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Effect of Plant Growth Promoting Rhizobacteria (PGPR) on Reduction Nitrogen Fertilizer Application in Rapeseed (*Brassica napus* L.)

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Abstract: In order to study the effect of *Plant Growth Promoting Rhizobacteria* (PGPR) on yield and yield components of rapeseed, a factorial experiment was conducted based on randomized completed block design with three replications in Ilam, Iran in 2011-2012 cropping season. The factors consisted of three levels of nitrogen fertilizer (100, 150 and 200 kg ha^{-1}) and bio-fertilizer (non-inoculation, *Azotobacter Chroococcum* and *Pseudomona Putid*). The results showed that nitrogen rates had significant effects on yield and yield components. Significant increase was observed in all characters with applying bio-fertilizers and increasing nitrogen from 100 to 200 kg ha^{-1}. The latter two treatments were in same statistical class in most characters. Applying *Azotobacter* and *Pseudomona* increased yield and yield components by 15.8 and 13.7%, respectively compared with control treatment. There were significant interaction between nitrogen levels and bio-fertilizers regarding yield, yield components and seed oil and protein content. The highest amount of yield obtained from applying 200 kg ha^{-1} with *Azotobacter*. Also, the highest seed oil and protein content was obtained by applying 200 kg ha^{-1} nitrogen, *Pseudomona*. There were no significant differences between 200 and 150 kg ha^{-1}. According to results of present study, it can be concluded that farmers can obtain the same rapeseed yield if they apply 150 kg.ha^{-1} of nitrogen with bio-fertilizers. In this way, decreasing nitrogen fertilizer help to reduce environment pollution and develop sustainable agriculture.

Key words: Rapeseed · Nitrogen fertilizer · Seed yield · PGPR

INRODUCTION

Brassica napus L. is one of the main agricultural plants cultivated across the world to extract oil and in terms of production it stands at the third rank after soybeans and palm oil. *Brassica napus* L. is a new product with special traits which can exploit seasonal rainfall like winter Plants and can be an appropriate alternative for wheat and oats [1]. Chemical fertilizers have several negative impacts on environment and sustainable agriculture. Therefore, bio-fertilizers are recommended in these conditions and growth prompting bacteria uses as a replacement of chemical fertilizers [2]. Plant Growth Promoting Rhizobacteria (PGPR) are a group of bacteria that actively colonize plant roots and increase plant growth and yield. The mechanisms by which PGPRs promote plant growth are not fully understood, but are thought to include: - the ability to produce phytohormons, - asymbiotic N2 fixation, against phytopathogenic microorganisms by production of siderophores, the synthesis of antibiotics, enzymes and/or fungicidal compounds and also - solubilisation of mineral phosphates and other nutrients [3]. Growth prompting bacteria including *Azotobacter*, *Azospirillum* and *Pseudomonas* [4, 5, 6].

Tilak [7] reported positive effects of double-inoculation of *Azotobacter* and *Azospirillum* on dry matter of maize and sorghum. Rai and Caur [8] studied *Azotobacter* and *Azospirillum* and double-inoculation and alone inoculation effects on wheat growth and yield.

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Table 1: Soil physical and chemical properties of experimental area

Soil texture	Available P (mg kg^{-1})	Available K (mg kg^{-1})	Total N (%)	Organic Carbon (%)	E.C(dS/m)	pH
Silty loam	4.2	312	0.09	1.12	0.60	7.4

