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Research Article

Evaluation of Potential Microbial Strains and Lentil Genotypes for Enhancing BNF and Production of Lentil Crop in Pothwar

Ehsan ul Haq¹, Hajra Parveen¹, Khan Bahadar², Ameer Uddin³, Muhammad Yousuf⁴, Ameer Hamza⁵, Naeem Iqbal⁶, Jahangir Ahmed⁷, Abdul Mannan Hamzah⁸, Ghulam Mujtaba⁹

¹Department of Agronomy, PMAS Arid Agriculture University Rawalpindi, Pakistan.

²Food and Agriculture Section, Ministry of Planning, Development and Special Initiatives, Islamabad, Pakistan.

³Agriculture Research, Awaran, Baluchistan, Pakistan.

⁴Directorate of Agriculture Research, Panjgur, Balochistan, Pakistan.

⁵Department of Plant Breeding and Genetics, PMAS Arid Agriculture University Rawalpindi, Pakistan.

⁶Department of Soil and Environmental Sciences, MNS University of Agriculture Multan, Pakistan.

⁷Directorate of Agriculture Research Oil Seeds ARI, Sariab Quetta, Pakistan

⁸Department of Entomology, PMAS Arid Agriculture University Rawalpindi, Pakistan.

⁹Institute of Soil and Environment Sciences, PMAS Arid Agriculture University Rawalpindi, Pakistan.

ABSTRACT

Lentil (*Lens culinaris* L) is Pakistan's second main winter season legume crop after chickpea and is known to be vital for health. Rhizobia have a natural capacity to fix atmospheric nitrogen in legume crops through a symbiotic relationship with legumes-rhizobium, producing nodules on their roots. It is important to maintain an optimal rhizobial population in the rhizosphere in order to enhance nodulation, N₂ fixation, and yield of lentil crop. The experiment was conducted to Evaluation of potential Microbial strains and lentil genotypes for enhancing BNF and production of lentil crop in Pothwar. Lentil varieties used were: V₁ = NIA-Masoor-2005, V₂ = Markaz-2009, V₃ = Punjab-Masoor-2009, V₄ = Chakwal-Masoor-2011, V₅ = NIA-Masoor-2016 and the five *rhizobial* strains, RS₀, RS₁, RS₂, RS₃, RS₄ and RS₅ (Their accession numbers lie with the Institute of Soil science, PMAS AAUR) were used. Inoculation of seed with *Rhizobium* strains significantly increased the plant height (17.58%), number of branches plant⁻¹ (25.67%), number of pods plant⁻¹ (15.42%), number of nodules plant⁻¹ (34%) and the N-content of shoots (12.70) of lentil crop as compared to control. Consequently, *rhizobium* strains contributed to improved soil productivity and could minimize the cost of production of next crop by reducing inputs in the form of nitrogen fertilizers. It has been found that *rhizobic* inoculation is more productive and better yields, as compared to control. Maximum growth and yield components was recorded in variety, V₅ (NIA-Masoor-2016) followed by variety, V₃ (Punjab-Masoor-2009) and minimum production was recorded in V₄ (Chakwal-Masoor-2011). NIA-Masoor-2016 had larger yielded better than Chakwal-Masoor-2011. It was also clear from the present results that interactive effect of *rhizobial* strains and lentil varieties produced the plants with more yield than control which might have resulted in higher nitrogen fixation and consequently higher dry matter production.

Keywords: Lentils, Legume, BNF, Rhizobia, Yield.

INTRODUCTION

In Pakistan, pulses are mostly grown on marginal lands with low rhizobial population. Majority pulses are grown in Punjab (Thal and Potohar) and Khyber



Correspondence

Ehsan ul Haq
ehsanulhaq620@gmail.com

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Pukhtunkhwa (Malakan) on 14.2 thousand hectares (Nourin et al., 2019). Punjab contributes 49.3 % of the country's total production. It was cultivated on an area of 17,700 and 12,400 hectares in 2014-15 and 2018-19 with total production of 9,000 and 6,400 tons respectively (Finance and India, 2018). Lentil (*Lens culinaris* L) is Pakistan's second main winter season legume crop after chickpea and is known to be vital for health. Because of their nitrogen-fixing capacity, the lentil has great nutritious value in cereal cropping systems. Its seeds have high micronutrients for human diet and its straw used for animal feed (Singh and Singh, 2018). It is cheap source of protein, calories, and some vitamins (Nourin et al., 2019). On an average, lentils contain around 25 % protein, while 340-346 calories account for 100 g of dried lentils.

In Pothwar, lentil is grown by resource-poor farmers in marginal soils with low fertility resulting in low yields than the potential of the cultivars. Pothwar soils are moisture and nutrient deficient due to low and erratic rainfalls and little use of nutrients especially nitrogenous and phosphatic fertilizers, which decline the yields of lentil crop in the rain-fed areas of Pothwar I (Iqbal et al., 2017). Application of chemical fertilizers at the time of sowing under the erratic and low rainfall does not guarantee the sustained nutrition availability to the plants due to losses of nitrogen (Abraham, 2015; Rani et al., 2014). Indiscriminate use of fertilizers and pesticides have been in cultivated areas resulted in depletion of soil quality in contort with fertility, biodiversity and productivity of microbial population (Singh et al., 2018). Lentil area has been significantly reduced in previous years due to poor economic lentil return relative to other winter crops (Nourin et al., 2019). The area as well as its production has been decreased gradually almost to 40% & 28% from 2015-18 mainly due to lack of adopting high yielding cultivars, low soil fertility and poor native microbial populations and shift of main lentil area to other crops in Pakistan (Ahmed et al., 2008).

