

## GROWTH PROMOTION OF TWO WHEAT CULTIVARS BY PLANT GROWTH-PROMOTING RHIZOBACTERIA

MUHAMMAD JAVED AND MUHAMMAD ARSHAD\*

*Directorate of Land Reclamation,  
Canal Bank, Moghalpura, Lahore, Pakistan*

### Abstract

Based upon their ability to produce auxins (indole-3-acetic-acid equivalents), 11 isolates of rhizobacteria were selected to evaluate their growth promoting activity. Seeds of 2 wheat cvs. Inqlab and LU 26S inoculated with these isolates were sown in the field under optimum fertilization (NPK @ 150-75-50 kg ha<sup>-1</sup>). Inoculation with rhizobacteria showed an increase in grain yields of cv. Inqlab and LU 26S by upto 15.3% and 18.5%, respectively, as compared to uninoculated control. Inoculation with rhizobacteria also significantly increased the number of tillers, straw weight and 1000-grain weight in both cultivars. Plant height was increased only in case of wheat cv. LU 26S.

### Introduction

Free living saprophytic rhizosphere bacteria have been investigated for their effects on growth and yield of crops throughout the 20th century. The term 'plant growth-promoting rhizobacteria (PGPR)' was coined for the subset of total rhizosphere bacteria colonizing plant roots upon inoculation which have positive effects on plant growth (Kloepper & Schroth, 1978). However, Frankenberger & Arshad (1995) are of the view that all those rhizobacteria which promote plant growth upon inoculation through any mechanism of action could be grouped under PGPR. Research on the use of PGPR to promote plant growth has increased dramatically over the last few years in the world. Chen *et al.*, (1994) reported upto 15% increase in yield of wheat due to inoculation with PGPR. A composite analysis of their results showed that when PGPR were used in geographical area outside the isolated source, efficacy was variable with *Pseudomonas* species. Significant increases in yields of wheat due to inoculation with PGPR had been reported (Chen *et al.*, 1994; Xia *et al.*, 1990; deFreitas & Germida, 1990; 1992; Javed, 1995; Javed *et al.*, 1996).

During the last few years the heavy use of chemical fertilizers had shown little impact on improvement in crop yields. A tangible line of approach to solve this problem may be the introduction of beneficial soil microbes in agriculture for seed and soil inoculation. Keeping in view the potential of PGPR for growth promotion, experiments were carried out to examine the growth enhancing ability of various rhizobacteria under soil and climatic conditions of Faisalabad, Pakistan.

### Materials and Methods

Field trials were carried out in the Department of Soil Science, University of  
\*Department of Soil Science, University Agriculture, Faisalabad, Pakistan.

**Table 1. Effect of inoculation with rhizobacteria on grain yield, straw, 1000-grain weights, tillers and plant height of wheat cv. Inqlab.**

Isolates	grain yield (g m <sup>-2</sup> )	%incr- ease	straw weight (g m <sup>-2</sup> )	%incr- ease	1000-gr- ain weight (g)	%incr- ease	tillers m <sup>-2</sup>	%incr- ease	plant height (cm)	%incr- ease
J0	367.6 de	-	1176 c	-	33.52 c	-	581 cd	-	122.6 <sup>NS</sup>	-
J1	392.8 b-e	6.8	1323 ab	12.5	37.02 ab	10.4	600 c	3.3	124.0	1.1
J2	399.6 abc	8.7	1348 ab	14.6	37.25 ab	11.1	650 ab	11.9	124.0	1.1
J3	377.6 cde	2.7	1293 ab	9.9	34.30 c	2.3	642 b	10.5	123.5	0.7
J7	424.0 a	15.3	1378 a	17.2	38.43 a	14.7	686 a	18.1	123.6	0.8
J11	416.6 ab	13.3	1313 ab	11.6	38.18 a	13.9	660 ab	13.6	122.4	-0.2
J12	398.6 a-d	8.4	1336 ab	13.6	37.03 ab	10.5	655 ab	12.7	123.8	1.0
J17	375.7 cde	2.2	1264 bc	7.5	34.18 c	2.0	558 d	-4.0	123.5	0.7
J22	363.0 e	-1.2	1251 bc	6.4	34.14 c	1.9	596 c	2.6	124.2	1.3
J24	391.9 b-e	6.6	1320 ab	12.2	36.14 b	7.8	656 ab	12.9	122.8	0.2
J26	368.8 cde	0.3	1296 ab	10.2	33.59 c	0.2	649 ab	11.7	124.9	1.9
J30	410.2 ab	11.6	1326 ab	12.8	38.06 a	13.5	667 ab	14.8	123.5	0.7
LSD at P=0.05	28.22		100.5		1.740		34.13			

