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Integrated Effects of Rhizobial Inoculum and Inorganic Fertilizers on Wheat Yield and Yield Components

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Abstract

An experiment was conducted in pots under natural condition with two factor factorial completely randomized design (CRD) (CRD) to investigate the integrated effect of rhizobia inoculums and inorganic fertilizers on growth and yield of wheat crop at The University of Agriculture Peshawar, during 2012-2013. The experiment was comprised of four inoculums (no, lentil, peas and chickpeas) and two NPK levels (recommended 120:90:60 kg·ha^{-1} & 20% less of recommended). It was observed that inoculation of wheat by rhizobia significantly increased tillers per plant by a maximum of 42%, plant height by 13%, grain per spike by 16%, 100 grain weight by 10%, biological and grain yield by 10% over un-inoculated (control) treatment. Among inoculums, peas inoculum was found to be the most efficient for all traits except plant height where chickpea inoculum performed better. Similarly recommended NPK significantly increased tillers per plant by 33%, plant height by 19%, grain per spike by 9%, 100 grain weight by 10%, biological yield by 8% and grain yield by 10% compared with 20% less of recommended NPK. Interactive effect of inoculum x NPK was significant for tillers per plant, grain per spike, grain yield and non-significant for plant height, 100 grain weight and biological yield. However, it was evident from the results that inoculation improved all traits both under recommended and 20% less of recommended NPK. The persistent good performance of peas and lentil rhizobial inoculation in wheat growth exhibited that this could be used as a plant growth promoting rhizobacteria for wheat and other cereal crops in prevailing soil and climatic conditions.

Keywords

Yield Components, Rhizobial Inoculum, Wheat, Recommended NPK, Inorganic Fertilizer

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1. Introduction

Wheat belongs to family *Poaceae* and genus *Triticum* and ranks third after maize and rice. On a worldwide scale, wheat covers about 30% of total cereal products [1]. Pakistan is the ninth largest wheat producing country in the world [2]. Wheat is grown on major areas in Pakistan but its average yield at farmers' fields is still less than the potential [3]. To increase the yields the farmers use mineral fertilizers [4], which are not only very costly but also act as pollutants in many ways. Throughout the world, researchers are facing these threats and they are trying to control this situation by producing alternative substitutes which are more economical, environment friendly and bring sustainable improvement. The best approach to improve crop yield is to use beneficial microbes or plant growth promoting rhizobacteria [5]. Rhizobia can be used as plant growth promoting rhizobacteria (PGPR) in non-legumes [6]. It enhances growth and yield of cereals by production of phytohormones IAA, gibberellins and cytokinins [7], siderophore (iron chelator) [8], enzymes [9], and improves supply of insoluble nutrients such as phosphorus [10] and iron by producing organic acids making these nutrients easily available for plants uptake [11]. It also helps to kill pathogens by producing antibiotics [12], HCN [13] and expolysaccharides [14], and improves morphological characteristics of inoculated roots [15] which increase nutrient and water use efficiency [16], mobilization and efficient uptake of nutrients [11]. Likewise, it increases stress resistance and induction of systemic stress [17] and drought tolerance/resistance by releasing substances like abscisic acid [18] or lumichrome which reduce leaf stomatal opening and decrease transpirational water losses from plants through the leaves. It also improves N uptake in rice plants [6]. Studies conducted so far regarding the impact of *Rhizobium* application for cereals are limited and showed specificity of different *Rhizobium* strains towards cultivar, soil, and environment. Therefore, this pot study was conducted to assess the potential of three *Rhizobium* species isolated from the root nodules of three locally growing rabbi legumes including peas, lentil and chickpea with two NPK levels (basal dose of NPK and 20% less than basal of dose NPK) for improving the yield of wheat under greenhouse conditions.