The cost of chemical fertilizers is becoming unaffordable for rainfed growers. There is great dire to augment or replace organic manures with chemical fertilizers or to seek biological means to increase soil production. Native manures are not available in the Pothwar region of Pakistan, and people use them as firewood instead of using them for soil fertility. Therefore, under agro-climatic conditions in the Pothwar area, the potential of rhizobia as legume inoculants needs to be explored. Lentil, which is a legume crop, may fix atmospheric nitrogen (78%) through root nodules by *Rhizobium* bacteria, which can reduce the burden of nitrogen fertilizer application to the crop (Haque et al., 2014). Over 85 percent of the lentil 's required nitrogen needs can be met by inoculating the crop seed with successful rhizobial strains in the field, increased yields to 2 tons ha⁻¹ (Bisen et al., 1980). A number of researchers performed experiments on inoculation of rhizobium in mungbean crops with and without fertilizers and observed increased nitrogen content of plants, number of nodules, production and production components.

Lentil can render high profits to the resource poor farmers of Pothwar region if grown through some suitable agro-techniques as well as the use of proper inoculants, its yield could be doubled thus increasing farmer's income significantly (Wang et al., 2012). The foremost problems of lentil production in rain-fed areas of Pothwar are moisture stress, poor soil fertility, non-availability of tested inoculants and lack of recommended varieties compatible with the available *rhizobial strains* and climate resilient for Pothwar region. The yield of legumes in Pakistan is 0.5 to 0.6 t ha⁻¹, which is smaller than most other countries in the world. In general, the yield of chickpea and peas increased by 20-40 percent, while for other food legumes, i.e. lentil, mung and mash, it increased by 10-20 per cent (Aslam et al., 2000).

Seed inoculation with suitable *Rhizobium* sp. is recommended to boot up lentil growth. The application of rhizobium inoculation improves the technology of leguminous growth, in particular by increasing nutrient uptake in lentil crop (Singh et al., 2016). This reduces the use of nitrogenous fertilizers and protect the sustainability of our ecosystem. Judiciously mixing *Rhizobium* stains with host plants and careful use of large viable inoculum is the only way to achieve optimum nitrogen fixation and yield of lentil crop. Moreover, *Rhizobium* inoculation increased plant height, grain yield and crop residues yield of plant significantly compared to un-inoculated control (Yaseen et al., 2016).

It is hypothesized that microbial strains have potential to improve the growth and yield attributes of lentil crop. This study aims to evaluate the lentil varieties and *Rhizobium* strains compatibility for N fixation, thereby enhancing lentil growth, yield and quality attributes.

MATERIALS AND METHODS

Germplasm collection

In the first year, the interactive studies of lentil varieties collected from different research institutes/stations viz. Pulses Research Institute, NARC, Islamabad, Pulses Section, BARI, Chakwal, Pulses Research Station, AARI, Faisalabad, Barani Agriculture Research Station, Jamra (Kohat) and Agri. Research Station, Tarnab, (Peshawar) and

rhizobium strains procured from Institute of Soil Science, PMAS Arid Agriculture University, Rawalpindi was conducted.

Experiment Undertaken:

A pot experiment was carried out during the 1st year (2019-20) at PMAS Arid Agriculture University, Rawalpindi to evaluate the interactive effect of *rhizobial* strains and lentil varieties. Lentil varieties used were:

V₁ = NIA-Masoor-2005

V₂ = Markaz-2009

V₃ = Punjab-Masoor-2009

V₄ = Chakwal-Masoor-2011

V₅ = NIA-Masoor-2016

and the five *rhizobial* strains, RS₀, RS₁, RS₂, RS₃, RS₄ and RS₅ (Their accession numbers lie with the Institute of Soil science, PMAS AAUR) were used. The earthen pots (with 30 cm high and 15 cm width dimension and each pot was filled with 5 kg of dry soil with 50:50 sand mixtures of clay.) was sandy loam containing 0.045 % total nitrogen, 3 ppm nitrate, 60 ppm potassium and 4.5 ppm phosphorus. The experiment was designed in a completely randomized design (CRD) in two factor factorial arrangement with three replicates of each treatment. Five lentil varieties, NIA-Masoor-2005, Markaz-2009, Punjab-Masoor-2009, Chakwal-Masoor-2011 and NIA-Masoor-2016 were planted in 30-10-2019.

For seed inoculation, inoculum paste containing *rhizobium* (five gram) was used to coat the seeds. In every pot ten seeds were sown. All agronomic inputs like irrigation, weeding, fertilizer, and management practices were uniformly applied to each pot.

Plant height (cm):

At maturity, plant height (PH) of three plants per pot was taken and their average was taken. The height of each plant was measured at the harvest stage from the ground level to the plant apex using measuring tape in each pot, and its average was taken.

Number of branches plant⁻¹:

Three plants were randomly selected from each plot and number of branches plant⁻¹ were recorded and their average was taken as the number of branches plant⁻¹.

