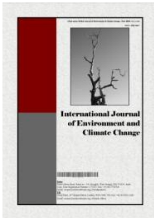

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Pre-Sowing Seed Treatment with Bio Inoculants and Micronutrients on Growth, Yield and Yield Attributing Traits of Lentil

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The field experiment entitled "Pre sowing seed treatment of bio inoculants and micronutrients on growth, yield and yield attributing traits of lentil (*Lens culinaris L.*)" var. Shekhar Masoor-2 was conducted during rabi at central reseach field of the Department of Genetics and Plant Breeding, Sam Higginbottom University of Agriculture, Technology & Sciences, Prayagraj, Uttar Pradesh, India during 2020 - 2021. The experiment consisted of 13 treatments which was laid in Randomized Block Design (RBD). Results revealed that seeds treated with T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) recorded maximum values in growth parameters viz., field emergence (%) 88.72%, plant height at 30, 60, 90 DAS with 6.97, 23.33, 45.2 cm Days to 50% flowering (72.67days), number of branches 5.07 branches per plant, number of pods per plant with 64.07 pods per plant, number of seeds per pod 1.85 seeds per pod. Similar results are observed in yield paramters where highest seed yield per plant was observed in T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) with 3.79 g and seed yield per plot 107.4 g.

Keywords: Ammonium molybdate; Phosphate solubilising bacteria; Pottasium mobilizing bacteria; Rhizobium; Yield parameters; $ZnSO_4$; Seed priming.

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1. INTRODUCTION

Lentil (*Lens culinaris L.*) is an important *Rabi* pulse crop extensively grown in India. It is an important annual leguminous crop which is locally called "Masoor" belongs to the family Fabaceae. As a leguminous crop, it utilizes atmospheric nitrogen to meet its partial nitrogen requirement and thus occupies an important place in crop rotation in different part of the country. It can adapt well to a wide range of climatic and soil condition. Lentil contains about 24-26% protein and 57-60% carbohydrates besides fat, calcium, phosphorus and iron. Lentil is also a good source of vitamin A and vitamin C. Lentil has got special importance in diet and is consumed in various ways [1].

India occupies second position of lentil production in the world after Canada and is the fifth most important pulse crop in India in terms of production after chickpea, pigeon pea, mungbean and urdbean (Singh et al., 2015). The major lentil producing states are Uttar Pradesh, Madhya Pradesh, Bihar, West Bengal, Rajasthan and Assam. In India the area under lentil was 1.42 million ha with a produce of 1.13 million tons while the productivity was 797 kg/ha. In Uttar Pradesh it is grown on 4.95 lakh ha area with total production of 4.41 lakh tons and average productivity of 891 kg/ha during 2018-19 [2].

Seed priming is a pre-sowing treatment which leads to a physiological state that allows seed to germinate more proficiently. The preponderance of seed a treatment are based on seed imbibition allowing the seeds to go through the first reversible stage of germination but does not allow radical protrusion through the seed coat. Seeds keeping their desiccation tolerances are then dehydrated and can be stored until final sowing [3] Heydecker et al., [4] Heydecker et al., [5].

Any improvement in agricultural system that results in higher production should reduce the negative environmental impact and enhance the sustainability of the system. One such approach is the use of bio inoculants, which can enhance the effectiveness of conventional mineral fertilizers. Long-term indiscriminate use of them invites the crucial problem of soil health disorder vis-à-vis reduced input use efficiency, more precisely, fertilizer use efficiency. Due to intensive cropping systems, soils are becoming deficient in macro as well as micronutrients. Hence the logical alternative is to increase the usage of bio inoculants and micronutrients.

Bio-priming is an innovative skill of seed treatment that assimilates biological (inoculation of seed with beneficial organism to protect seed) and physiological facets (seed hydration) of disease control [6]. Bio-priming is directly involved in the enrichment of plant development by the excretion of compounds and mineral solubilisation [7], Balaji et al., [8]. *Rhizobium* inoculation is well known agronomic practice to ensure adequate nitrogen of legumes instead of N-fertilizers [9]. There is a good possibility to increase crop production through inoculation of effective nitrogen fixing bacteria to the seed. Pulse crop production in most of agricultural systems is dependent on symbiotic nitrogen fixation, efficiency of which depends on rhizobium strain and host cultivar interaction which is influenced by several environmental and soil edaphic factors. Soil may be lacking in effective rhizobial strains or their population may be too low to form sufficient nodules. Hence, it is necessary to inoculate the seed with the most efficient strain of *rhizobium* species to get more nitrogen fixation there by improve pulse crop production [10].