2. Materials and Methods

A pot experiment was conducted to investigate the integrated effect of different rhizobial species isolated from winter legumes and inorganic fertilizers on growth and yield of wheat crop at the University of Agriculture, Peshawar during winter 2012-2013. The experiment was comprised of four levels of rhizobial inoculums (control, lentil, peas and chick peas inoculum) and two levels of NPK fertilizers (120:90:60 and 96:72:48 kg·ha^{-1}) making a total of 8 treatments per replication. The experiment was laid out in Completely Randomized Design with three replications. Pots having 10cm radius and 30 cm depth were filled with five kg soil. At the time of sowing, pots were amended with required amount of P, K and half of N fertilizer in solution form, 23 g of Urea, 24.5 g of DAP and 15 g SOP were dissolved in 500 ml solution. In this way, 10 and 8 ml of prepared solution was added to pots receiving basal dose and 20% less than basal dose of NPK, respectively. Half of N fertilizer was applied at tillering stage in the same manner. Rhizobial inoculum of lentil, peas and chick pea were obtained from National Agricultural Research Council, Islamabad. The seeds of wheat variety Siran were soaked in 20% concentrated sugar solution, inoculated with required but equal amount of rhizobial inoculums, dried in shade and were sown @ of 8 seeds per pot . Thinning of the pots was performed after 14 days of emergence to maintain 4 seedlings per pot. The pots were irrigated with tap water as per moisture condition. Pots were randomized on weekly basis to avoid side, shade and temperature effect. Data were recorded on agronomic parameter including tillers per plant, plant height, grain per spike, 100 grain weight, biological and grain yield. Grain and biological yield per pot were converted into Kg·ha^{-1} on the basis of soil mass Kg·ha^{-1} (as soil mass for upper 15 cm per hectare is 2×10^6 kg). Data recorded were statistically analyzed using ANOVA technique appropriate for completely randomized design and means were compared using LSD test at 5% level of probability [19].

3. Results and Discussions

A pot study was carried out in two factor factorial completely randomized design (CRD) to investigate the integrated effect of rhizobia inoculums and inorganic fertilizers on growth and yield of wheat crop. The soil used for this experiment was silty loam in texture, alkaline in reaction, low in organic matter (0.73%), highly calcareous in nature (13.5%), non-saline (0.46 d·sm^{-1}), and low in total nitrogen (0.08%) and AB-DTPA extractible Phosphorous (2.25 mg·kg^{-1}) contents (**Table 1**, **Table 2**).

Table 1. Physico-chemical characteristics of soil under investigations.

Property	Units	Concentration
Textural class	-	Silt loam
pH	-	7.56
Electrical conductivity (EC_e)	$d\cdot sm^{-1}$	0.46
Lime	%	13.5
Organic matter content	%	0.73
Total nitrogen content	%	0.08
AB-DTPA extractable P	$mg\cdot kg^{-1}$	2.25

3.1. Tillers per Plant

Significant differences were observed among inoculum for tillers per plant (**Table 3**). Maximum mean tillers per plant of 3.49 was recorded for peas inoculum followed by 3.44 of lentil and 3.05 of chick pea while, minimum mean tillers per plant of 2.77 was recorded for non-inoculated control treatment. These results demonstrated that inoculated treatments produced significantly higher (up to 42%) mean tillers per plant than non-inoculated treatments suggested that *Rhizobium* inoculation had increasing and positive impact on tillers per plant in wheat as reported by [20]. Significant variation of mean tillers per plant in inoculated treatments showed that inoculums varied in their potential as PGPR (plant growth promoting rhizobacteria) for non-leguminous crop as reported by [21] and it may be due to their adoptability to prevailing soil and climatic conditions as reported by [22] who found that *Rhizobium* strains behave differently according to the soil used. Recommended NPK produced significantly higher (up to 33%) tillers per plant than 20% less NPK, it may be due to the inadequacy of NPK in treatments with 20% less NPK. Interactive effect of inoculum × NPK showed (**Figure 1**) significantly higher,maximum mean tillers per plant of 4.46 for treatment of pea's inoculum with recommended NPK while minimum mean tillers per plant of 2.44 were recorded for un-inoculated treatment with 20% less NPK. Un-inoculated treatment with full NPK produced mean tillers per plant of 2.60 that were statistically similar to that of 2.33, 2.33 and 2.27 for inoculated treatments of peas, chickpea and lentil with 20% less NPK respectively. These results suggested that inoculation of wheat seed with Peas, chickpeas and Lentil Inoculums can contribute 20% NPK to the system and thus may reduce the use of mineral NPK fertilizer up to 20% for wheat crop under prevailing soil conditions, this may be due to the ability of *Rhizobium* inoculums to act as PGPR (plant growth promoting rhizobacteria), as it solubilize/mobilize and bind nutrients from organic and inorganic sources as reported by [23].