Number of pods plant⁻¹:

The number of pods plant⁻¹ was counted at maturity from the three selected plants pot⁻¹. The average value of these plants was taken.

Fresh shoot weight (g):

From each pot, three plants were taken at 50 percent flowering. The shoot of each plant was separated, and then fresh shoot weight (g) were taken & average was calculated.

Dry shoot weight (g):

From each pot, three plants were taken at 50 percent flowering. The shoot of each plant was separated and dried in an oven at 70°C for 72 hours. Then dried shoot weight (g) taken & average was calculated.

Fresh roots weight (g):

From each pot, three plants were taken at 50 percent flowering. The roots of each plant were separated then fresh root weight (g) was taken & average was calculated.

Dry roots weight (g):

From each pot, three plants were taken at 50 percent flowering. The roots of each plant were separated and dried in an oven at 70°C for 72 hours. Then dried root weight (g) taken & average was calculated.

Number of nodules plant⁻¹:

Plants from each pot were uprooted for root nodulation at 50 percent flowering and their nodules were counted and then averaged.

Nitrogen contents of shoot:

At flowering stage, three plants were picked from each pot for the nitrogen content of the shoot. Oven dried samples have been ground and taut. The overall nitrogen content of the shoots was measured using the total nitrogen (Jackson, 1962) formula.

Statistical Analysis

The data recorded were subjected to statistical analysis using Statistix 8.1 software. In order to compare the differences between treatment means, the least significant difference test at 5 % probability level was used (James et al., 1997).

RESULTS

Plant Height (cm)

Plant height (PH) is the vertical spatial distribution of the plant and is a key index of plant growth and development. Data (Table 1) regarding plant height as affected by different *rhizobial* strains and lentil varieties showed significant effect ($P < 0.05$). The results of the present study indicated that the tallest plants (29.96 cm) were recorded with RS₅ which was statistically at par with RS₃ (29.87 cm) followed by RS₂ (28.55 cm). However, the lowest plant height (25.54 cm) was recorded with RS₀ (control). Furthermore, it was also clear from the present results that are RS₅ significantly produced the plants with more plants height than all the other treatments.

Similarly, among the lentil varieties the highest plant height was recorded in NIA-Masoor-2016 (31.41 cm) followed by Markaz- 2009 (28.58 cm) while the lowest plant height was recorded in Punjab Masoor-2009 (25.13 cm).

The interactive effect of *rhizobial* strains and lentil varieties were also statistically significant ($P < 0.005$). Data presented in the table (1) indicated that RS₃ with NIA-Masoor-2016 produced the plants with maximum plant height (33.76 cm) that was statistically at par with RS₅ with the same lentil varieties measuring plant height (33.40cm). Furthermore, the minimum plant height was recorded in RS₀ with NIA-Masoor 2005 (24.66 cm). The increased plant height with *rhizobial* strains might be due to the improving a growth and nutrient uptake. (Singh et al., 2018) and (Huang and Erickson, 2007) also reported the similar finding that application of *rhizobium* inoculation significantly increased the plant height associated with in enhanced soil fertility and nutrient uptake. Moreover, the difference in plant height among the lentil varieties might be due to the genotypic variability. These results were in line (Gull et al., 2004) where they reported the increase plant height is highly associated with varietal traits.

Number of Branches Plant⁻¹

The number of branches plant⁻¹ is an important yield contributor to the lentil parameter. Data (Table 2.) regarding number of branches plant⁻¹ as affected by different *rhizobial* strains and lentil varieties showed significant effect ($P < 0.05$). The results of the present study indicated that the maximum number of branches plant⁻¹ (3.77) were recorded with RS₅ which was statistically at par with RS₃ (3.58). However, the lowest number of branches plant⁻¹ (3.13) was recorded with RS₀ (control). Furthermore, it was also clear from the present results that are RS₅ significantly produced the plants with more number of branches plant⁻¹ than all the other treatments.

Similarly, among the lentil varieties the highest number of branches plant⁻¹ was recorded in NIA-Masoor-2016 (4.38) while the lowest number of branches plant⁻¹ was recorded in Punjab Masoor-2009 (3.06).

The interactive effect of *rhizobial* strains and lentil varieties were also statistically significant ($P < 0.005$). Data presented in the table (2) indicated that RS₅ with NIA-Masoor-2016 produced the plants with maximum number of branches plant⁻¹ (5.00) that was statistically at par with RS₂ and RS₃ with the same lentil varieties measuring number of branches plant⁻¹ (4.50 and 4.50). Furthermore, the minimum number of branches plant⁻¹ was recorded in RS₂ with Chakwal-Masoor-2011 (2.40). The increased number of branches plant⁻¹ with *rhizobial* strains might be due to the improving a growth and nutrient uptake. (Singh et al., 2018) also reported the similar finding that application of *rhizobium* inoculation significantly increased the number of branches plant⁻¹ associated with in enhanced plant growth and nutrient uptake. Number of branches per plant of lentil varieties increased due to varietal trait and genotypic variability. These results were in line with those of (Rana and Solanki, 2015) have reported similar variations in the number of branching potential of different varieties of the lentils.

