm. Rhizobium produces amino acids and growth promoters (auxin, cytokinin and GA) which fix atmospheric nitrogen leading to improvement of the

Table 2b. Mean performances of different treatments for characters in French Bean (Post harvest).

Treatment symbols	Yield of pod per plant (g)	Seed yield per plot (g)	Seed yield per h (q/h)	Seed index (g)	Biological yield (g)	Harvest index (%)
T_0	113.42	367.99	147.2	33.95	120.61	1.22
T_1	122.66	490.93	196.37	34.91	115.13	1.7
T_2	126.72	511.17	204.47	36.28	114.15	1.79
T_3	128.43	527.99	211.2	36.15	111.33	1.9
T_4	146.38	594.54	237.81	34.32	113.62	2.1
T_5	147.77	623.39	249.35	34.76	119.09	2.09
T_6	174.85	910.91	364.36	35.78	117.3	3.11
T_7	201.68	1087.53	435.01	39.06	104.83	4.15
T_8	167.5	867.1	346.84	34.79	111.74	3.1
T_9	174.54	706.09	282.43	35.42	119.08	2.37
T_{10}	134.1	616.48	246.59	37.47	116.36	2.12
T_{11}	164.18	706.13	282.45	34.43	114.5	2.47
T_{12}	131.27	592.62	237.05	35.44	111.57	2.12
Grand Mean	148.73	616.75	264.7	35.6	114.56	2.32
F Test	S	S	S	S	S	S
CV	25.28	202.49	2.64	35.59	1.14	2.32
SE	4.04	32.42	80.99	1.58	4.52	0.77

Legends : T_0 - control, T_1 - PSB 12%, T_2 – PSB 10%, T_3 - PSB 8%, T_4 - *Azospirillum* 12%, T_5 - Azospirillum 10%, T_6 - Azospirillum 8%, T_7 - Rhizobium 12%, T_8 - Rhizobium 10%, T_9 - Rhizobium 8%, T_{10} - Trichoderma 12%, T_{11} - Trichoderma 10%, T_{12} - Trichoderma 8%.

vegetative growth. Similar results were reported by [12] that rhizobium fixed nitrogen in adequate amount significantly increases number of branches per plant resulted in more plant height.

Number of primary branches

The mean performance of number of primary branches was ranged between 2.54 to 4.75 with the mean value of 3.47. It was found that the maximum number of primary branches was under T_7 rhizobium at 12% for 6hrs, *i.e.* 4.75 followed by T_6- Rhizobium at 10% for 6hrs, *i.e.* 4.57 and the minimum number of primary branches was under T_1 PSB at 12% for 6hrs, *i.e.* 2.58. The significant increase in number of branches plant per plant was due to the presence of nutrients in manures and fertilizers. Rhizobium encourages proliferation of root system and better absorption of nutrients resulted in increased branches. The results of the present study confirm the findings of [13] in French bean.

Days to 50% flowering

The mean performance days to 50% flowering was ranged between 44.00 to 47.66 with the mean value of 46. It was found that the maximum days to 50% flowering was found in T_0 (control) *i.e.* 47.66 followed by T_3 PSB 8% for 6hrs (45.00) and the minimum days to 50% flowering was found in T_7 rhizobium 12% for 6hrs (44.00). [14] reported that control has more days to 50% flowering in French bean.

Number of pods per plant

The mean performance of number of pods per plant was ranged between 7.66 to 14.50 with the mean value of 10.84 It was found that the maximum number of pods per plant was under T_7 rhizobium at 12% for 6hrs, *i.e.* 14.50 followed by T_9 rhizobium at 8% for 6hrs *i.e.* 13.26, and the minimum number of pods per plant was found in T_0 (Control *i.e.* 7.66) The number of pods per plant is most important character which contributes directly to yield. This indicates that 100% RDF through produce some impact on growth but yield attributing characters were much influenced by supplemental organic source which changed the soil condition making it favorable for influencing the economic traits controlling yield. Increase in number of pods per plant, were also reported by [15].

Number of seeds per pod

The mean performance of number of seeds per pod was ranged between 3.24 to 5.39 with the mean value of 4.37. It was found that the maximum number of seeds per pod was under T_7 rhizobium at 12% for 6hrs, *i.e.* 5.39 followed by T_6 rhizobium at 10%, for 6hrs, *i.e.* 5.21 and the minimum number of seeds per pod was under T_0 (control), *i.e.* 3.24. [16] reported that 100% of rhizobium treatment showed greater number of seed per pod in soybean.