N2-fixing may be important for plant nutrition by increasing N uptake by the plants and playing a significant role as plant growth promoting rhizobacteria (PGPR) in the biofertilization of crops. Plant growth-promoting rhizohacteria (PGPR) are able exit a beneficial upon plant growth. Nitrogen fixation and P solubilization [9] production of antibiotic [4] and increased rood dry weight are the principal mechanism for the PGPR. A number of different bacteria promote plant growth, including *Azotobacter sp*., *Azospirillum sp*., *Pseudomones sp*., *Bacillus sp*. *Acetobacter sp* [5]. Plant growth promoting rhizobacteria (PGPR) are a group of bacteria that actively colonize plant roots and increase plant growth and yield. The mechanisms by which PGPRs promote plant growth are not fully understood, but are thought to include: - the ability to produce phytohormons, - asymbiotic N_2 fixation, -against phytopathogenic microorganisms by production of siderophores, the synthesis of antibiotics, enzymes and/or fungicidal compounds and also - solubilisation of mineral phosphates and other nutrients [3]. In Behl *et al.* [10] experiment integrated use of *Azotobacter* and micorhiyza caused to increasing seed yield, seed number, 1000-seed weight and bio- yield of wheat. Zahir *et al.* [11] reported 19.8 % increase in seed yield of maize due to dual inoculating seed with *Azotobacter* and *Pseudomona*. Tilak *et al.* [7] reported improving seed yield of maize due to duel inoculation of *Azotobacter* and *Azospirillum*. Boddy and Dbereinezer [12] also reported that yield and nitrogen content increased due to inoculation of wheat with *Azospirillum*. Inoculation with *Azotobacter* sp., *Enterobacter* sp. or *Klebsiella* sp. increased number of root hairs, tillering ratio, dry matter concentration, N-uptake or yields of wheat [13]. Other reasons are related to producing amino acids, carbohydrates, organic acids and growth simulating materials [14]. With due attention to irregular chemical fertilizers consumption and to rapeseed importance as new oily plant and lack of supported and comprehensive information about growth reaction of this plant to bio-fertilizer, this study, thus, conducted in order to evaluate bio-fertilizer on yield and yield components of rapeseed in nitrogen levels.

MATERIALS AND METHODS

In order to investigate and study bacterial bio-fertilizer application on yield and yield components of rapeseed (Hayola 401), a factorial experiment was conducted based on randomized completed block design with three replications in Ilam, Iran in 2011-2012 cropping season. Soil physical and chemical properties of experimental area are shown in Table 1. The factors consisted of three levels of fertilizer (100, 150 and 200 kg ha^{-1}) and bio-fertilizer (non-inoculation, *Azotobacter Chroococcum* and *Pseudomona Putid*).

Experiment plots were seeded with *Hayola 401* cultivar with 25 cm row to row distance and 10 cm between plants. Rapeseed was planted manually in October 2011. Seeds were sown 4 cm deep. Weeds were removed by hand.

Plots consisted of six lines with 4 meter length, 30 cm row spacing and 10 cm plant spacing. Seeds were moistured with 2% sugar water and inoculated with 7 gram inoculation including 10^7 alive and active bacteria before planting. After planting, irrigation was applied as required during the growing season. The rapeseed was harvested in April 2012. In order to measuring agronomic traits (i.e. pods s per plant, seeds per pod and 1000-seed weight) 10 plants were randomly selected in each plot. In order to measure seed yield, all lines of each plot except marginal ones and 50 cm from beginning and end of the lines were harvested at maturity stage and appropriate seed moisture. Seed oil and protein content were measured using Nuclear Magnetic Resonance Spectrophotometer (NMR) and Micro Kejeldahl digestion, respectively. Statically analysis was conducted using MSTAT-c software. Mean comparison was also conducted with Duncan's Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

Analyzing seed yield variance showed that there was a significant difference between nitrogen fertilizer levels (Table 2). Seed yield was increased with applying nitrogen fertilizer (Table 3). However, there was no significant difference between 150 and 200 kg ha^{-1}. Bio-fertilizers had significant effect on seed yield. In due attention to Table results, comparing mean bio-fertilizers application caused to increasing seed yield in compared with control treatment. *Azotobacter* and *Pseudomona* improved seed yield by 15.8 and 13.7%, respectively, in compared with non- inoculation treatment. These results are in agreement with those obtained by Singh *et al.* [14] who believed to maximize production of different wheat cultivars which

Table 2: Analysis of variance (mean of squares) for traits under study in plant growth promoting rhizobacteria (PGPR) under nitrogen fertilizer application in rapeseed