Phosphate solubilizing bacteria partly solubilizes inorganic and insoluble phosphate and improves applied phosphorus use efficiency stimulating plant growth by providing hormone, vitamin and other growth promoting substances [11]. Zinc plays a vital role in the synthesis of protein and nucleic acid and help in the utilization of nitrogen and phosphorus in plant. It also promotes nodulation and nitrogen fixation of leguminous crops. Molybdenum plays a vital role in nitrogen fixation through its effect on nitrogenase enzyme of nodule in leguminous plants Togay et al., [12]. The chief role of molybdenum is activate nitrate reductive enzyme during nitrogen metabolism [13] Kumar et al., [14] Tahir et al., [15]. From the above study, the experiment was undertaken to study the suitable pre sowing seed treatments of lentil var. KLB-303 (Shekhar Masoor-2).

2. MATERIALS AND METHODS

The field experiment was carried out in a Randomized Block Design (RBD) with three replications during *Rabi*, 2020-2021 at the central research field, Department of Genetics and Plant Breeding, Naini Agricultural Institute, Sam Higginbottom University of Agriculture Technology and Sciences, Prayagraj (U.P.) India, which is located at 25^{o}39'42''N latitude, 81^o 67'56''E longitude and 98 m altitude above the mean sea level. The soil was sandy loam in texture, low in organic carbon and medium in available nitrogen, phosphorous and low in potassium. The recommended dose of fertilizer was applied 20:40:20 kg N, P_2O_5 and K_2O. Nutrient sources were Urea, DAP, MOP to fulfil the requirement of Nitrogen, phosphorous and potassium. Gypsum used to fulfil the requirement of sulphur.The genetically pure seeds of lentil var. KLB-303 (Shekhar Masoor-2) are bold seeded and moderately tolerant to wilt and rust, seeds are brown and small and were subjected to various pre sowing seed treatments like.

1. T_0 (control)
2. T_1 *Rhizobium* @ 5g
3. T_2 Phosphate solubilizing bacteria (PSB) @ 5g
4. T_3 Potash mobilizing bacteria (KMB) @ 5g
5. T_4 ($ZnSO_4$) @ 0.1% for 8hrs
6. T_5 Ammonium molybdate @ 0.1% for 8 hrs
7. T_6 (*Rhizobium* + PSB)
8. T_7 (*Rhizobium* + PSB + KMB)
9. T_8 ($ZnSO_4$ + Ammonium molybdate)
10. T_9 (*Rhizobium* + $ZnSO_4$ + Ammonium molybdate)
11. T_{10} (PSB + $ZnSO_4$ + Ammonium molybdate)
12. T_{11} (KMB + $ZnSO_4$ + Ammonium molybdate)
13. T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate)

Seeds were presoaked in distilled water before treating with bio inoculants at room temperature for 8hrs, Dry formulation of *Rhizobium*, PSB, KMB at the rate 5 gm Kg^{-1} of seed each is added to pre-soaked lentil seeds. The treated seeds are taken in polythene bags, heaped and covered with moist jute sack to maintain high humidity and maintained for 2 hr at approximately 25–32°C. For the preparation of 0.1% (w/v) solution of the $ZnSO_4$, ammonium molybdate, 1gm each of chemicals $ZnSO_4$ and ammonium molybdate were taken in a beaker seperately and 1000 ml. of distilled water is added with constant stirring. After preparation of solution $ZnSO_4$ and ammonium molybdate the lentil seeds are soaked in each solution for 8 hrs at room temperature (25±1°C). Untreated seed is called as control. After priming for 8 hrs the solution were drained out from the beaker and were air dried to intial moisture. After drying, seeds are sown in experimental research plot.

