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THE STUDY EFFECT OF NITROGEN, *AZOTOBACTER* SPP. AND *AZOSPIRILLUM* SPP. ON PHENOLOGICAL AND MORPHOLOGICAL TRAITS OF DURUM WHEAT CULTIVARS IN DEHLORAN REGION, IRAN

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ABSTRACT. This experiment was carried out in the form of factorial split in the model random complete block design in three replicates. In this design three kinds of durum wheat including Yavarous, Karkheh, Seimareh were planted in the main plots and three levels of 40, 80 and 120 kg net nitrogen in hectare which it had been provided from source of urea and three levels of bio fertilizer including *Azotobacter* spp. and *Azospirillum* spp. and without using bio fertilizer (control) as factorial were cultivated in secondary plots. Before cultivation, amount of seeds which it must be mixed with together *Azospirillum* spp. and *Azotobacter* spp. bacteria's, it was mixed together water and sugar with 2% density of wet and in proportion of 2 kg in 100 kg seeds were mixed with seeds. The irrigation of blocks was separately done for preventing from mixture of bacteria. This research will have a new asped for/in order to effect of biological fertilizers in replacement with/to fertilizers and it hasn't demonstrate its comparison on durum wheat in the region by now. Finally, with interpretation of results obtained from this research defined that positive effect of biological fertilizers on growth that before they have verified about plants, also, they are true about grains such as wheat. So, considering the obtained results from this research, it is seemed that application of suitable biological fertilizers can be effective in increase of function, improvement of growing traits of wheat and decrease nitrogen fertilizer.

Key words: Nitrogen fertilizers; Biological fertilizers; Phenological; Morphological; Durum wheat.

INTRODUCTION

Over using of chemical fertilizers for increasing production caused more costs of production associated with the degradation of soil and water resources in worldwide include Iran. Serious environmental degradation due to the misuse of chemical fertilizers caused a critical attention and interest in a healthy crop production in sustainable farming

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systems. Nowadays development of sustainable agricultural systems is a key to combat with the disaster (Ardakani, 2009). Alimadadi *et al.* (2010) reported that one of the strategies for improving crop production and protecting the environment is providing plant growth via using more soil microorganisms and bio-fertilizers instead of chemicals. According to the low level of chickpea yield around the world, it seems that there are still possible methods to increase crop production via farming and breeding methods. The fungi can increase the uptake of water and nutrients, especially phosphorus, zinc, copper and nitrogen via the wide hyphae network (Clark and Zeto, 2000). In many cases, in addition the increasing of production, it plays an important role in maintaining ecological balance in soil (Abbott and Murphy, 2007). Xie *et al.* (2011) reported that N, P, and K are among the most important nutrients for plant growth, and their diverse concentrations have a significant influence on plant growth and soil-plant interactions. Some earlier researchers reported that grain yield per plant was positively and significantly correlated with plant height, cob length, leaf area, number of rows, grain number per cob and 1000-grain weight (Devi *et al.,* 2001; Mohsan *et al.,* 2002; Viola *et al.,* 2003; Farzaneh *et al.,* 2009). The objective of the present paper was to determine the study effect of nitrogen and bio-fertilizer on phenological and morphological traits of durum wheat cultivars in Dehloran region.