(Average of 5 replicates)

Means sharing the same letter(s) don't differ significantly at P=0.05

J0= no inoculation

NS= Non-significant

Agriculture, Faisalabad, Pakistan during the years 1992-94. Thirty eight isolates of rhizobacteria were isolated from the maize rhizosphere soil by using the glucose peptone medium. These isolates were screened on the basis of auxin (IAA-equivalents) production which was determined by inoculating 25 mL broth culture of each isolate and incubated at  $28 \pm 1^\circ\text{C}$  for 48 hours. The cultures were filtered and auxin production was measured on spectronic 20 at 535 nm (Sarwar *et al.* 1992). Eleven isolates were selected to study their effects on growth and yield of wheat. Seeds of two wheat cvs., Inqlab and LU 26S were inoculated with selected isolates. Seeds (1 kg) of each cultivar were treated with peat based preparation (100 mL broth culture having  $10^7$ - $10^8$  cfu mL<sup>-1</sup> +50 g sterilized peat +10% sugar solution). Controls (J0) were treated with slurry without inoculum. Inoculated and uninoculated seeds after drying in the shade were sown in the field.

Seeds of both the cvs. were sown on sandy clay loam soil having pH 7.8; ECe 1.9 dS m<sup>-1</sup>; SAR 4.29 (mmol L<sup>-1</sup>)<sup>1/2</sup>; OM 0.83% and CEC 6.62 cmol(+) kg<sup>-1</sup> soil. In both the trials treatments were replicated 5 times using randomized complete block design with net plot size 2.13x5.49 m<sup>2</sup>. NPK @ 150-75-50 kg ha<sup>-1</sup> were applied as urea, single super phosphate and potassium sulphate, respectively. Seeds of two cultivars were sown in lines by single row cotton seed drill keeping row to row distance of 30 cm. At maturity crops were harvested from one m<sup>2</sup> from each plot centre and data regarding plant height, number of tillers m<sup>-2</sup>, straw weight, grain yield and 1000-grain weight were recorded.

## Results

a) Cv. Inqlab: Inoculation of wheat cv. Inqlab with some of the isolates significantly affected all the parameters studied except plant height (Table 1). Isolate J7 showed maximum increase in grain yield (15.3% higher than control) and it was statistically at par with isolates J30, J12, J11 and J2, which respectively produced 11.6, 8.4, 13.3 and 8.7% higher yields over control. Straw weight was significantly increased over control with all the isolates except J17 and J22. Maximum increase in straw weight was observed with isolate J7 which was statistically at par with isolates J30, J26, J24, J12, J11, J3, J2 and J1. The increase in straw weight in response to inoculation with these isolates ranged from 9.9 to 17.2% over the control. Inoculation of wheat with all the selected isolates significantly increased 1000-grain weight over control except isolates J26, J22, J17 and J3. Maximum increase in 1000-grain weight was observed with isolate J7 (14.7% higher than control) and it differed non-significantly with isolates J30, J12, J11, J2 and J1, which respectively yielded 13.5, 10.5, 13.9, 11.1 and 10.4%, more 1000-grain weights as compared to control.

Plant height was not significantly affected due to inoculation with rhizobacteria, but number of tillers m<sup>-2</sup> significantly increased. Maximum number of tillers were recorded with isolate J7 (18.1% higher than control) and these were statistically at par with tillers produced by isolates J30, J26, J24, J12, J11 and J2.

b) Cv. LU 26S: Inoculation of wheat cv. LU 26S with different isolates significantly affected the grain yield, straw weight, 1000-grain weight, tillers m<sup>-2</sup> and plant height (Table 2). Maximum grain yield was recorded with isolate J2 (18.5% higher than

Table 2. Effect of inoculation with rhizobacteria on grain yield, straw, 1000-grain weights, tillers and plant height of wheat cv. LU 26S.