Number of pods plant⁻¹

Number of pods plant⁻¹ is the most important yield attribute in lentil crop. Significant increase in number of pod plant⁻¹ was recorded due to *rhizobial* inoculation. Data (Table 3) regarding number of pod plant⁻¹ as affected by different *rhizobial* strains and lentil varieties showed significant effect ($P < 0.05$). The results of the present study indicated that maximum number of pod plant⁻¹ (43.58) were recorded with RS₅ which was statistically at par with RS₃ (42.86) followed by RS₁ (39.02). However, the lowest number of pod plant⁻¹ (37.20) was recorded with RS₀ (control). Furthermore, it was also clear from the present results that are RS₅ significantly produced the plants with a greater number of pod plant⁻¹ than all the other treatments.

Similarly, among the lentil varieties the more number of pod plant⁻¹ was recorded in NIA-Masoor-2016 (44.79) followed by Markaz-2009 (43.57) while the lowest number of pod plant⁻¹ was recorded in Punjab-Masoor-2009 (36.38). These results are in line with those reported by (Huang and Erickson, 2007).

The interactive effect of *rhizobial* strains and lentil varieties were also statistically significant ($P < 0.005$).

Table 1. Interactive effect of *rhizobium* strains and lentil varieties on plant height (cm)

Rhizobial strains	Varieties					Mean
	NIA-Masoor-2005	Markaz-2009	Punjab-Masoor-2009	Chakwal-Masoor-2011	NIA-Masoor-2016	
RS ₀	24.66 mn	26.73 hijk	23.46 no	24.06 mno	28.80 gh	25.54 e
RS ₁	27.76 ghi	29.70 def	26.00 kl	26.80 hijk	32.50 b	28.55 b
RS ₂	26.50 jk	27.56 ghij	24.30 mno	25.13 lm	30.76 cd	26.85 c
RS ₃	29.56 def	30.50 cde	26.73 hijk	28.80 fg	33.76 a	29.87 a
RS ₄	26.56 ijk	26.66 ijk	23.40 o	24.60 mno	29.26 ef	26.10 d
RS ₅	31.16 c	30.36 cde	26.93 hijk	27.93 gh	33.40 ab	29.96 a
Mean	27.70 c	28.58 b	25.13 e	26.22 d	31.41 a	

Means followed by different letter is a comparison are significant at $P < 0.05$.

LSD for Treatments = 0.339, LSD for Varieties = 0.554, LSD for Treatments* Varieties = 1.358, CV = 2.97

Table 2. Interactive effect of *rhizobium* strains and lentil varieties on number of branches plant⁻¹

Rhizobial strains	Varieties					Mean
	NIA-Masoor-2005	Markaz-2009	Punjab-Masoor-2009	Chakwal-Masoor-2011	NIA-Masoor-2016	
RS ₀	2.82 fgh	3.25 defg	3.17 defg	2.73 gh	3.67 cd	3.13 c
RS ₁	2.90 efgh	3.42 cdef	2.92 efgh	3.25 defg	3.67 cd	3.23 c
RS ₂	3.25 defg	2.80 fgh	2.73 gh	2.40 h	4.50 ab	3.14 c
RS ₃	3.42 cdef	3.50 cde	3.25 defg	3.25 defg	4.50 ab	3.58 ab
RS ₄	2.92 efgh	2.73 gh	2.90 efgh	3.25 defg	3.92 bc	3.14 c
RS ₅	3.58 cd	3.50 cde	3.25 defg	3.50 cde	5.00 a	3.77 a
Mean	3.15 b	3.20 b	3.04 b	3.06 b	4.38 a	

LSD for Treatments = 0.3516, LSD for Varieties = 0.2472, LSD for Treatments* Varieties = 0.6055, CV = 10.96

*Means followed by different letter is a comparison are significant at $P < 0.05$.

Data presented in the table (3) indicated that RS₅ with NIA-Masoor-2016 produced the plants with maximum number of pod plant⁻¹ (47.99) that was statistically at par with RS₃ with the same lentil varieties measuring number of pod plant⁻¹ (46.33). Furthermore, the minimum number of pod plant⁻¹ was recorded in RS₀ with Punjab-Masoor 2009 (32.22). The increased plant height with *rhizobial* strains might be due to the improving a growth and nutrient uptake. (Huang and Erickson, 2007) also reported the similar finding that application of *rhizobium* inoculation significantly increased the number of pod plant⁻¹ associated with in enhanced soil fertility and nutrient uptake. Moreover, the difference in number of pod plant⁻¹ among the lentil varieties might be due to the genotypic variability. These results were in line (Gull et al., 2004) where they reported the increase number of pod plant⁻¹ is highly associated with varietal traits.

Fresh Shoot Weight Plant⁻¹

Data regarding to fresh shoot weight plant⁻¹ of lentil crop was significantly increased due to inoculation with *rhizobial* strains. Data (Table 4) regarding fresh shoot weight plant⁻¹ as affected by different *rhizobial* strains and lentil varieties showed significant effect ($P < 0.05$). The results of the present study indicated that the maximum fresh shoot weight plant⁻¹ (6.88) were recorded with RS₅ followed by RS₃ (6.62). However, the lowest fresh shoot weight plant⁻¹ (5.03) was recorded with RS₀ (control). Furthermore, it was also clear from the present results that are RS₅ significantly produced the plants with more fresh shoot weight plant⁻¹ than all the other treatments. Similar result was reported by (Islam et al., 2013) he stated that the fresh weight of shoot plant⁻¹ of all inoculated plots was higher than uninoculated.