The Experiment was laid out in Randomized Block Design, with 13 treatments which are replicated thrice. Date of sowing was on 27th November 2020. In the period from germination to harvest several growth parameters were recorded at frequent intervals along with it after harvest several yield parameters were recorded those include viz. field emergence, plant height (cm) at 30, 60, 90 DAS, days to 50% flowering, numbers of branches per plant, numbers of pods per plant, number of seeds per pod, test weight (g), seed yield per plant (g), seed yield per plot (g), biological yield (g) and harvest index (%). The mean data of each character replicated three times was worked out statistically by the method of Analysis of Variance (Fisher 1936) using RBD (Randomize Block Design by Panse and Sukhatme, [16].

3. RESULTS AND DISCUSSION

Results revealed significant differences in seeds subject to pre sowing treatments. The study revealed that among different treatments T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) showed maximum growth and yield attributing traits. The mean values, the coefficient of variation (CV), standard error of difference (SE), the critical difference (CD) at 5% and range of 13 seed priming treatments for 13 quantitative characters are presented in Table 1 which revealed a wide range of variation for all the traits studied. The mean performance of different priming treatments with respect to different characters is described as under.

Growth Attributes: The mean performance of field emergence (%) ranged from 76.19 to 88.72 with a grand mean of 83.07%. Perusal of Table 1 indicated that maximum field emergence (%) recorded under T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (88.72%) and least in T_0 (control) (76.19%). Among the significant treatments, Treatment T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (88.72%) was significantly higher in comparison to control as well as other significant treatments.The results are in agreement with findings of Biswas et al., [17].

The mean performance of days to 50% flowering ranged from 62.33 to 74.0 with a grand mean of 69.41 days. Among these treatments, T_8 ($ZnSO_4$ + Ammonium molybdate) (67.67days), T_3 Potash mobilizing bacteria (KMB) (5gm) (65.67 days) was significantly lower as compared to control and other treatments.

The mean performance of plant height (cm) at 90 DAS ranged from 35.47 to 45.2 with a grand mean of 40.67 cm. Maximum plant height (cm) was recorded in T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (45.2 cm). All treatments were found significantly higher in comparison to control. Among these treatments T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (45.2 cm) was found significantly higher in comparison to control and other treatments. The similar results were observed by Singh et al., [18], Amit kumar Tiwari et al., [19], Rabieyan et al. (2011).

The mean performance of number of branches per plant ranged from 3.6 to 5.07 with a grand mean of 4.36. Maximum number of primary branches per plant recorded under T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (5.07). Among the significant treatments, T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (5.07) followed by T_{10} (PSB + $ZnSO_4$ + Ammonium molybdate) (4.67), T_9 (*Rhizobium* + $ZnSO_4$ + Ammonium molybdate) (4.53), T_{11} (KMB + $ZnSO_4$ + Ammonium molybdate) (4.53), T_8 ($ZnSO_4$ + Ammonium molybdate) (4.53) were found significantly higher in comparison to control. Similar results were observed by Ali et al., [20], Hussain et al. [21].

Yield Attributes: The mean performance of number of pods per plant ranged from 39.47 to 64.07 with a grand mean of 51.83. For number of pods per plant, all the treatments were significantly higher in comparison to control except T_1. Maximum number of pods per plant recorded under T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (64.07). Among the treatments, T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (64.07) followed by T_{11} (KMB + $ZnSO_4$ + Ammonium molybdate) (60.6) were found significantly higher in comparison to control and other treatments. Similar findings are reported by Rabieyan et al. (2011), Bandoupadhayay SK [22].

The mean performance of number of seeds per pod ranged from 1.54 to 1.85 with a grand mean of 1.67. For number of seeds per pod, all the treatments were significantly higher in comparison to control. Maximum number of seeds per pod recorded under T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (1.85). Among the treatments, for number of seeds per pod T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (1.85), T_{11} (KMB + $ZnSO_4$ + Ammonium molybdate) (1.82), T_{10} (PSB + $ZnSO_4$ + Ammonium molybdate) (1.78) were found significantly higher in comparison to control and other treatments.

The mean performance of test weight (g) ranged from 25.7 to 36.05 with a grand mean of 30.61. For test weight (g), all the treatments were significantly higher in comparison to control. Maximum test weight (g) recorded under T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (36.05g). Among the treatments, for test weight (g), T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (36.05g) was found significantly higher in comparison to control and other treatments. Similar results are also reported by Zaman et al. [23] and Yasari et al. [24].