		MS					
S.O.V	d.f	Seed yield	Pods per plant	Seeds per pod	1000-seed weight	Oil content	Protein content
Replication	2	147579.3	85.1	31.1	59.2	32.1	7.4
Nitrogen fertilizer (N)	2	1486514.8**	210.1**	412.1**	215.3**	36.1**	13.5**
Inoculation (I)	2	524685.5**	109.2**	512.2**	240.9**	19.8**	6.5**
N×I	4	76164.9**	18.1**	43.7**	65.1**	9.1**	3.5**
Error	16	14271	3.3	3.2	7.9	1.9	1.1
CV (%)		18.2	13.4	10.6	9.1	8.4	6.5

*: Significant at 0.05 level, **: Significant at 0.01 level

Table 3: Main effect of nitrogen fertilizer and Plant growth promoting rhizobacteria on seed yield, number of pods per plant, number of seed per pods, 1000-seed weight oil content and protein

Treatments	Seed yield (kg ha^{-1})	Pods per plant	Seeds per pods	1000-seed weight	Oil content (%)	Protein content (%)
Nitrogen fertilizer (kgha^{-1})						
100	2850.1b	75.1c	16.9c	3.3b	42.5b	20.1c
150	3232.3a	84.2ab	19.7a	3.5a	44.1a	21.3ab
200	3332.1a	89.3a	20.1a	3.65a	44.3a	21.7a
Inoculation						
No- inoculation	2820.8c	77.5c	17.2c	3.1c	42.7c	19.6c
Azotobacter	3351.8a	87.5a	19.5b	3.7a	43.3b	21.5a
Pseudomona	3271.6b	83.1b	20.05a	3.5b	43.2a	21.3b

Means, in each column, followed by similar letter are not significantly different at the 5% probability level- using Duncans Multiple Range Test

inoculated with *Azotobacter* under normal conditions. Results obtained from this study showed that with using bio-fertilizer inoculation methods, not only seed yield can be improved but also consumption of nitrogen chemical fertilizer reduces remarkably. Seed yield improving factors causes a tinted rated treatment include increasing plant accessibility to nutrients using dual use chemical fertilizers and more their absorption by plant and as a result of improving growth and photosynthesis by increasing leave area. Nanda *et al.* [15] reported that grain maize inoculation with *Azospirillum* and *Azotobacter* significantly increased forage yield of the plant. Similar results were obtained by Lucangeli and Bottini [16] who reported that secreting growth regulating and prompting in attars such as uxin, gibberllin by *Azospirillum* and secreting uxin, gibberllin and cytokinin by *Azotobacter* and assisting these bacteria with maize rhizome are most important interacting to improving growth hand grain yield of maize. The positive effects of PGPRs on the yield and growth of crops such as wheat, maize, soybean and sugar beet were explained by N_2 fixation ability, P solubilizing capacity and phytohormons production [17, 13]. Interaction effect between chemical fertilizer and bio-fertilizer was affect on seed yield, but the highest and lowest seed yield was obtained from 200 kgha^{-1} and inoculation with *Azotobacter* and 100 kgha^{-1} and non-inoculation treatment, respectively (Fig. 1).

Pods per plant was significantly influenced by nitrogen fertilizer and bio-fertilizer treatment as well (Table 2). Pods per plant was increased with increasing nitrogen fertilizer. This can be attributed to improving water absorption and plants nourishing due to nitrogen. On the other hand, there were no significant differences between 150 kgha^{-1} and 200 kgha^{-1}. The lowest number of pods per plant was observed in non-inoculation treatment and bacteria treatments caused to increasing pods per plant. Effect of bio-fertilizers on pods per plant and flowering levels were positive in this experiment. In other word, using nitrogen fertilizer at appropriate levels provide better nutrient uptake and plant photosynthesis through improving bio-fertilizers activity which results in better flowering and poding. Also, positive effect of using bio-fertilizer can be attributed to increase water and nutrient uptake due to development and expansion of roots and also to biological nitrogen fixation by bio-fertilizers. Interaction effect between chemical fertilizer and bio-fertilizer was affect on pods per plant. The highest and lowest number of pods per plant was obtained with applying 150 or 200 kg ha^{-1} by *Azotobacter* treatments and 100 kg ha^{-1} and non-inoculation treatment, respectively (Fig. 2). Lower nitrogen levels accompanied by bacteria and higher nitrogen levels accompanied by coexistent bacteria were set at same group. This indicates that these bacteria are