MATERIALS AND METHODS

This experiment was conducted in the Dehloran, Ilam, Iran. (47°15′35″E, 32°25′45″N, and 180 m above sea level and average precipitation 226.8 mm year^{-1}). The experiment was randomized complete block design (RCBD) in a split-plot arrangement, with three replicates and three factors (three cultivars, three levels of nitrogen fertilizer and three biologic fertilizer). Treatments were included: in this design three cultivars of durum wheat including Yavarous, Karkheh, Seimareh were planted in the main plots and three levels of 40, 80 and 120 kg net nitrogen in hectare which it had been provided from source of urea and three levels of bio fertilizer including *Azotobacter* spp. and *Azospirillum* spp. and without using bio fertilizer (control) as factorial were cultivated in secondary plots. Before cultivation, amount of seeds which it must be mixed with together *Azospirillum* spp. and *Azotobacter* spp. bacteria's, it was mixed together water and sugar with 2% density of wet and in proportion of 2 kg in 100 kg seeds were mixed with seeds. The irrigation of blocks was separately done for preventing from mixture of bacteria. Each plot consisted of six rows with 5 m length and 1.4 m width. Investigated parameters were the plant height, harvest index (%), grain yield (kg ha^{-1}), biological yield, protein of grain (%), 1000 grain weight (g) and grain spike. The data recorded were analyzed statistically by using statistical software package, MSTATC and graphs were developed using Excel. Mean comparisons were performed using an ANOVA protected least significant difference (Duncan) ($P < 0.05$) test.

Table 1 - Analysis of variance (factorial) for studied traits

S.O.V	df	Plant height (cm)	Grain spike	1000 grain weight (g)	Grain yield (kg ha^{-1})	Biological yield (kg ha^{-1})	Harvest index (%)	Protein of grain (%)
Replication	2	1.25	48.29	7.3	8.49	4.25	86.17	23.4
Cultivar (A)	2	7.11*	3.43^{ns}	4.5**	3.42^{ns}	39673280*	4.11^{ns}	39.13**
Error 1	4	12.14	3.17	98.17	812522	5927457	21	56
Nitrogen (B)	2	19.66**	17.19**	59.27**	3.16**	3.16**	38.2^{ns}	20.1**
Bio fertilizer (C)	2	27.13**	48.42**	8.38**	5.14*	5.14*	26.3^{ns}	8^{ns}
A*B	4	23.21*	42.24**	12.15*	618653^{ns}	618653^{ns}	60.2^{ns}	5*
A*C	4	75.5^{ns}	7.2^{ns}	24.1^{ns}	57.16^{ns}	57.16^{ns}	7.1*	1^{ns}
B*C	4	3.2^{ns}	29.13*	86.8^{ns}	29.4*	29.4*	31.6^{ns}	1^{ns}
A*B*C	8	31.9^{ns}	4.3^{ns}	92.5^{ns}	6.45^{ns}	6.45^{ns}	31.5^{ns}	7^{ns}
Error 2	48	35.1	78.6	7.6	2.38	2.38	31.3	2
C.V (%)	-	3.14	4.11	11.6	8.16	8.16	7.11	5.7

*, ** and ns, indicate significant difference at 5%, 1% probability level, and no significantly, respectively.

Table 2 - The effect of cultivars on studied traits

Cultivar	Plant height (cm)	Grain spike	1000 grain weight (g)	Grain yield (kg ha^{-1})	Biological yield (kg ha^{-1})	Harvest index (%)	Protein of grain (%)
Yavarous	80.59^{a}	27.44^{a}	45.19^{a}	4771^{a}	10361^{a}	46.15^{a}	10.47^{b}
Karkheh	69.96^{b}	21.11^{a}	36.63^{b}	4161^{a}	8493^{b}	49.19^{a}	11.29^{ab}
Seimareh	69.18^{b}	19.66^{a}	41.25^{ab}	4032^{a}	8089^{b}	50.03^{a}	11.87^{a}

Table 3 - The effect of nitrogen, *Azotobacter* spp. and *Azospirillum* spp.

Nitrogen fertilizer	Plant height (cm)	Grain spike	1000 grain weight (g)	Grain yield (kg ha^{-1})	Biological yield (kg ha^{-1})	Harvest index (%)	Protein of grain (%)
40 kg	68.29^{c}	19.7^{b}	37.8^{b}	3982^{b}	8231^{b}	48.68^{a}	11^{c}
80 kg	73.29^{b}	24.07^{a}	41.16^{ab}	4342^{a}	8928^{b}	48.90^{a}	11.2^{b}
120 kg	78.14^{a}	24.37^{a}	44.11^{a}	4640^{a}	9783^{a}	47.78^{a}	11.42^{a}
Bio fertilizer							
Control	63.05b	16.32b	33.29b	3422b	7991a	48.38a	11.17a
Azobactor	74.22a	23.37a	40.98a	4358a	9053a	48.55a	11.18a
Azospirillum	74.74a	23.55a	42.8a	4414a	9162a	48.49a	11.27a