The mean performance of biological yield (g) ranged from 113.27 to 183.13 with a grand mean of 153.67. For biological yield (g), all the treatments were significantly higher in comparison to control except T_2 and T_3. Among the treatments, biological yield (g), T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (183.13g), T_6 (*Rhizobium* + PSB) (179.2g), T_{11} (KMB + $ZnSO_4$ + Ammonium molybdate) (174.47g), T_9 (*Rhizobium* + $ZnSO_4$ + Ammonium molybdate) (169.13g), T_{10} (PSB + $ZnSO_4$ + Ammonium molybdate) (166.73g), T_8 ($ZnSO_4$ + Ammonium molybdate) (164.47g), T_7 (*Rhizobium* + PSB + KMB) (160.73g) were found significantly higher in comparison to control and other treatments.Similar findings are reported by Meena et al., [25],Wondimu et al., [26].

The mean performance of seed yield per plant (gm) ranged from 2.94 to 3.79 with a grand mean of 3.3g. For seed yield per plant (g), all the treatments were significantly higher in comparison to control except T_1, T_2 and T_4. Among the treatments, for seed yield per plant (g), T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (3.79g), T_{11} (KMB + $ZnSO_4$ + Ammonium molybdate) (3.59g) was found significantly higher in comparison to control and other treatments.Similar findings are reported by Mishra and Tiwari [27] and Hossain et al., (2010).

The mean performance of seed yield per plot (gm) ranged from 71.55 to 107.4 with a grand mean of 86.87g. For seed yield per plot (gm), all the treatments were significantly higher in comparison to control. Among the treatments, for seed yield per plot (gm), all the treatments were found significantly higher than control. Among the significant treatments, T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) (107.4g) was found significantly higher in comparison to control and other treatments. Similar findings are reported by Kumar et al., [28], Hafeez et al. (2000) and Shah et al. (2000).

Table 1. Mean performance of lentil for Growth, Yield and Yield attributes

S. No.	Symbols	Field emergence (%)	Plant height at 30 DAS (cm)	Plant height at 60 DAS (cm)	Plant height at 90 DAS (cm)	Days to 50% flowering	Number of branches plant^{-1}	Number of pods plant^{-1}	Number of seeds pod^{-1}	Seed yield plant^{-1} (gm)	Seed yield plot^{-1}	Test Wt. (gm)	Biological yield (gm)	Harvest index (%)
T0	Control	76.19	5.66	20.73	35.47	73.67	3.6	39.47	1.54	2.94	71.55	25.7	113.27	64.27
T1	*Rhizobium* @ 5g	77.64	6.2	20.8	40.07	64.67	4.33	43.13	1.55	2.97	72.29	29	136.47	53.82
T2	Phosphate solubilizing bacteria (PSB) @ 5g	82.34	5.73	20.27	40.6	64.67	4.27	46.27	1.63	3.11	79.92	29.5	119.6	66.96
T3	Potash mobilizing bacteria (KMB) @ 5g	80.15	6.13	21.53	38.73	65.67	4.4	45.33	1.57	3.32	86.33	28.4	124.13	69.62
T4	$ZnSO_4$ @ 0.1%	81.27	6.2	22.1	40.67	73.33	4.2	49.33	1.58	3.17	78.4	29.9	149.93	52.45
T5	Ammonium molybdate) @ 0.1%	82.05	5.8	22.03	40.6	70	4.4	48.2	1.65	3.27	85.98	30.6	156.47	55.3
T6	(*Rhizobium* + PSB)	82.33	6.4	21.63	41.07	73.67	4.13	50.73	1.61	3.28	87.48	31.62	179.2	48.9
T7	(*Rhizobium* + PSB + KMB)	83.21	5.94	20.83	40.27	**74**	4.07	57.53	1.68	3.32	87.4	30.3	160.73	54.72
T8	($ZnSO_4$ + Ammonium molybdate)	83.67	5.87	21.27	41.2	67.67	4.53	54.8	1.69	3.3	90.17	30.55	164.47	54.83
T9	(*Rhizobium* + $ZnSO_4$ + Ammonium molybdate)	86.49	6	22.47	40.73	68.67	4.53	56.4	1.77	3.38	91.31	31.8	169.13	54.14
T10	PSB + $ZnSO_4$ + Ammonium molybdate	87.47	6.65	22.17	40.8	62.33	4.67	57.87	1.78	3.4	91.91	32.5	166.73	55.19
T11	(KMB + $ZnSO_4$ + Ammonium molybdate)	88.37	6.23	22.43	43.27	71.33	4.53	60.6	1.82	3.59	99.16	32.04	174.47	57.46
T12	(*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate)	**88.72**	**6.97**	**23.33**	**45.2**	72.67	**5.07**	**64.07**	**1.85**	**3.79**	**107.4**	**36.05**	**183.13**	**58.7**
	Mean	83.07	6.14	21.66	40.67	69.41	4.36	51.83	1.67	3.3	86.87	30.61	153.67	57.41
	C.V.	1.9	6.15	4.02	3.12	4.16	8.93	4.59	3.38	4.26	6.59	5.2	8.8	10.52
	F ratio	18.23	2.94	2.99	9.19	5.99	2.36	28.58	10.44	7.99	9.15	7.01	8.91	2.97
	F Prob.	0	0.01	0.01	0	0	0.04	0	0	0	0	0	0	0.01
	S.E.	0.91	0.22	0.5	0.73	1.67	0.23	1.37	0.03	0.08	3.3	0.92	7.81	3.49
	C.D. 5%	2.66	0.64	1.47	2.14	4.87	0.66	4.01	0.1	0.24	9.64	2.68	22.78	10.18
	C.D. 1%	3.61	0.86	1.99	2.9	6.6	0.89	5.43	0.13	0.32	13.07	3.64	30.88	13.79