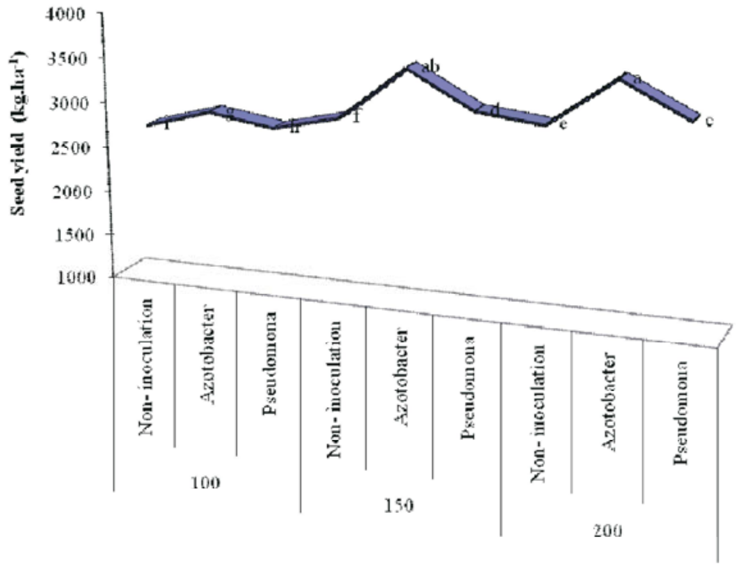

Fig. 1: Response of seed yield to nitrogen chemical fertilizer and bio-fertilizer application

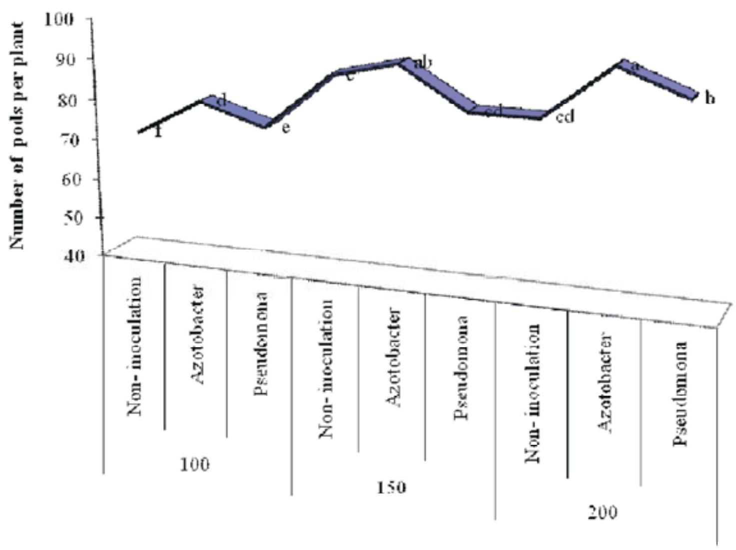

Fig. 2: Response of number of pods per plant to nitrogen chemical feratilizer and bio-fertilizer application

active in rhizospher and can minimize the need of applying nitrogen fertilizer. This is probably resulted from synthesize of bacteria simulating plant growth promoting rhizobacteria and also nitrogen fixation by these bacteria. This current investigation confirms the earlier works. It revealed that under conditions, seed treatment with PGPR improved seed germination, seedling vigor, seedling emergence and seedling stand over the control. Corresponding enhancement of seed germination parameters by PGPR has been reported in other crop plants [18]. Naseri and Mirzaei [19] also indicated positive effect of bio-fertilizers on number of head per plant in safflower.

