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Effect of Seed Priming with Plant Growth Promoting Rhizobacteria (PGPR) on Agronomic Traits and Yield of Barley Cultivars

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Abstract: In order to evaluate the effects of seed priming with Plant Growth Promoting Rhizobacteria (PGPR) on grain yield and agronomic traits of barley cultivars, a factorial experiment based on randomized complete block design with three replications was conducted in 2009. Factors were: seed priming with Plant Growth Promoting Rhizobacteria in three levels containing, without priming (as control), priming with *Azotobacter*, *Azospirillium*, *Azotobacter* + *Azospirillium* three plus barley cultivars (Makoei, Bahman and CB-7910). The results showed that seed priming with Plant Growth Promoting Rhizobacteria affected plant height, spike length, number of spike per area, grains per spike, 1000-grain weight and grain yield, significantly. Maximum of these traits were obtained by the plots in which seeds were inoculated with *Azospirillium* bacteria. The highest grain yield (3063.4 $kg.ha^{-1}$) was obtained in cultivar of Makori. According to the results of this experiment, application of PGPR bacteria (especially Azospirillum) had an appropriate performance and could increase grain yield to an acceptable level, so it could be considered as a suitable substitute for chemical fertilizer in organic agricultural systems.

Key words: Barley · PGPR · Grain yield

INTRODUCTION

The use of biological fertilizers, especially plant growth promoting bacteria, are the most important strategy to increase production in sustained agricultural systems [1]. These bacteria are called plant growth promoting bacteria because of their great impact on crop growth [2]. Mechanisms that bacteria use to drive growth are not fully understood, but generally the production of growth promoting hormones, participation in biological survival of Nitrogen, controlling plant pathogens by producing antibiotics enzymes, mass production of growth promoting hormones specially different types of auxins, jibberllin and cytokinin, growth stop, or complete elimination of pathogens in soil can be noted. The use of the different breeds of bacteria which promote plant growth is very important, since the provision of adequate amounts of nutrients can provide access to high performance. Using these bacteria in biological fertilizers and agricultural bio-control has been proved in various field and *in vitro* experiments. *Azotobacter* and *Azospirillium* are of useful soil living bacteria to which crops reported to have different responses based on bacteria breed and climate of the region. In cases yield increase was about 12 to 39 percent [1]. In addition to stabilizing Nitrogen survival in the soil and soil phosphor solution, they affect plant growth by producing considerable amount of plant growth promoting bacteria specially auxins, jibberellin and cytokinin [3]. Since the solution power of barely root is less than other crops specially wheat, rye [2], it seems that growth promoting rhizobacteria (PGPR) affects growth rate by increasing the solubility of minerals in soil and absorption of nutrition by barely root [4]. It was reported that the increases in leaf appearance rate was due to use of bacteria *Azospirillum* and pseudomonas. Similar results have been reported by Ribaudo *et al*. [5]. Increasing of wet upper part of the plant and number of leaf in maize [6] and barely [7]. The extentions of leaf area and delay in leaf senescence in barely have been reported by different researchers [7, 8]. Shaukat *et al*. [9] stated that the races of *Azosprilium*, *Pseudomonas* and *Azotobacter* had significant effects on

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germination and seedling growth. Various experiments showed that inoculating plants with *Azospirillium* caused significant changes in different growth parameters such as increase in dry matter weight, nutrition absorption, tissue nitrogen, plant high, leaf size and root length in crops [10]. The bacteria increase germination, root length and weight of the aerial, accelerate the establishment of plants, root elongation, increase number of embryonic and lateral roots, leaf area expansion, delayed leaf senescence, chlorophyll and protein content and ultimately increase crop yield and quality of different plants [7]. Cakmakci *et al*.[11] concluded that inoculation of barely with PGPB increased mineral nitrogen concentration in soil and phosphorus and nitrogen in barely seed. The results of this experiment showed that the aerial plant weight increased by 28.8 to 45.2% due to bacteria inoculation depending on the type of bacteria. Kloepper and Beauchamp [12] reported that wheat yield increased between 30-43% after inoculation with bacteria. Cakmakci *et al.* [7] in separate studies stated that inoculation of plants with *Azospirillum* can cause significant changes in parameters as increase in total biomass and leaf area. significant increase in growth and yield by plant growth promoting bacteria inoculation have been shown in separate experiments by Asghar *et al*. [13] and Biswas *et al*. [14]. Kloepper *et al.* [12] stated that yield of plants such as rice, corn and sugar cane when inoculated with the bacteria increased by 10 to 30 percent. Dobbelaere *et al*. [8] and Cakmakci [7] stated that the bacteria cause increase in leaf area, root and shoot weight and delay in leaf senescence. The experiments done on PGPB inoculation showed that yield and dry matter accumulation in plants like rice [15], barely [7], wheat [4, 11], corn [16] and sugarcane [11] increase.

The present experiment was done to investigate the effect of implying PGPB through inoculation of barely seeds with pure breeds of *Azotobacter* and *Azospirillum*, on growth, yield and other some agronomic characteristics of barley cultivars in Ardabil climate.