Isolates	grain yield (g m <sup>-2</sup> )	%incr-ease	straw weight (g m <sup>-2</sup> )	%incr-ease	1000-grain weight (g)	%incr-ease	tillers m <sup>-2</sup>	%incr-ease	plant height (cm)	%incr-ease
J0	527.1 cde	-	897.2 c	-	50.03 h	-	427 ef	-	111.5 c -	-
J1	574.8 a-d	9.0	992.6 b	10.6	51.15 fg	2.2	439 def	2.8	119.6 ab	7.3
J2	624.5 a	18.5	1053.3 a	17.4	54.64 a	9.2	504 a	18.0	119.0 ab	6.7
J3	526.5 cde	-0.1	998.2 b	11.2	50.52 gh	1.0	458 cde	7.2	118.4 ab	6.2
J7	592.2 ab	12.4	1029.7 ab	14.8	53.48 b	6.9	498 ab	16.6	116.9 ab	4.8
J11	575.7 a-d	9.2	1006.6 ab	12.2	52.20 cde	4.3	481 abc	12.6	117.8 ab	5.6
J12	548.7 bcd	4.1	992.3 b	10.6	51.95 c-f	3.8	468 a-d	9.6	121.2 a	8.7
J17	530.4 cde	0.6	986.5 b	9.9	51.50 ef	2.9	463 b-e	8.4	118.4 ab	6.2
J22	482.4 e	-8.5	903.2 c	0.7	49.94 h	-0.2	415 f	-2.8	115.6 bc	3.7
J24	588.0 ab	11.6	1010.4 ab	12.6	52.51 cd	5.0	483 abc	13.1	117.0 ab	4.9
J26	548.8 bcd	4.1	993.8 b	10.8	51.54 def	3.0	454 cde	6.3	117.8 ab	5.6
J30	582.0 abc	10.4	1009.2 ab	12.5	52.65 bc	5.2	486 abc	13.8	118.0 ab	5.8
LSD at P=0.05	49.41		47.14		0.905		35.66		4.910	

Means sharing the same letter(s) don't differ significantly at P=0.05

J0= no inoculation

control) and it differed significantly with control and all other isolates except J30, J24, J11, J7 and J1. Straw weight was significantly increased due to inoculation with all the isolates studied, except isolate J22. Maximum straw weight was recorded with isolate J2 (17.4% higher than control) and it was statistically at par with isolates J7, J11, J24, and J30, which respectively produced 14.8, 12.2, 12.6 and 12.5%, higher weights compared to control. Inoculation with different isolates also significantly increased 1000-grain weight over the control except isolates J22 and J3. Maximum increase in 1000-grain weight was recorded with isolate J2, which increased it by 9.2% over control and this isolate differed significantly from all other isolates. Isolates J7 and J30 increased the 1000-grain weights by 6.9 and 5.2%, respectively, over control and these differed significantly from control and non-significantly from each other.

Plant height measured at maturity and tillers  $m^{-2}$  were significantly increased over control due to inoculation with some of the rhizobacterial isolates. Maximum plant height was observed with isolate J12 (8.7% higher than control) and it was statistically equal to isolates J30, J26, J24, J17, J11, J7, J3, J2 and J1. Number of tillers  $m^{-2}$  of wheat were increased significantly over control with all the isolates studied except isolate J22. Maximum number of tillers were produced with isolate J2 (18.0% higher than control) and these were statistically equal to the tillers produced by isolates J30, J24, J12, J11 and J7.

## Discussion

Selected rhizobacteria, isolated from the maize rhizosphere and applied as seed coating produced significant increase in yield of two wheat cvs. Inqlab and LU 26S by upto 15.3% and 18.5%, respectively. Chen *et al.*, (1994) reported increase in yield of wheat upto 15.0%. Our results are also in accordance with the findings of Xia *et al.*, (1990) and Vancura (1989) who reported mean increases in yield of wheat by upto 14.7 and 15.0%, respectively due to inoculation with PGPR, in different field trials. In the present study, the response of cultivars to rhizobacterial inoculation were different. This might be due to genetic make up of the cultivars. These results are in accordance with the findings of deFreitas & Germida (1990), Grayston *et al.*, (1991) and Chanway *et al.*, (1988) who observed the specificity of PGPR for certain soils and cultivars.

Inoculation with rhizobacteria also significantly increased the number of tillers, straw weight and 1000-grain weight in both cultivars but plant height only in case of LU 26S. These results are in conformity with the findings of different workers who observed that inoculation of wheat with different PGPR enhanced plant height (deFreitas & Germida, 1990), tiller formation (Xia *et al.*, 1990; Vancura, 1989) and plant dry weight (Javed, 1995; deFreitas & Germida, 1992).