Table 3. Interactive effect of *rhizobium* strains and lentil varieties on number of pods plant⁻¹

Rhizobial strains	Varieties					Mean
	NIA-Masoor-2005	Markaz-2009	Punjab-Masoor-2009	Chakwal-Masoor-2011	NIA-Masoor-2016	
RS ₀	35.55 jkl	41.10 fgh	32.22 m	35.88 jk	43.22 cdef	37.20 c
RS ₁	37.33 ij	42.77 def	33.55 lm	36.66 ij	44.77 bcd	39.02 b
RS ₂	36.22 j	43.55 cde	33.99 klm	35.66 jkl	42.99 cdef	38.48 bc
RS ₃	40.22 gh	46.22 ab	41.33 efg	40.22 gh	46.33 ab	42.86 a
RS ₄	36.33 j	42.55 def	33.77 m	37.33 ij	43.44 cde	39.12 b
RS ₅	38.88 hi	45.22 bc	43.22 cdef	42.55 def	47.99 a	43.57 a
Mean	37.42 c	43.57 b	36.38 d	38.05 c	44.79 a	

LSD for Treatments = 1.1272, LSD for Varieties = 0.9061, LSD for Treatments* Varieties = 2.2195, CV = 3.38

*Means followed by different letter is a comparison are significant at $P < 0.05$.

Table 4. Interactive effect of *rhizobium* strains and lentil varieties on fresh shoot weight plant⁻¹ (g)

Rhizobial strains	Varieties					Mean
	NIA-Masoor-2005	Markaz-2009	Punjab-Masoor-2009	Chakwal-Masoor-2011	NIA-Masoor-2016	
RS ₀	5.30 kl	6.47 ef	2.92 n	4.94 l	5.51 jk	5.03 d
RS ₁	5.64 ijk	6.39 efg	3.20 n	5.24 kl	7.15 bc	5.52 c
RS ₂	5.83 hij	6.53 ef	3.15 n	5.28 kl	7.35 bc	5.63 c
RS ₃	6.98 cd	7.42 bc	4.47 m	5.98 ghi	8.26 a	6.62 b
RS ₄	5.89 hij	6.63 de	3.20	5.29 kl	7.42 b	5.69 c
RS ₅	7.32 bc	7.13 bc	5.24 kl	6.12 fgh	8.61 a	6.88 a
Mean	6.16 c	6.76 b	3.70 e	5.48 d	7.39 a	

LSD for Treatments = 0.2023, LSD for Varieties = 0.1778, LSD for Treatments* Varieties = 0.4354, CV = 7.49

*Means followed by different letter is a comparison are significant at $P < 0.05$

Similarly, among the lentil varieties the highest fresh shoot weight plant⁻¹ was recorded in NIA-Masoor-2016 (7.39)

while the lowest fresh shoot weight plant⁻¹ was recorded in Punjab-Masoor-2009 (3.70).

The interactive effect of *rhizobial* strains and lentil varieties were also statistically significant ($P < 0.005$). Data presented in the table (4) indicated that RS₅ with NIA-Masoor-2016 produced the plants with maximum fresh shoot weight plant⁻¹ (8.61) that was statistically at par with RS₃ with the same lentil varieties measuring fresh shoot weight plant⁻¹ (2.26). Furthermore, the minimum fresh shoot weight plant⁻¹ was recorded in RS₀ with Punjab-Masoor-2009 (2.92). It was also clear from the present results that interactive effect of *rhizobial* strains and lentil varieties produced the plants with more fresh shoot weight plant⁻¹ than control which might have resulted in higher nitrogen fixation and consequently higher dry matter production.

Dry Shoot Weight Plant⁻¹ (g)

Data regarding to dry shoot weight plant⁻¹ of lentil crop was significantly increased due to inoculation with *rhizobial* strains. Data (Table 5) regarding dry shoot weight plant⁻¹ as affected by different *rhizobial* strains and lentil varieties showed significant effect ($P < 0.05$). The results of the present study indicated that the maximum dry shoot weight plant⁻¹ (1.97 g) were recorded with RS₅ followed by RS₃ (1.89). However, the lowest dry shoot weight plant⁻¹ (1.44) was recorded with RS₀ (control). Furthermore, it was also clear from the present results that are RS₅ significantly produced the plants with more dry shoot weight plant⁻¹ than all the other treatments. Similar result was reported by (Islam et al., 2013) he stated that the dry weight of shoot plant⁻¹ of all inoculated plots was higher than uninoculated. Similarly, among the lentil varieties the highest dry shoot weight plant⁻¹ was recorded in NIA-Masoor-2016 (2.11) while the lowest dry shoot weight plant⁻¹ was recorded in Punjab-Masoor-2009 (1.05).