Table 4 - Interaction effects (Cultivars × Nitrogen of fertilizer) on studied traits

Treatments	Plant height (cm)	Grain spike	1000 grain weight (g)	Grain yield (kg ha^{-1})	Biological yield (kg ha^{-1})	Harvest index (%)	Protein of grain (%)
V1*F1	75.77^{b}	23.23^{b}	41.42^{bc}	4268^{bc}	9343^{bc}	45.78^{bc}	10.27^{g}
V1*F2	81.44^{a}	30.44^{a}	45.16^{ab}	4866^{a}	10322^{b}	47.22^{abc}	10.44^{g}
V1*F3	84.55^{a}	28.55^{a}	49.01^{a}	5180^{a}	11418^{a}	45.43^{c}	10.68^{f}
V2*F1	63.88^{d}	18.55^{c}	33.41^{e}	3938^{cd}	7901de	49.98^{ab}	11.14^{e}
V2*F2	71.11^{bc}	20.66^{bc}	35.84^{de}	4213^{bc}	8551^{cde}	49.36^{abc}	11.3^{de}
V2*F3	74.88^{b}	24.11^{b}	40.63^{c}	4330^{b}	9026^{c}	48.22^{abc}	11.42^{cd}
V3*F1	65.22^{d}	17.44^{c}	38.58^{cd}	3739^{d}	7450^{e}	50.28^{a}	11.58^{c}
V3*F2	67.33^{cd}	21.11^{bc}	42.47^{bc}	3947^{cd}	7911de	50.13^{a}	11.86^{b}
V3*F3	75^{b}	20.44^{bc}	42.7^{bc}	4410^{b}	8905^{cd}	49.67^{abc}	12.16^{a}

V1, V2 and V3 indicate Yavarous, Karkheh and Seimareh, respectively; F1, F2 and F3 indicate 40, 80 and 120 kg ha^{-1} nitrogen, respectively.

Table 5 - Interaction effects (Cultivars × biological of fertilizer) on studied traits

Treatments	Plant height (cm)	Grain spike	1000 grain weight (g)	Grain yield (kg ha^{-1})	Biological yield (kg ha^{-1})	Protein of grain (%)
V1*B1	77.55^{b}	25.66^{ab}	43.03^{ab}	4686^{a}	10156^{a}	10.44^{c}
V1*B2	81.22^{ab}	28^{a}	45.55^{a}	4829^{a}	10460^{a}	10.44^{c}
V1*B3	83^{a}	28.66^{a}	47^{a}	4799^{a}	10467^{a}	10.52^{c}
V2*B1	67.88^{c}	20^{c}	35.12^{e}	4022^{b}	8308^{b}	11.23^{b}
V2*B2	71.44^{c}	22.11^{bc}	36.45^{de}	4207^{b}	8596^{b}	11.26^{b}
V2*B3	70.55^{c}	21.22^{c}	38.31^{cde}	4253^{b}	8574^{b}	11.36^{b}
V3*B1	66.88^{c}	18.22^{c}	39.73^{bcd}	3868^{b}	7718^{b}	11.83^{a}
V3*B2	70^{c}	20^{c}	40.93^{bc}	4037^{b}	8104^{b}	11.84^{a}
V3*B3	70.66^{c}	20.7^{c}	43.1^{ab}	4191^{b}	8444^{b}	12.92^{a}

V1, V2 and V3 indicate Yavarous, Karkheh and Seimareh, respectively; B1, B2 and B3 indicate control, *Azotobacter* and *Azospirillum*, respectively.