Seeds number per pod was influenced by nitrogen fertilizer and bio-fertilizer treatments and their interaction as well (Table 2). This trait was increased by increasing nitrogen level. The lowest seeds per pod belonged to non-inoculation treatment. Bio-fertilizers caused to increasing seeds per pod; both kinds of bio-fertilizers had same effects on this trait. Also interaction of this traits indicated, seeds per pod increase due to multiplying nitrogen fertilizer, both bio-fertilizers (*Azotobacter* and *Pseudomona*) have significant and positive effects on this trait. The highest seeds per pod produced in 200 kgha^{-1} with *Azotobacter* (Fig. 3). In general *Azotobacter* followed by nitrogen fertilizer can increase seed yield and yields

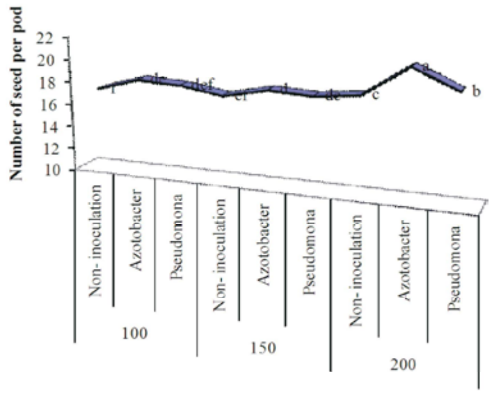

Fig. 3: Response of number of seed per pod to nitrogen chemical fertilizer and bio-fertilizer application

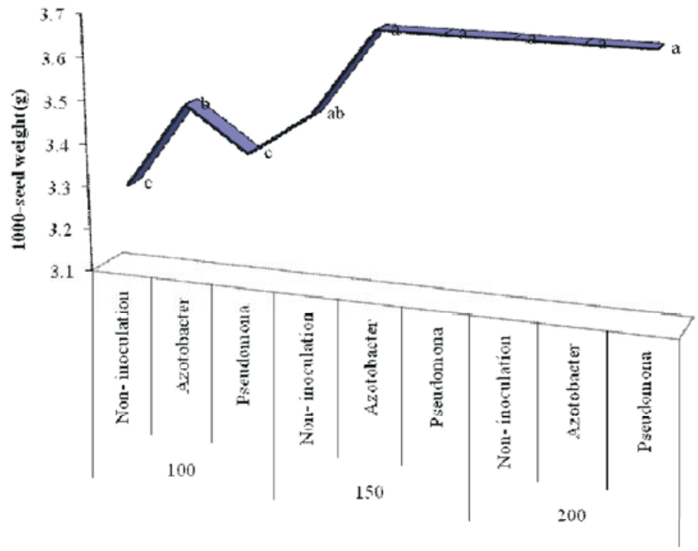

Fig. 4: Response of 1000-seed weight to nitrogen chemical fertilizer and bio-fertilizer application

components by positive influence on macro elements absorption such as N, P and K [20], micro elements such as Zn and Fe [21], improving water distributing in plant, developing nitrate reductase activity and finally producing plant hormones which have an important role in plant growth. This results is in agreement with the findings of Narula *et al.* [20] who reported that producing indole acetic acid followed by cytokinin by *Azotobacter* cause to increasing preserved matters by growing rhizomes and increasing leaf and root weight and as a result cause to increasing reproductive organs such as grain number. Fulchieri and Frioni [22] reported that increasing grain due to inoculating seed wits *Azospirillum*. Asghar *et al*. [23] indicated that *Azospirillum*, *Pseudomonas* and *Azotobacter* strains could affect seed germination and seedling growth.