Yield of pods per plot (g)

The mean performance of yield of pods per plot was ranged between 113.42g to 201.68g with the mean value of 148.73. It was found that the maximum yield of pods per plot was found in T_7 rhizobium at 12% for 6hrs, *i.e.* 201.68g followed by T_6 *azopirillum* at 8% 6hrs, *i.e.* 174.85g and the minimum yield of pods per plot was under T_0 (control), *i.e.* 113.42g.

Seed yield per plot (gm)

The mean performance of seed yield per plot was ranged between 367.99 to 1087.53 with the mean value of 161.75 It was found that the maximum seed yield per plot was found in T_7 rhizobium at 12% for 6hrs, *i.e.* 1087.53g followed by T_6 Azospirillum at 8% for 6hrs, *i.e.* 910.91g and the minimum seed yield per plot was under T_0 (control), *i.e.* 367.99g.

Seed yield per hectare (q/h)

The mean performance of seed yield per hectare was ranged between 147.20 to 435.01 with the mean value of 264.70. It was found that the maximum seed yield per hectare was under T_7 rhizobium at 12% for 6hrs, *i.e.* 435.01 followed by T_6 *Azospirillum* at 8% for 6hrs, *i.e.* 364.36 and the minimum seed yield per hectare was found in T_0 (control), *i.e.* 147.20. Rhizobium treatment enhanced the seed yield parameters might be due to increased availability of N and P in the root zone during the vegetative growth of crop through more atmospheric nitrogen fixation by rhizobium. [17] reported that the seeds treated with bio-fertilizers producing high yield and enhance the plant growth.

Seed index (g)

The mean performance, Seed index was ranged between 33.95 to 39.06 with the mean value of 35.60g. It was found that the maximum seed index was found in T_7 rhizobium at 12% for 6hrs, *i.e.* 39.06g followed by T_{10} *Trichoderma* at 12% for 6hrs (37.47g) and the minimum seed index was under T_0 (control), *i.e.* 33.95g. [11] reported in French bean that rhizobium + PGPR treatment resulted in more seed index. Similarly, [16] reported in soybean that 100% of rhizobium treatment resulted in more seed weight.

Biological yield (g)

The mean performance, biological yield was ranged between 104.83 to 120.61 with the mean value of114.56. It was found that the maximum biological yield was found in control *i.e.* 120.61g followed by T_9 rhizobium at 8% for 6hrs, *i.e.* 119.08g and the minimum biological yield was observed in T_7 rhizobium at 12% for 6hrs, *i.e.* 104.83g. [12] reported in french bean that rhizobium treatment has been found to improve the yield. It may be due to more leaves and plant height, as according to the source sink relationship, more carbohydrates were produced due to a greater number of leaves, sank into the root zone and hence more production.

Harvest index (%)

Harvest index was ranged between 1.22 to 4.15 with the mean value of 2.32%. Maximum harvest index was found in T_7 *rhizobium* at 12% for 6hrs, *i.e.* 4.15% followed by T_8 rhizobium at 10% for 6hrs, *i.e.* 3.10% and the minimum harvest index was found in control, *i.e.* 1.22%. [16] reported in soybean that 100% of *rhizobium* treatment resulted in more harvest index. This effect of higher harvesting index might be due to the presence of good vegetative growth by utilizing both organic and inorganic source of nutrients available for the plants at different growth periods. The highest harvest index might be due to the larger seed size with higher sink capacity [18].

CONCLUSION

It was concluded from the present study that the seeds of French bean variety Ellora Number 11 were treated with *rhizobium* @12% with 6 hours (T_7) showed significant increase in terms of growth, yield and yield attributing traits followed by *Azosprillum* @ 8% with 6 hours (T_6) as compared to control (untreated) seeds. Therefore, pre sowing seed treatment with *rhizobium* was found more productive and eco-friendly. The farmers can reap the benefits of pre sowing seed treatments as it minimizes the time for plant emergence, expense of re-seeding, additional irrigation and fertilization, weed management, ineffectiveness of cultural practices used on a stand of non-uniform growth. Seeds treated with bio-fertilizers demonstrated remarkable effect on germination, growth and yield parameters of Frenchbean. All the bio-fertilizer treated seeds are likely to play an important role in production of food which is free from toxic and chemical residues were beneficial for humans, cattle and environment. Since all the treatments showed better results and therefore the bio-fertilizer treatments stimulates the growth and yield in crops.

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