The interactive effect of *rhizobial* strains and lentil varieties were also statistically significant ($P < 0.005$). Data presented in the table (5) indicated that RS₅ with NIA-Masoor-2016 produced the plants with maximum dry shoot weight plant⁻¹ (2.46) that was statistically at par with RS₃ with the same lentil varieties measuring dry shoot weight plant⁻¹ (2.36). Furthermore, the minimum dry shoot weight plant⁻¹ was recorded in RS₀ with Punjab-Masoor-2009 (0.83). According to Zafar et al. (2012) he stated that application of *microbial* strains greatly increases dry weight plant⁻¹. Similar result was reported by (Singh et al., 2016) he stated that application of microbial strains improves dry weight plant⁻¹ over control.

Table 5. Interactive effect of *rhizobium* strains and lentil varieties on dry shoot weight plant⁻¹ (g)

Rhizobial strains	Varieties					Mean
	NIA-Masoor-2005	Markaz-2009	Punjab-Masoor-2009	Chakwal-Masoor-2011	NIA-Masoor-2016	
RS ₀	1.52 kl	1.84 ef	0.83 n	1.41 l	1.58 jk	1.44 d
RS ₁	1.61 ijk	1.82 efg	0.91 n	1.50 kl	2.05 bc	1.58 c
RS ₂	1.67 hij	1.86 ef	0.90 n	1.51 kl	2.10 bc	1.61 c
RS ₃	2.00 cd	2.11 bc	1.28 m	1.71 ghi	2.36 a	1.89 b
RS ₄	1.68 hij	1.89 de	0.91	1.51 kl	2.13 b	1.62 c
RS ₅	2.09 bc	2.03 bc	1.50 kl	1.75 fgh	2.46 a	1.97 a
Mean	1.76 c	1.92 b	1.05 e	1.56 d	2.11 a	

LSD for Treatments = 0.0579, LSD for Varieties = 0.0508, LSD for Treatments* Varieties = 0.1245, CV = 4.49

*Means followed by different letter is a comparison are significant at $P < 0.05$.

Fresh Root Weight Plant⁻¹ (g)

Data (Table 6) regarding fresh root weight plant⁻¹ as affected by different *rhizobial* strains and lentil varieties showed significant effect ($P < 0.05$). The results of the present study indicated that the maximum fresh root weight plant⁻¹ (1.37) were recorded with RS₅ which was statistically at par with RS₃ (1.31). However, the lowest fresh root weight plant⁻¹ (0.91) was recorded with RS₀ (control). Furthermore, it was also clear from the present results that RS₅ significantly produced the plants with more fresh root weight plant⁻¹ than all the other treatments.

Similarly, among the lentil varieties the highest fresh root weight plant⁻¹ was recorded in NIA-Masoor-2016 (1.29) while the lowest fresh root weight plant⁻¹ was recorded in Chakwal-Masoor-2011 (1.03) which was statistically at par with NIA-Masoor-2005 (1.03). The interactive effect of rhizobial strains and lentil varieties were also statistically significant ($P < 0.005$). Data presented in the table (7.6) indicated that RS5 with NIA-Masoor-2016 produced the plants with maximum fresh root weight plant⁻¹ (1.65) that was statistically at par with RS3 with the same lentil genotype measuring fresh shoot weight plant⁻¹ (1.62). Furthermore, the minimum fresh root weight plant⁻¹ was recorded in RS0 with Punjab-Masoor-2009 (0.85). Similar result was reported by (Zafar et al., 2012) he stated that application of microbial strains increases fresh root weight plant⁻¹.

Table 6. Interactive effect of *rhizobium* strains and lentil varieties on fresh root weight plant⁻¹ (g)

Rhizobial strains	Varieties					Mean
	NIA-Masoor-2005	Markaz-2009	Punjab-Masoor-2009	Chakwal-Masoor-2011	NIA-Masoor-2016	
RS ₀	0.89 kl	0.85 l	0.85 l	0.90 jkl	1.09 efgh	0.91 c
RS ₁	0.95 ijkl	1.03 fg hi	0.89 kl	0.94 jkl	1.12 defg	0.98 b
RS ₂	1.01 ghijk	1.08 efgh i	0.31 ijkl	0.97hijkl	1.12 defg	1.02 b
RS ₃	1.15 def	1.35 bc	1.25 cd	1.17 de	1.62 a	1.31 a
RS ₄	1.03 fghij	1.09efgh	0.98 hijkl	0.95 ijkl	1.14 defg	1.03 b
RS ₅	1.17 de	01.43 b	1.35 bc	1.25 cd	1.65 a	1.37 a
Mean	1.03 c	1.14 b	1.04 c	1.03 c	1.29 a	

LSD for Treatments = 0.0627, LSD for Varieties = 0.0554, LSD for Treatments* Varieties = 0.1358, CV = 4.50

*Means followed by different letter is a comparison are significant at $P < 0.05$.

Dry Root Weight Plant⁻¹ (g)

Data (Table 7) regarding dry root weight plant⁻¹ as affected by different *rhizobial* strains and lentil varieties showed significant effect ($P < 0.05$). The results of the present study indicated that the maximum dry root weight plant⁻¹ (0.45) were recorded with RS₅ which was statistically at par with RS₃ (0.43). However, the lowest dry root weight plant⁻¹ (0.30) was recorded with RS₀ (control). Furthermore, it was also clear from the present results that RS₅ significantly produced the plants with more dry root weight plant⁻¹ than all the other treatments.