MATERIALS AND METHODS

The experiment was conducted in Research filed of College of Agricultural, Islamic Azad University, Ardebil Branch in 2009-2010. The height of the experiment site from the sea level was 1350 m. The mean annual precipitation was 367 mm and the most rainfall concentrated between winter and spring. The soil texture of the Research Site was silty loam with a pH of 7.8.

In this experiment, three autumn cultivars of barley, Makuei, Bahman and CB-7910 were obtained from Agricultural support services of Ardabil. Factors were: seed priming with plant growth promoting rhizobacteria in three levels containing, without priming (as control), priming with *Azotobacter*, *Azosprilium*, *Azotobacter*+*Azosprilium* plus barley cultivars in three levels (Makoei, Bahman and CB-7910). The indigenous soil bacteria studied in this experiment and the inoculum was prepared by the Department of Soil and Water Research Institute of Biology. Seven grams of seed inoculum for inoculation of the bacteria alive is used each of which had 10^7 alive bacteria. The solution of Arabic gum of 15 percent by weight - volume for better adhesion to the seed inoculums was used. The factorial experimental design was a randomized complete block with three replications. In order to prepare the ground using atmospheric - stacker, stack width of 60 cm and sowing the seeds of each cultivar through the open slot on the stack as rigid in working with the density of 350 seeds m was done. Controlling weeds during the growing season was done manually. In order to evaluate the elements of yield and some other related characteristics, such as Number of grains per spike, spike length, plant height, grain weight, ear number per square meter at the end of the main lines 10-15 plants per plot were randomly chosen. Yield was estimated in half a square meter, when crop was harvested. The experimental data were then analyzed using SAS statistical software. The averages were compared using Duncan test at 5% level.

RESULTS AND DISCUSSION

The results of analysis of variance and the effect of priming plant growth promoting bacteria on grain yield, yield components and agronomic traits in barley are given in Table 1.

Plant Height: seed inoculation with PGPR significantly increased the plant height in barley cultivars. Data regarding the effect of barley cultivars and seed inoculation with PGPR on plant height are given in Table 2. In general, the maximum plant height (76.5 cm) was obtained to seed inoculation with Azospirillum, while the least value (63.7 cm) was recorded at control treatment. Increase in plant height, with seed priming in plants such as wheat [4], barley [7], corn [17] and other plants [10] have been reported. Means of comparisons for barley cultivars indicated that the maximum (73.2 cm) plant height was recorded for Makuei cultivar and minimum value (63.2 cm) was recorded for CB-7910 cultivar (Table 2).

Table 1: Analysis of variance for the effects of seed priming with PGPR on experimented traits in barley cultivars

		MS					
S.O.V	df	Plant height	Spike length	Number of spike per area	Number of grains per spike	1000- grain weight	Grain yield
Replication	2	207.8**	1.3**	313725.2	36.1**	83.4**	3678.4**
Barely cultivars (BV)	2	76.1**	0.3**	1976.4**	97.7**	9.9**	1184.6**
Seed priming with PGPR	3	270.9**	0.935**	3983.86**	215.3**	60.6**	1935.8**
BV * PGPR	6	6.1**	0.0388ns	1.137ns	1.193*	0.136ns	15.61ns
Error	22	0.01060	0.0323	58.99	0.284	0.176	201.48

* and **: Significant at 5% and 1% levels respectively

Table 2: Means comparison for yield and agronomic traits of barley cultivars as affected by seed priming with PGPR

Treatment		Plant height (cm)	Spike length (cm)	Number of spike per area (m^2)	Number of grains per spike	1000- grain weight (gr)	Grain yield ($kg.ha^{-1}$)
Barely cultivar	CB-7910	63.29 b	6.15 c	450.1 c	42.4c	49.2 b	2917.4 c
	Bahman	68.6 ab	6.3 b	465.2 b	45.2b	51.0 a	2784.9 b
	Makuei	69.06 a	6.49 a	476.4 a	51.8a	51.1 a	3063.4 a
Plant Growth Promoting Rhizobacteria	Control	63.73 d	5.92 c	441.3 d	44.7b	46.8 b	2763.6 d
	Aztobacter	68.66 c	6.18 b	446.3 c	45.4b	49.6 ab	2904.9 c
	Azosprilium	76.59 a	6.63 a	457.3 a	56.2a	52.9 a	3603.3 a
	Azosprilium + Aztobacter	72.54 b	6.51 a	450.6 b	48.2a	50.1 a	3072.2 b

Means with the same column and factor, followed by the same letter are not significantly difference ($p < 0.05$) using Duncan's multiple range test