The isolates used in this study were tentatively identified as Pseudomonads on the basis of physiological and morphological characteristics. The ability of the *Pseudomonas* spp., to rapidly colonize the rhizosphere of most plants makes them an attractive taxonomic group of soil bacteria for bacterization studies. Kloepper (1994) reported that the precise mode of action for commercially used PGPR inoculants are yet not known for all strains. The production of siderophores, antibiotics, several extracellular metabolites and induced systemic resistance are the mechanisms of indirect growth promotion

in addition to direct growth promotion as evidenced by increased root hair development. However, Frankenberger & Arshad (1995) are of the view that all those rhizobacteria which promote plant growth upon inoculation through any mechanism of action could be grouped under PGPR.

The impact of agricultural chemicals on soil, water, plant and ultimately on human environment contain a global dimension and the situation would worsen as the use of fertilizer multiplies further over the years (Sial *et al.*, 1993). Under such circumstances the use of such techniques should be encouraged for environmentally safe agriculture.

## References

- Chanway, C.P., L.M. Nelson and F.B. Holl. 1988. Cultivar-specific growth promotion of spring wheat (*Triticum aestivum* L.) by coexistent *Bacillus* species. *Can. J. Microbiol.*, 34: 925-929.
- Chen, Y., R. Mei, S. Lu, L. Liu and J.W. Kloepper. 1994. The use of yield increasing bacteria as plant growth-promoting rhizobacteria in Chinese agriculture. In: *Management of Soil-Borne Diseases*. (Eds.) V.K. Gupta and R. Utkhede. M/S Narosa Publishing House, New Delhi, India.
- deFreitas, J.R. and J.J. Germida. 1990. Plant growth promoting rhizobacteria for winter wheat. *Can. J. Microbiol.*, 36: 265-272.
- deFreitas, J.R. and J.J. Germida. 1992. Growth promotion of winter wheat by fluorescent pseudomonads under growth chamber conditions. *Soil Biol. Biochem.*, 24: 1127-1135.
- Frankenberger, W.T. Jr. and M. Arshad (Eds.). 1995. *Phytohormones in Soils: Microbial Production and Functions*. Marcel Dekker Inc., New York NY.
- Grayston, S.J., J.H. Stephens and L.M. Nelson. 1991. Field and greenhouse studies on growth promotion of spring wheat inoculated with co-existent rhizobacteria. p.11-16. In: *Plant Growth Promoting Rhizobacteria-Progress and Prospects* (Eds.) C. Keel, B. Koller and G. Défago. IOBC, Switzerland.
- Javed, M. 1995. *Potential of plant growth-promoting rhizobacteria for enhancing yields of agricultural crops*. Ph.D. Thesis, Dept. of soil Sci., Univ. of Agri. Faisalabad, Pakistan.
- Javed, M., M. Arshad and A. Hussain. 1996. Improving growth and yield of wheat with plant growth promoting rhizobacteria. *Pak. J. Soil Sci.*, 12: 95-100.
- Kloepper, J.W. 1994. Plant growth-promoting rhizobacteria (other systems). p.135-166. In: *Azospirillum/Plant Associations*. (Ed.) Y. Okon, Boca Raton Ann Arbor London, Tokyo.
- Kloepper, J.W. and M.N. Schroth. 1978. Plant growth promoting rhizobacteria on radishes. p.879-882. In: *Proc. 4th Int. Conf. Plant Pathogenic Bacteria*, Vol. 2, Station de Pathologie, INRA. Angers (Ed.). Gibert-Clary, Tours, France.
- Sial, J.K., S. Mahmood, N. Ahmad and M.S. Sabir. 1993. Nitrate nitrogen management for ground water quality. p. 48-54. In: *Int. Symp. on Environmental Assessment and Management of Irrigation and Drainage Projects*. April, 1993, Lahore.
- Sarwar, M., M. Arshad, D.A. Martens and W.T. Frankenberger Jr. 1992. Tryptophan-dependent biosynthesis of auxins in soil. *Plant Soil*, 147:207-215.
- Vancura, V. 1989. Inoculation of plants with *Pseudomonas putida*. p.185-190. In: *Interrelationships between Microorganisms and Plants in Soil*. (Developments in Soil Sci. 18). (Eds.) V. Vancura and F.Kunc. Elsevier Amsterdam, Netherland.
- Xia, L., X. Ding, S. Xiao, S. Han and R. Mei. 1990. Mechanism of PGPR.III. Influence of PGPR on physiology, resistance, quality and yield of wheat. *Agricultural Sci. in Hunan*, 108:26-29.