The mean performance of harvest index (%) ranged from 48.9 to 69.62 with a grand mean of 57.41%. Maximum harvest index (%) recorded under T_3 Potash mobilizing bacteria (KMB) (69.62%). Among the treatments, no treatment was found significantly higher in comparison to control for Harvest index (%). Maximum Harvest index (%) was recorded in T_3 Potash mobilizing bacteria (KMB) @ 5g Kg^{-1}seed (69.62%), followed by T_2 Phosphate solubilizing bacteria (PSB) @ 5g Kg^{-1}seed (66.96%), T0 (Control) (64.27%).

The increase in growth and yield parameters with the treatment of T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) can be explained by the fact that biopriming with *Rhizobium*, PSB, KMB significantly influenced the growth characteristics. Biopriming is the treatment of seeds with beneficial microorganisms such as *Rhizobium* is used to improve plant growth. *Rhizobium* a genus of Gram (-ve) soil bacteria that help fix atmospheric nitrogen with the roots of plants legumes. The bacteria colonize the plant cells within root nodules where they convert atmospheric nitrogen in to ammonia using the enzyme nitrogenase and then supplying the plant with nitrogenous organic compounds such as glutamine. The plant, in turn, supplies bacteria with organic compounds produced by photosynthesis. This mutually beneficial relationship is true for all rhizobia, of which the genus rhizobium is a typical example. This mutual association seems to be the reason for the increase in growth characteristics. Similar results are also reported by Zaman et al. [23], Gangwar, S. and Dubey, M. [29]. and Yasari et al. [24]. Increased yield by the PSB could be due to the greater availability of nutrients in the soil and better nodulation under the influence of inoculation resulting in better growth and development which might be attributed to better mobilization of phosphorus and increased allocation parts and also hormonal balance on the plant system. Similar findings are observed in Menaria (2004) in soybean, Rajput et al. [30] in garden pea and Jain et al. [31] in chick pea, Poonia, T.C. and Pithia, M.S. [32], Prajapati K. [33], Priyamvada C [34].

4. CONCLUSION

It may be concluded from the present investigation that different priming treatments were affecting growth significantly. Priming with T_{12} (*Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) was found better growth and yield parameters in comparison to control and other treatments. Thus, application of T_{12} (combination of *Rhizobium* + PSB + KMB + $ZnSO_4$ + Ammonium molybdate) is useful for improving yield in lentil. Therefore, this study reveals combination of bio inoculants and micronutrients were capable to increase growth and yield parameters. Further investigations under field conditions might be needed to clarify the role of bio inoculants and micronutrients in lentil and other crops for commercial cultivation by farmers.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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