1000-seed weight showed an increase by multiplying nitrogen fertilizer amount, but between 150 and 200 kgha^{-1} wasn't observe a significant difference. 1000-seed weight also increased due to inoculating seed with studied bacteria in compared with non- inoculation. Bio-fertilizers improved photosynthesis maybe by increasing water and nutrients absorption leading to produce more assimilate and improve plant growth and thus, 1000-seed weight increased in comparison with non-inoculation treatment. This trait interaction showed that lowest 1000-seed weight produced at bio-fertilizer non-inoculation and non-utilizing nitrogen fertilizer treatments. As showed in Fig. 4, the highest 1000-seed weight was observed at 150 and 200 kgha^{-1} nitrogen fertilizer followed by *Azotobacter* and *Pseudomona*. Applying 150 kg N ha^{-1} provided better nourishment condition to activity and reproducing *Azotobacter* and *Pseudomona*, because these bacteria need this element to grow, develop and fix nitrogen. Bio-fertilizer treatments provided more suitable condition to improving bioactivities of soil and caused to increasing 1000-seed weight through absorbing nutrients by root in compared with control treatment. This result is

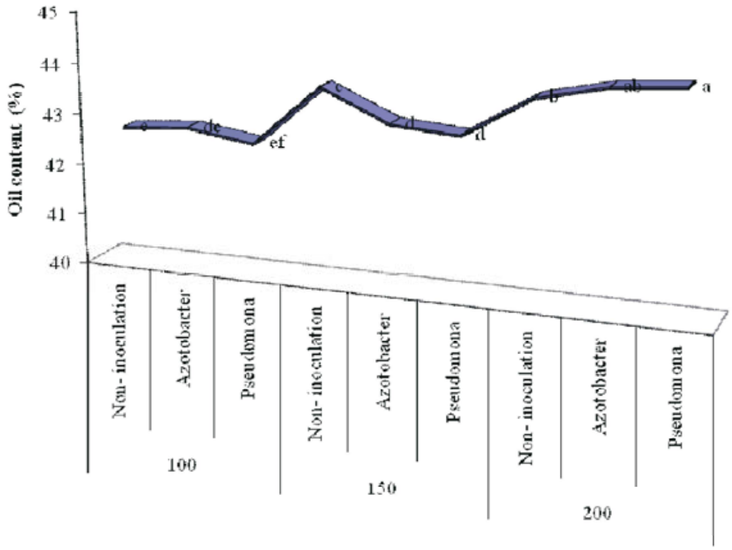

Fig. 5: Response of oil content to nitrogen chemical fertilizer and bio-fertilizer application

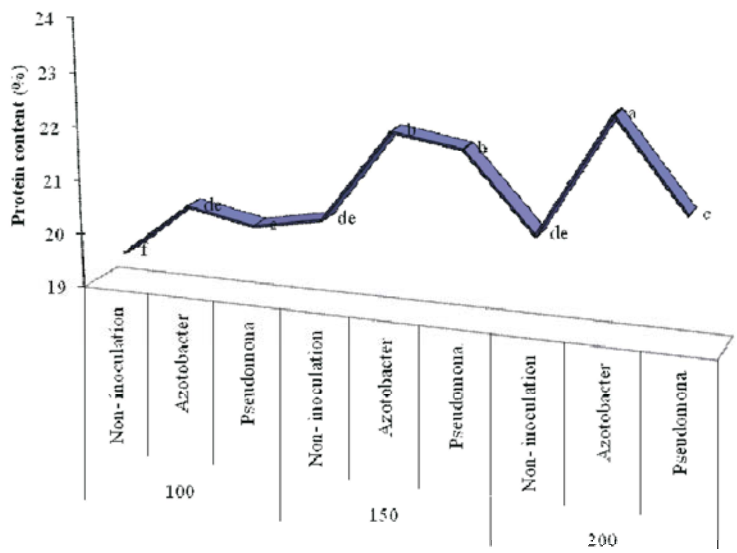

Fig. 6: Response of protein content nitrogen chemical fertilizer and bio-fertilizer application

in agreement with the findings of Yasari and Patwardhan [24] who indicated that application of *Azotobacter* and *Pseudomona* increased canola weight of 1000-seed. Meshram and Shende [25] suggested that PGPR increases root surface area and thus promotes intake of N, P, K, other nutrients and water and consequently above ground weight of plants.