3.2. Plant Height (cm)

Significant differences were recorded in inoculum for plant height (**Table 3**). Plant height of 73.33, 69.99 and 72.66 cm were recorded for chickpeas, peas and lentil inoculum respectively which were statistically at par, however minimum plant height of 64.66 was observed for control. These results revealed that inoculation enhanced plant height by 9% over non-Inoculated control. [24] also observed increase in wheat plant height by rhizobial inoculation. Treatments with full NPK showed significantly higher plant height of 72.25 cm than that of 68 cm of 20% less NPK, this may be due to the inadequate availability of NPK at 20% less NPK. Interaction of inoculum and NPK for plant height was non-significant but it showed a trend which demonstrated that inoculation increase plant height at both recommended and 20% less of recommended NPK. These findings suggested that inoculation of rhizobia along with full NPK is more effective than inoculation with 20% less NPK.

3.3. Grain per Spike

Significant differences were observed in mean grains per spike for main effect of inoculum (**Table 4**). Maximum mean grains per spike of 43.77 were recorded for peas inoculum. Lentil inoculum showed 41.78 grain per spike which was significantly higher than that of 40.66 grain per spike of chickpea and lower than that of peas

Table 2. ANOVA and Sum of Squares of Tillers per plant, Plant height, Grain per spike, 100 grain weight, Grain yield and Biological yield.

SOV	DF	Sum of Square					
		Tillers per plant	Plant Height	Grain per Spike	100 grain weight	Grain yield	Biological yield
Inoculums	3	4.64**	279.33**	112.39**	0.618**	501,965**	3,733,736**
NPK	1	5.34**	104.17**	61.25**	1.219**	739,206**	2,250,325**
Inoculums* NPK	3	0.76**	7.30*	4.73**	0.082*	204,637**	109,634*
Error	16	1.02	66.47	5.84	0.718	191,584	2,110,745
Total	23	11.78	457.27	184.2	2.637	1,637,391	8,204,440
CV	-	7.66	2.91	1.55	4.49	1.00	4.47

**Significant and *Non Significant.

Table 3. Tillers per plant and plant height as influenced by applied treatments.

Inoculum	Tillers per	% Increase	Plant height	% Increase
	Plant	Decrease	(cm)	Decrease
Control	2.77c	-	64.66b	-
Lentil	3.44b	24	72.66a	12
Peas	3.49a	42	69.99a	8
Chickpeas	3.05c	10	73.33a	13
LSD (0.05)	0.31		3.4	
NPK				
Full	3.77a	-	72.25a	-
20% less	2.83b	−33	68.00b	−19
Sig. level	*		*	
			I*NPK Interaction	
Sig. level	*(Figure 1)		NS	

*Means with different letter(s) in columns are significantly different at $P \leq 0.05$.

Table 4. Grain per spike and 100 grain weight as influenced by applied treatments.

Inoculum	Grain per	% Increase	100 grain weight	% Increase
spike	Decrease	(g)	Decrease	
Control	37.55d	-	4.51b	-
Lentil	41.78b	8	4.76ab	6
Peas	43.77a	16	4.95a	10
Chickpeas	40.66c	4	4.65a	3
LSD (0.05)	0.74		0.26	
NPK				
Full	40.66a	-	4.94a	-
20% less	37.46b	−9	4.49b	−10
Sig. level	*		*	
		I*NPK Interaction		
Sig. level	*(Figure 2)		NS	

*Means with different letter(s) in columns are significantly different at $P \leq 0.05$.

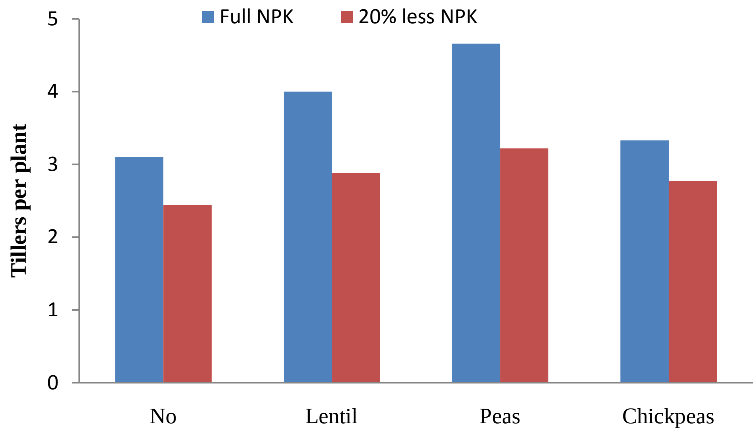

Figure 1. Tillers per plant as influenced by interaction of inoculum and NPK.