Table 3: Mean comparisons of the interaction effect

Treatment	Plant height (cm)	Spike length (cm)	Number of spike per area (m^2)	Number of grains per spike	1000- grain weight (gr)	Grain yield ($kg.ha^{-1}$)
CB-7910 (Control)	66.2g	5.7i	275.7c	42.3i	46.1k	2752.4c
CB-7910 + AZ	71.4d	5.8h	286.9bc	48.1h	48.5h	2851.9bc
CB-7910 + AS	46.3a	6.5c	299.4ab	52.5e	49.2g	3064.7ab
CB-7910 + AZAS	73.4b	6.2e	302.1ab	48.3g	52.1c	3072.8ab
Bahman (Control)	65.3h	6.1f	286.5bc	49.1f	46.6j	2853.8bc
Bahman + AZ	68.1f	6.4d	300.7ab	52.1e	49.5g	3054.1ab
Bahman + AS	72.3c	6.7a	314.9a	61.2c	52.8b	3145.2a
Bahman + AZAS	69.2e	6.6b	311.2a	56.5d	51.1d	3115.4a
Makuei (Control)	61.2i	5.9g	273.5c	57.1d	47.5i	2734.7c
Makuei + AZ	68.3f	6.1f	284.8bc	60.5c	50.3e	2832.6bc
Makuei + AS	73.2b	6.6b	298.2ab	66.6a	53.5a	3062.7ab
Makuei + AZAS	71.1d	6.4d	296.8ab	63.2b	52.1c	3025.5ab

For a given means within each column of each section followed by the same letter are not significantly different ($p<0.05$)

Spike Length: The effect of seed inoculation with PGPR on spike length was significant (Table 1). The maximum spike length (6.63 cm) was obtained using Azospirillum and the minimum spike length (5.92 cm) was gained in control plots (Table 2). Means comparisons indicated that maximum spike length (6.49 cm) was observed for Makuei cultivar and minimum value (6.15 cm) was observed for CB-7910 cultivar. Similar results have been reported by Bashan *et al*. [10] and Cakmakci *et al*. [7].

Number of Spike per Area: number of spike per area was significantly affected by seed priming with PGPR and barley cultivars. (Table 1). The maximum number of spike per area (457.3) was obtained by sole Azospirillum application and minimum number of spike per area (441.3) was gained in control plots (no PGPR priming) (Table 2). These results are in agreement with De Freitas [4] who concluded that number of spike per area in wheat was highest at inoculation with PGPR. It seems that biological control of seedling diseases and proper nutrition has increased the number of spike per area [3, 15]. Mean comparison also indicated that Makuei had more number of spike per area compared with other cultivars.

Number of Grains per Spike: The number of grains per spike was significantly affected by seed priming with PGPR. The maximum number of grains per spike (56.2) was observed in Azospirillum treatment and the minimum number in Azotobacter and control treatments. There was a significant difference between cultivars regarding this trait (Table 2). Maximum and minimum number of grains per spike was observed in Makuei and CB-7910, respectively. Comparison of Average number of interactions with bacteria priming and cultivar showed that the highest number of grains per spike (69.3) belonged to Makuei when priming seed with Azosprilium and the lowest (42.6) belonged to the cultivar CB-7910 without priming. De Freitas [4] reported that the number of grains per spike significantly increased with the effect of different levels of PGPR priming. Similar results has been reported by Cakmakci *et al.* [7] about barely.

One Thousand Grain Weight (1000-Grain Weight): The effect of seed priming with PGPR on 1000-grain weight was significant (Table 1). The maximum 1000-grain weight (52.9 gr) was observes in seed priming of *Azospirillum* and the minimum 1000-grain weight (46.8 gr) in control treatment which was not significantly different from seed priming of *Azotobacter*. Maximum 1000- grain weight (51.1 gr) was produced by cv. Makuei while minimum (49.2 gr) by cv. CB-7910. Golami *et al*. [17] reported that grain weight increased with seed priming with PGPR in maize. Increasing of 1000-grain weight in barley with PGPR priming has been reported by cakmakci *et al*. [7].

Grain Yield: Grain yield is the main target of crop production. The grain yield was significantly affected by both barley cultivars and seed priming with PGPR. Seed priming with PGPR significantly increased the grain yield. The grain yield varied between 2763.6 kg. ha^{-1} in without priming till 3603.3 kg. ha^{-1} in seed priming with Azospirillum (Table 2). Maximum grain yield was produced by Makuei cultivar (3063.4 kg.ha^{-1}) while minimum by CB-7910 cultivar (2917.4 kg.ha^{-1}). Cakmakci *et al*. [7] also reported that PGPR increased plant growth and grain yield significantly in barely. Increasing in grain yeild has been reported by Dobbelaere *et al*. [8]. Significant increase in growth and yield of plants in response to PGPR has been reported in separate experiments by Biswas *et al*. [14], Kloepper *et al*. [12] stated that yield in plants such as rice, corn and sugar cane inoculated with bacteria had 10 to 30 percent increase. The experiment showed that yield an dry matter accumulation when inoculating with PGPR increased in plants such as rice [15], barely [7], wheat [4], corn [16] and sugarcane [11].

CONCLUSIONS

In general, it can be said that all the characteristics were influenced by experimental factors. Plant growth promoting bacteria had a positive effect on all the studied characteristics specially the implication of azosprilium caused significant increase in many of the studied characteristics. Thus, it can be suggested that in order to increase grain yield, dry matter accumulation and other growth indices such as crop growth rate, relative growth rate, Makuei cultivar should be inoculated with Azosprilium bacteria in conditions of the rigion.

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