Results obtained from analysis variance indicated that there are significant differences between nitrogen fertilizer, bio-fertilizer and their interaction effects on oil and protein content (Table 2). The lowest oil and protein content was observed in non-utilizing nitrogen fertilizer treatment. Oil and protein content increased with using nitrogen fertilizer, but statistical significant differences weren't observed between 150 and 200 kgha^{-1}. The highest oil and protein content belongs to bio-fertilizers followed by nitrogen fertilizer. Shehata and El-Khawas [26] reported a significant increase in oil content of sunflower with applying bio-fertilizer. Nitrogen fertilizer ×bio-fertilizer interaction indicated that increasing nitrogen and bio-fertilizers increases oil and protein content, as shown in Fig. 5 & 6. Interaction effect between chemical fertilizer and bio-fertilizer was affect on oil and protein content, the highest and lowest oil and protein content were obtained from 200 kgha^{-1} and inoculation with *Azotobacter* and 100 kgha^{-1} and non-inoculation treatment, respectively.

CONCUSION

In general, using bio-fertilizers and manage integrated nourishment quantitatively and qualitatively is one of the efficient ways to improve plants production and environment would have a better condition if chemical fertilizers consumption reduce. Recent studies indicated that using bio-fertilizers also improving soil physiological structure and also increase organic matters content and nitrogen available to coexistent plant. Of course, before it is recommended to massive production and widely application it is necessary to implement and replicate this experiment in different regions.

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REFERENCES

1. Naghizadeh, M. and B. Hasanzadeh, 2012. Effect of plant density on yield, yield components, oil and protein of canola cultivars in Hajiabad. Advances in Environmental Biology, 6(3): 1000-1005.
2. Wu, S.C., Z.H. Caob, Z.G. Lib, K.C. Cheunga and M.H. Wong. 2005. Effects of bio-fertilizer containing N-fixer, P and K solubilizers and AM fungi on maize growth: a greenhouse trial. Geoderma, 125: 155-166.
3. Gholami, A., S. Shahsavani and S. Nezarat, 2009. The Effect of Plant Growth Promoting Rhizobacteria (PGPR) on Germination, Seedling Growth and Yield of Maize. World Academy of Science, Engineering and Technology, 49: 19-24.
4. Zahir, A., Z.M. Arshad and W.F. Frankenberger, 2004. Plant growth promoting rhizobacteria. Advances in Agronomy, 81: 97-168.
5. Turan, M., N. Ataoglu and F. Sahin, 2006. Evaluation of the capacity of phosphate solubilizing bacteria and fungi on different forms of phosphorus in liquid culture. Sustainable Agricultural, 28: 99-108.
6. Banerjee, M., R.L. Yesmin and J.K. Vessey, 2006. Plant-growth promoting rhizobacteria as bio-fertilizers and biopesticides. pp: 137-181. In: Handbook of microbial bio-fertilizers'. Ed., Rai, M., K., Food Production Press, U.S.A.
7. Tilak, K.V.B., C.S. Singh, V.K. Roy and N.S. Rao, 1982. *Azospirillum brasilense* and *Azotobacter chroococcum* inoculum: effect on yield of maize and sorghum. Soil Bio and Biochem., 14: 417- 418.
8. Rai, S.N. and A.C. Caur, 1998. Characterization of *Azotobacter* Spp. and effect of *Azospirilum lipoferum* on the yield and N-uptake of wheat crop. Plant and Soil, 109: 131-134.
9. Zaidi, A. and S. Mohammad, 2006. Co-inoculation effects of phosphate solubilizing micro- organisms and *Glomus fasciculatum* on green grambradyrhizobium symbiosis. Agricultural Science, 30: 223-230.
10. Behl, R.K., H. Sharma, V. Kumar and K.P. Singh, 2003. Effect of duel inoculation of VA micorrhyza and *Azotobacter chroococcum* on above flag leaf characters in wheat. Aechives of Agronomy and soil Science, 49(1): 25-31.
11. Zahir, AZ., M. Arshad and A. Khalid. 1998. Improving maize yield by inoculation with plant growth promoting rhizobacteria. Soil Sci. J. Pak., 15: 7-11.
12. Boddey, R.M. and J. Dobereinezr. 1998. Nitrogen fixation associated with grasses and cereals. Plant and Soil, 102: 53-65.
13. Salantur, A., A. Ozturk, S. Akten, F. Sahin and F. Donmez, 2005. Effect of inoculation with non-indigenous and indigenous rhizobacteria of Erzurum (Turkey) origin on growth and yield of spring barley. Plant and Soil, 275: 147-156.
14. Singh, R., R.K. Behl, K.P. Singh, P. Jain and N. Narula, 2004. Performance and gene effects for wheat yield under inoculation of arbuscular mycorrhiza fungi and *Azotobacter chroococcum*. Haryana Agricultural University. Hisar, India. Plant Soil Environ., 50 (9): 409-415.
15. Nanda, S.S., K.C. Swain, S.C. Panda, A.K. Mohanty and M.A. Alim, 1995. Effect of nitrogen and biofertilizers in fodder rainfed upland conditions of Orisa. Current Agricultural Research, 8: 45-47.
16. Lucangeli, C. and R. Bottini, 1997. Effects of *Azospirillum* spp. On endogenous gibberellin content and growth of maize (*Zea mays* L.) treated with uniconazol Symbiosis, 23: 63-71.
17. Ozturk, A., O. Caglar and F. Sahin. 2003. Yield response of wheat and barley to inoculation of plant growth promoting rhizobacteria at various levels of nitrogen fertilization. J. Plant. Nutr. Soil Sci., 166: 262-266.