Similarly, among the lentil varieties the highest dry root weight plant⁻¹ was recorded in NIA-Masoor-2016 (0.43) while the lowest dry root weight plant⁻¹ was recorded in Chakwal-Masoor-2011 (0.34) which was statistically at par with NIA-Masoor-2005 and Punjab-Masoor-2009.

The interactive effect of rhizobial strains and lentil varieties were also statistically significant ($P < 0.005$). Data presented in the table (7) indicated that RS₅ with NIA-Masoor-2016 produced the plants with maximum dry root weight plant⁻¹ (0.55) that was statistically at par with RS₃ with the same lentil genotype measuring dry shoot weight plant⁻¹ (0.54). Furthermore, the minimum dry root weight plant⁻¹ was recorded in RS₀ with Punjab-Masoor-2009 (0.28).

Number of Nodules Plant⁻¹

Data (Table 8) regarding number of nodules plant⁻¹ as affected by different rhizobial strains and lentil varieties showed significant effect ($P < 0.05$). The results of the present study indicated that the maximum number of nodules plant⁻¹ (23.00) were recorded with RS₅ followed by RS₃ (21.07). However, the lowest number of nodules plant⁻¹ (17.14) was recorded with RS₀ (control). Furthermore, it was also clear from the present results that are RS₅ significantly produced the plants with more number of nodules plant⁻¹ than all the other treatments.

Similarly, among the lentil varieties the highest number of nodules plant⁻¹ was recorded in NIA-Masoor-2016 (24.96) while the lowest number of nodules plant⁻¹ was recorded in Punjab Masoor-2009 (16.07) which was statistically at par with Chakwal-Masoor-2011 (16.46).

The interactive effect of *rhizobial* strains and lentil varieties were also statistically significant ($P < 0.005$). Data

presented in the table (8) indicated that RS₅ with NIA-Masoor-2016 produced the plants with maximum number of nodules plant⁻¹ (28.67) followed by RS₃ with the same lentil genotype measuring number of nodules plant⁻¹. Furthermore, the minimum number of nodules plant⁻¹ was recorded in RS₀ with Punjab-Masoor-2009 (14.22) that was statistically at par with Punjab- Masoor-2009. Number of nodules plant⁻¹ increased due to varietal trait and inoculation with *rhizobial* strains.

Table 7. Interactive effect of *rhizobium* strains and lentil varieties on dry root weight plant⁻¹ (g)

Rhizobial strains	Varieties					Mean
	NIA-Masoor-2005	Markaz-2009	Punjab-Masoor-2009	Chakwal-Masoor-2011	NIA-Masoor-2016	
RS ₀	0.29 kl	0.29 l	0.28 l	0.30 jkl	0.36 efgh	0.30 c
RS ₁	0.31 ijkl	0.34 efghi	0.30 kl	0.31 ijkl	0.37 defg	0.32 b
RS ₂	0.33 ghijk	0.36 efgh	0.31 ijkl	0.32 hijkl	0.37 defg	0.34 b
RS ₃	0.38 def	0.45 bc	0.41 cd	0.39 de	0.54 a	0.43 a
RS ₄	0.34 fghij	0.36 efgh	0.32 hijkl	0.31 ijkl	0.38 defg	0.34 b
RS ₅	0.39 de	0.48 b	0.45 bc	0.41 cd	0.55 a	0.45 a
Mean	0.34 c	0.38 b	0.34 c	0.34 c	0.43 a	

LSD for Treatments = 0.0210, LSD for Varieties =0.0185, LSD for Treatments* Varieties =0.0454, CV = 7.48

*Means followed by different letter is a comparison are significant at P<0.05.

Table 8. Interactive effect of *rhizobium* strains and lentil varieties on number of nodules plant⁻¹

Rhizobial strains	Varieties					Mean
	NIA-Masoor-2005	Markaz-2009	Punjab-Masoor-2009	Chakwal-Masoor-2011	NIA-Masoor-2016	
RS ₀	15.55 jk	19.16 efg	14.22 k	14.22 k	22.66 d	17.14 d
RS ₁	18.00 gh	20.11 e	15.11 jk	15.66 ijk	23.77 cd	18.53 c
RS ₂	19.89 e	19.66 ef	15.12 jk	16,12 ij	24.22 cd	19.00 c
RS ₃	19.78 e	23.11 d	17.22 hi	18.22 fgh	27.00 b	21.07 b
RS ₄	19.00 efg	20.11 e	15.44 jk	15.33 jk	23.44 d	18.66 c
RS ₅	22.66 d	25.11 c	19.33 efg	19.22 efg	28.67 a	23.00 a
Mean	19.15 c	21.19 b	16.07 d	16.46 d	24.96 a	

LSD for Treatments = 0.8046, LSD for Varieties =0.6329, LSD for Treatments* Varieties =1.5502, CV = 4.83

*Means followed by different letter is a comparison are significant at P<0.05.