Table 6 - Interaction effects (Nitrogen of fertilizer × Nitrogen of fertilizer) on studied traits

Treatments	Plant height (cm)	Grain spike	1000 grain weight (g)	Grain yield (kg ha^{-1})	Biological yield (kg ha^{-1})	Protein of grain (%)
F1*B1	66.11^{f}	19.88^{b}	37.03^{e}	3877^{e}	7960^{e}	10.96^{d}
F1*B2	69.77^{def}	19.3^{b}	37.73^{de}	4041de	8214de	10.98^{d}
F1*B3	69^{ef}	20.11^{b}	38.64^{cde}	4028de	8520^{cde}	11.06^{cd}
F2*B1	71.11^{cde}	21.88^{ab}	38.93^{cde}	4222^{cde}	8801^{bcde}	11.16^{bcd}
F2*B2	74.22^{bcd}	25.4^{a}	41.68^{bcd}	4337^{bcd}	8826^{bcde}	11.18^{bcd}
F2*B3	74.55^{bcd}	24.8^{a}	42.86^{abc}	4467^{abc}	9158^{abcd}	11.26^{abc}
F3*B1	75.11^{bc}	22.11^{ab}	41.91^{bcd}	4477^{abc}	9421^{abc}	11.39^{ab}
F3*B2	78.66^{ab}	25.3^{a}	43.52^{ab}	4695^{ab}	10121^{a}	11.39^{ab}
F3*B3	80.66^{a}	25.6^{a}	46.9^{a}	4749^{a}	9807^{ab}	11.49^{a}

F1, F2 and F3 indicate 40, 80 and 120 kg ha^{-1} nitrogen, respectively; B1, B2 and B3 indicate control, *Azotobacter* and *Azospirillum*, respectively.

RESULTS AND DISCUSSION

Result of analysis of variance (*Table 1*) showed that there was significant difference among different cultivars, nitrogen and bio fertilizer in the majority of traits expect of some traits.

The data showed that the highest plant height, 1000 grain weight and biological yield were attained from Yavarous of cultivar but the highest protein of grain was attained from Seimareh of cultivar (*Table 2*).

Our results showed that all growth parameters increased gradually by increasing nitrogen and application of *Azospirillum* as compared with the untreated plants (*Table 3*). These results were in agreement with those reveled by Gezing *et al.* (2001) and Osman *et al.* (2005).

Interaction effects (Cultivars × Nitrogen of fertilizer) on traits showed that the highest plant height, biological yield, grain yield and 1000 grain weight (g) were attained from V1 × F3, the highest grain spike was attained from V1 × F2, the highest harvest index was attained from V3 × F1 and the highest protein of grain was attained from V3 × F3 (*Table 4*).

F1, F2 and F3 indicate 40, 80 and 120 kg ha^{-1} nitrogen, respectively.

Interaction effects (Cultivars × biological of fertilizer) on traits showed that the highest plant height, grain spike, 1000 grain weight (g) and biological yield were attained from V1 × B3, the highest grain yield was attained from V1 × F2, the highest harvest index was attained from V3 × B1 and the highest protein of grain was attained from V3 × B3 (*Table 5*).

Interaction effects (Nitrogen of fertilizer × Nitrogen of fertilizer) on traits showed that F3 × B3, increased plant height, grain spike, 1000 grain weight (g), grain spike and grain yield as compared to the other treatments (*Table 6*) and the highest biological yield was attained from F3 × B2 (*Table 6*). Application of biochimical and bio fertilizer on phenological and morphological traits have been identified that can extend phonological and morphological traits in many crops (Farzaneh *et al.,* 2009; Alizadeh *et al.,* 2008).

CONCLUSION

The results showed that with application of *Azospirillum* spp. and *Azotobacter* spp. bacteria's in wheat cultivation can reduce the use of nitrogen fertilizer. Therefore with right combination of chemical fertilizer and biological fertilizer we can achieve to the expected yield, with lower danger for environment. The results of the present study are important in application of *Azospirillum* spp. and *Azotobacter* spp. bacteria's in low input and sustainable farming systems.

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