18. Naserirad, H., A. Soleymanifard and R. Naseri, 2011. Effect of integrated application of bio-fertilizer on grain yield, yield components and associated traits of maize cultivars. American-Eurasian J. Agric. and Environ. Sci., 10(2): 271-277.
19. Naseri, R. and A. Mirzaei. 2010. Response of yield and yield components of Safflower (*Carthamus tinctorius* L.) to seed inoculation with *Azotobacter* and *Azospirillum* and different nitrogen levels under dry land condition. American-Eurasian J. Agric. and Environ. Sci., 9(4): 445-449.
20. Narula, N., V. Kumar, R.K. Behl, A. Deubel, A. Gransee and W. Merbach, 2001. Effect of P-solubilizing *Azotobacter chroococcum* on N, P and K uptake in P- responsive wheat genotypes grown under greenhouse conditions. J. Plant Nutr. 163: 393-398.
21. Wani, S.P., S. Chandrapalaiah, M.A. Zambre and K.K. Lee, 1988. Association between nitrogen-fixing bacteria and pearl millet plants, responses mechanisms and resistance. Plant and Soil, 110: 284-302.
22. Fulchieri, M. and L. Frioni, 1994. *Azospirillum* inoculation on maize (*Zea mays* L.): Effect on yield in a field experiment in central Argentina. Soil Biology and Biochemistry, 26: 921-923.
23. Asghar, H.N., Z.A. Zahir, M. Arshad and A. Khaliq, 2002. 'Relationship between in vitro production of auxins by rhizobacteria and their growth promoting activities in *Brassica juncea*. L'. Bio. Fertil. Soil, 35: 231-237.
24. Yasari, E. and A.M. Patwardhan, 2007. Effects of *Aztobacter* and *Azospirillium* inoculations and chemical fertilizers on growth and productivity of Canola. Asian J. Plant Sci., 6(1): 77-82.
25. Meshram, S.U. and S.T. Shende, 1993. Total nitrogen uptake by maize with *Azotobacter* inoculation. Plant and Soil., 69: 275-280.
26. Shehata, M.M. and S.A. EL-Khawas, 2003. Effect of two bio-fertilizers on growth parameters, yield characters, nitrogenous components, nucleic acids content, minerals, oil content, protein profiles and DNA banding pattern of sunflower yield. Pakistan Journal of Biological Sciences, 6(14): 1257-1268.