inoculum. Minimum mean grains per spike of 37.55 were recorded for non-inoculated (control) treatment. These results showed that wheat seed inoculated with peas *Rhizobium* inoculum produced significantly higher mean grains per spike (up to 16%) than non-inoculated treatments, thus suggested that *Rhizobium* inoculation had increasing and positive impact on grains per spike in wheat as reported by [25]. Significant variation of mean grains per spike in inoculated treatments showed that inoculums varied in their potential as PGPR (plant growth promoting rhizobacteria) for non-leguminous crop as reported by [21] and it may be due to their adoptability to prevailing soil and climatic conditions as reported by [22] who found that *Rhizobium* strains behave differently according to the soil used. Recommended NPK produced significantly higher mean grains per spike of 40.66 than 37.44 of 20% less NPK, it may be due to the inadequacy of NPK in treatments with 20% lees of recommended NPK. Interaction of inoculum and NPK was significant for grain per spike (**Figure 2**). On the average maximum grain per spike of 43.77 were recorded for Peas inoculum with full recommended NPK while minimum grain per spike of 35.55 was recorded for control (no inoculum) with 20% less of recommended dose of NPK. It was also evident from the result that inoculation of rhizobia increased grain per spike at both recommended and 20% less of recommended NPK.

3.4. 100 Grains Weight

Data recorded for mean 100 grain weight (**Table 4**) showed significant differences for main effect of inoculums, NPK and non-significant variation for the interaction of inoculum and NPK. Maximum mean 100 grain weight of 4.95 g was recorded for peas inoculum followed by 4.76 g of lentil and 4.65 g of chick pea while minimum mean 100 grain weight of 4.51 g was recorded for non-inoculated (control) treatments. These results demonstrated that inoculated treatments produced significantly higher (up to 10%) 100 g weight than non-inoculated treatments suggested that *Rhizobium* inoculation had increasing and positive impact on 100 grain weight as reported by [19]. Significant variation of mean 100 grain weight in inoculated treatments showed that inoculums varied in their potential as PGPR (plant growth promoting rhizobacteria) for non-leguminous crop as reported by [21]. Full NPK produced significantly higher (up to 10%) mean 100 grain weight of 4.94 g than 4.49 g of 20%less NPK. The interactive effect of inoculum and NPK was non-significant but showed a trend that inoculation of wheat by rhizobia enhanced 100 grain weight at both full and 20% less of recommended NPK.

3.5. Grain Yield

Differences recorded for inoculum, NPK and their interaction were significant (**Table 5**). Maximum grain yield of 2949 $kg·ha^{-1}$ was recorded for peas inoculum followed by 2769 $kg·ha^{-1}$ of lentil, 2616 $kg·ha^{-1}$ of chickpeas while minimum grain yield of 2588 $kg·ha^{-1}$ was recorded for un-inoculated treatments. These results demonstrated that inoculated treatments enhanced grain yield (up to 14%) significantly over non-Inoculated treatments, same results were also found by [26]. Significant variation among the inoculums for grain yield may be due to variation in the adaptability of inoculum to prevailing soil and climatic condition as reported by [22]. Treatments with full NPK showed significantly higher (up to 14%) grain yield of 2909 $kg·ha^{-1}$ than that of 2558 $kg·ha^{-1}$ of

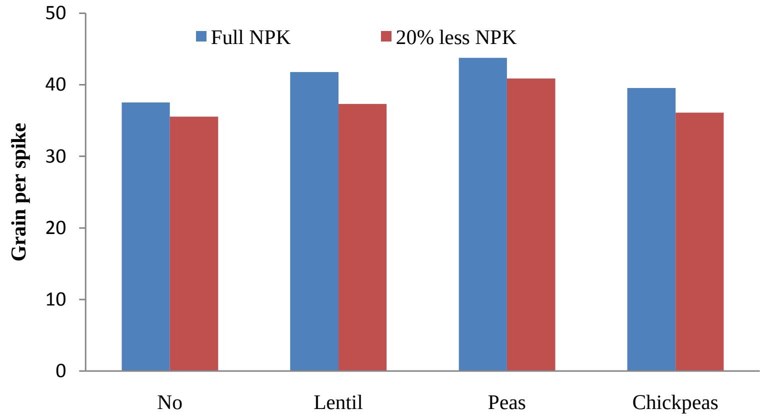

Figure 2. Grain per spike as influenced by interaction of inoculum and NPK.