Nitrogen contents of the shoot (ppm)

Data (Table 9) regarding nitrogen contents of the shoot as affected by different *rhizobial* strains and lentil varieties showed significant effect (P < 0.05). The results of the present study indicated that the maximum nitrogen contents of the shoot (2.75) were recorded with RS₅ followed by RS₃ (2.67). However, the lowest nitrogen contents of the shoot (2.44) were recorded with RS₀ (control). Furthermore, it was also clear from the present results that are RS₅

significantly produced the plants with more nitrogen contents of the shoot than all the other treatments. Similarly, among the lentil varieties the highest nitrogen contents of the shoot was recorded in NIA-Masoor-2016 (2.77) while the lowest nitrogen contents of the shoot were recorded in Chakwal-Masoor-2011 (2.34). The interactive effect of *rhizobial* strains and lentil varieties were also statistically significant ($P < 0.005$). Data presented in the table (9) indicated that RS₅ with NIA-Masoor-2016 produced the plants with maximum nitrogen contents of the shoot (2.94) followed by RS₄ with the same lentil varieties measuring nitrogen contents of the shoot (2.85). Furthermore, the minimum nitrogen contents of the shoot were recorded in RS₀ with Chakwal-Masoor-2011 (2.17). Nitrogen contents of the shoot of lentil varieties increased due to varietal trait and inoculation with *rhizobial* strains. These results are in line with those of (Gull et al., 2004). (Singh et al., 2018) also reported the similar finding that application of *rhizobium* inoculation significantly increased the nitrogen contents of the shoot associated with in enhanced plant growth and nutrient uptake. These results were in line with those of (Rana and Solanki, 2015)) have reported similar variations in the crop growth potential of different genotypes of the lentils.

Table 9. Interactive effect of *rhizobium* strains and lentil varieties on nitrogen content of shoot plant⁻¹

Rhizobial strains	Varieties					Mean
	NIA-Masoor-2005	Markaz-2009	Punjab-Masoor-2009	Chakwal-Masoor-2011	NIA-Masoor-2016	
RS ₀	2.31 pq	2.52 klm	2.51 lm	2.17 s	2.69 ef	2.44 f
RS ₁	2.40 o	2.59 hij	2.56 ijkl	2.23 r	2.70 ef	2.49 e
RS ₂	2.43 mn	2.61 ghi	2.63 gh	2.26 qr	2.73 de	2.53 d
RS ₃	2.55 jkl	2.72 e	2.74 de	2.47 mn	2.85 b	2.67 b
RS ₄	2.47 mn	2.63 gh	2.66 fg	2.33 p	2.73 de	2.56 c
RS ₅	2.66 fg	2.78 cd	2.80 bc	2.58 hijk	2.93 a	2.75 a
Mean	2.47 c	2.64 b	2.66 b	2.34 d	2.77 a	

LSD for Treatments = 0.0191, LSD for Varieties = 0.0237, LSD for Treatments* Varieties = 0.0580, CV = 1.37

*Means followed by different letter is a comparison are significant at $P < 0.05$.

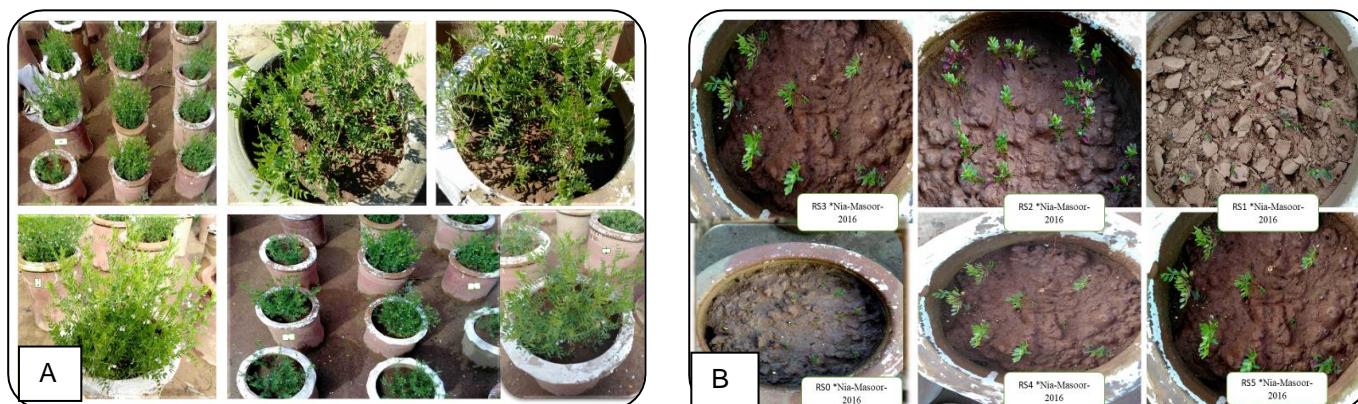


Figure 1. (A) General view of the experiment at flowering stage (interaction of rhizobial strains and lentil genotypes), (B) General view of the experiment at germination stage (interaction of rhizobial strains and lentil varieties)

CONCLUSION

It was concluded that the (NIA-Masoor- 2016) and variety, V₃ (Punjab- Masoor-2011) have a very high yield potential and that these varieties can be further evaluated for zoning success in Pothwar. Consequently, *rhizobium* strains contributed to improved soil productivity and could minimize the cost of production of next crop by reducing inputs in the

form of nitrogen fertilizers. It has been found that *rhizobic* inoculation is more productive and better yields, as compared to control. Such promising varieties will eventually contribute to the production of new commercial lentil varieties.

AUTHOR CONTRIBUTIONS

All authors contributed equally to this research.

COMPETING OF INTEREST

The authors declare no competing interests.

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