Table 5. Grain and biological yield of wheat as influenced by applied treatments.

Inoculum	Grain yield	% Increase	Biological yield	% Increase
	(Kg·ha^{-1})	Decrease	(kg·ha^{-1})	Decrease
Control	2588c	-	7479b	-
Lentil	2769b	7	8245a	10
Peas	2949a	14	8650a	14
Chickpeas	2616c	1	8231a	10
LSD (0.05)	133.93		444	
NPK				
Full	2909a	-	8433a	-
20% less	2558b	−14	7820b	−8
Sig. level	*		*	
		I*NPK Interaction		
Sig. level	*(Figure 3)		NS	

*Means with different letter(s) in columns are significantly different at P ≤ 0.05.

20% less NPK, this may be due to the inadequate availability of NPK at 20% less NPK. Significant interactive effect of inoculum and NPK exhibited that, on the average maximum plant grain yield of 3280 were recorded for Peas inoculum with full recommended NPK, while minimum of 2475 was recorded for control (no inoculum) with 20% less of recommended dose of NPK. It was also evident from the result that inoculation of rhizobia increased grain yield at both recommended and 20% less of recommended NPK. These results suggested that *Rhizobium* inoculation can be used as PGPR (plant growth promoting rhizobacteria) for wheat crop in prevailing soil and climatic conditions.

3.6. Biological Yield

Variations observed for biological yield as affected by inoculum and NPK were significant. Statistically at par biological yield of 8650, 8245 and 8231 kg·ha^{-1} were recorded for peas, lentil and chickpeas inoculums respectively, while minimum of 7479 kg·ha^{-1} was recorded for un-inoculated (control) treatment. These results demonstrated that inoculated treatments enhanced biological yield (up to 14%) significantly over non-Inoculated treatments (**Table 5**) same results were also found by [24]. Significant variation among the inoculums for biological yield may be due to variation in the adaptability of inoculum to prevailing soil and climatic condition as

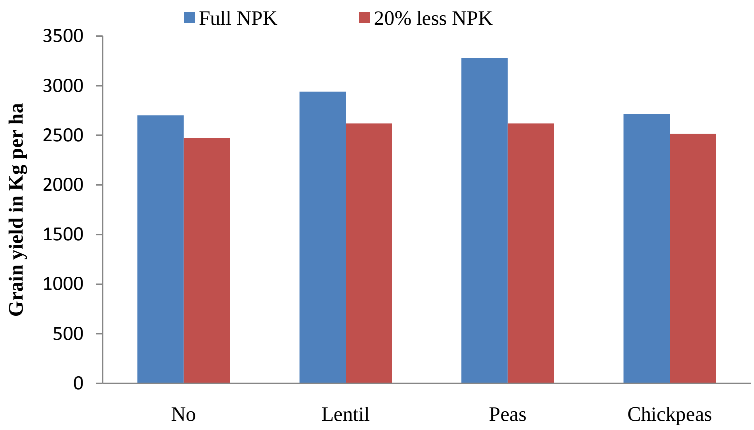

Figure 3. Grain yield as influenced by interaction of inoculum and NPK.

reported by [22]. Treatments with recommended NPK showed significantly higher (up to 8%) biological yield of 8433 kg·ha^{-1} than that of 7820 kg·ha^{-1} of 20% less 0f recommended NPK, this may be due to the inadequate availability of NPK at 20% less NPK. 1st order interaction of NPK and inoculum (NPK x inoculums) for biological yield indicated that non-inoculated treatment with full NPK produced significantly lower biological yield than chick pea, peas and Lentil Inoculum with 20% less of recommended NPK. These findings suggested that inoculation of wheat seed with Peas and Lentil Inoculums can contribute 20% NPK and thus may reduce the use of mineral NPK fertilizer up to 20% for wheat crop under prevailing soil conditions. These findings suggested that inoculation of rhizobia along with recommended NPK is more effective than inoculation with 20% less of recommended NPK.

4. Conclusion

It was concluded that inoculation of wheat with rhizobia enhanced wheat yield component at both recommended and 20% less of recommended NPK. Furthermore, it was also demonstrated by results that 20% less of recommended NPK along with peas and lentil inoculation had similar or better role than full recommended NPK alone in growth and yield of wheat crop.

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