

## Effect of Bio-Bacterial (Azetobacter, Azorhizobioum, Azospirillum) on Yield and Yield Components of Rice in Bandar-Anzali, North of Iran

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**Abstract:** A factorial test in the framework of designing complete random blocks with 3 replications was conducted in 2011 in Bandar-Anzali (North of Iran) to study the effect of assimilating stabilizer bacteria of nitrogen (Azetobacter, Azorhizobioum and Azospirillum) on the yield and yield components of rice. Given factors of this study include Azetobacter, Azorhizobioum, Azospirillum bacterium in two levels each (with and without bacteria). Results showed that Azospirillum treatment had a meaningful effect on the weight qualifications of a thousand grains rice with 5% probability level. Reciprocal effect of Azetobacter and Azospirillum on the qualification of the grain number in a rice cluster in 1% probability level was meaningful. Reciprocal effect of Azetobacter and Azorhizobioum on the length of the stem and number of grains in the rice cluster in 5% probability level became meaningful. Reciprocal effect of Azospirillum and Azorhizobioum on the nitrogen content of the rice grain in 5% probability level was meaningful. Reciprocal effect of Azetobacter, Azospirillum and Azorhizobioum on the operation of the grain and rice harvest index in 5% probability level and on the nitrogen content of rice grain in 1% probability level became meaningful.

**Key words:** Rice, Azetobacter, Azospirillum, Azorhizobioum, yield, yield components

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### INTRODUCTION

Rice is a key nutrient product all over the world. Rice grain and its resultant products provide almost the 40% food of half of the people in the world (Rabiee, 1996). Bacterial biologic fertilizers are proper alternative or complements for chemical fertilizers and can have useful effects on the improvement of the soil physical and chemical characteristics (Astarai and Kouchaki, 1996). Rice grain yield increases with the increase of nitrogen (Carreres *et al.*, 2000) but planting efficacy decreases with the increase of consumed nitrogen; in other words, grain yield increase less with the increase of consumed nitrogen. Increased nitrogen fertilizer from 60-120 kg ha<sup>-1</sup> increased the number of clusters in square meters but increasing it to 150 kg ha<sup>-1</sup> resulted in the decrease of the number of clusters (Faraji and Mirlohi, 1998). Reddy (1986) reported that increased nitrogen from 0-180 kg ha<sup>-1</sup> increased the number of tiller in each pile, the number of filled grains in each cluster and the weight of each grain and also the total number of stems. The effect of Azetobacter fertilization (Ram *et al.*, 1985), especially with Azospirillum (Rai and Gaur, 1988) on the growth and

yield of wheat has been reported positive and meaningful. Positive effect of fertilizing Azetobacter on other products such as corn and rice is also reported (Daneshmand *et al.*, 2006). Rai and Gaur (1988) studied the effects of Azetobacter and Azospirillum, alone and together on the growth and yield of wheat in which the highest grain yield was when these two bacteria were added to the soil, mixed. Abdus-Sattar *et al.* (2006), studied the effect of fertilizing Azetobacter with Azospirillum on rice cultivation in 5 levels of nitrogen fertilizer (120, 100, 80, 60, 0 kg N ha<sup>-1</sup>) and Blue Green Alga (BGA) cultivated with rice in 4 levels of nitrogen fertilizer (40, 30, 20 kg N ha<sup>-1</sup>) in two experimental fields in Bangladesh. Maximum grain yield was registered with Azospirillum in 100 kg ha<sup>-1</sup> nitrogen. A research was conducted in India on the effect of fertilizing Azetobacter on grain or seedlings of wheat, rice, onion, eggplant, tomato and cabbage and yield increase was observed in all cases however this effect was meaningful just for rice, cabbage and eggplant (Subba-Rao, 1988). Ebrahimi in a study on the effect of the amount of nitrogen fertilizer (0, 30, 60, 90 kg ha<sup>-1</sup> nitrogen (from urea fertilizer source) and Azospirillum bacterium (fertilization and unfertilization) on rice

genotype showed that nitrogen fertilizer and Azospirillum consumption in comparison with control treatment (nonconsumption of nitrogen fertilizer and Azospirillum) meaningfully increased grain yield. Soil increased productivity by biologic fertilizers such as Azetobacter, Azospirillum and Pseudomonas increases and develops growth features of the medicinal plant, *Nigella sativa* such as length, number of lateral bough, number of capsules each bush and grain yield (Shaalan, 2005). Babaei found out that fertilizing sunflower seed with Azospirillum increased length of stem stalk and stem and stalk weight in comparison to unfertilization situation. Parsaei-Mehr in a study on the effect of biologic and Azospirillum fertilizers on increasing consumption nitrogen in wheat cultivation concluded that Azospirillum bacteria along with Azetobacter had a positive and meaningful effect on maximum leave level index, grain yield and protein percentage. The goal of this research is to study the effect of biologic fertilizers on the yield and yield components of rice in North of Iran.

**MATERIALS AND METHODS**

This research with the goal of studying the effect of biologic fertilizers on the yield of rice in 2010-2011 was conducted in Bandar-Anzali (North of Iran). So, a factorial test was conducted with the basic plan of complete random block with 3 repetitions. Annual rainfall amount in the studied site was 1563 mm. Examined factors included Azospirillum, Azetobacter and Azorhizobioum bacteria in two levels each (existence and nonexistence of bacterium). Healthy and monotonous symbols of Hashemi rice was considered with 20×20 spacing and four seedlings in each pile. Amount of fertilizer was selected considering the soil test, plant need and source examination. To provide used fertilizer sources in rice cultivation, potassium solphat fertilizer was scattered in the field and then cultivated transplants without any fertilizer and put transplants with biologic fertilizers in solutions, made of these fertilizers

and then cultivating these transplants began (bacterial biologic fertilizers was given to the stem by fertilizer fertilization). MSTAT-C software was used for statistical analysis of data and data mean comparison test was conducted with Tooke method in 5% probability level.

**RESULTS AND DISCUSSION**

**1000 kernel weight:** Results show that effect of Azospirillum fertilizer on the 1000 Kernel weight in 5% probability level is meaningful (Table 1) and Azospirillum usage treatment with thousand kernel weight mean, 25.49 g showed more meaningful increase than control treatment which indicates that Azorhizobioum bacterium did not have any meaningful effect on thousand rice kernel weight, on its own (Table 2). Yasari and Patwardhan (2007) stated the increase of weight of a thousand corn grain through fertilization with *A. lipoferum*. Gupta and Samnotra (2004) reported that using Azetobacter and Azospirillum of a kind of biological fertilizer increased the weight of a thousand Colza seed because of increase in absorbing nutrient elements by the plant because of fertilization with Nitroxine. Yasari and Patwardhan (2007) reported a positive and meaningful increase in the weight of a thousand sesame seed because of using Azetobacter and Azospirillum. Gholami *et al.* (2009) also expressed an increase in the weight of corn grain in all fertilized treatments with Azospirillum.

**Grain yield of rice:** Reciprocal effect of Azetobacter, Azospirillum and Azorhizobioum on the yield of rice grain is meaningful in 5% probability level (Table 1). It increased the yield in the mean comparison test of using Azorhizobioum in Azetobacter attendances which shows that simultaneous usage of them has a better effect on grain yield. The highest grain yield was achieved in Azetobacter-Azorhizobioum treatment with 290.77 g m<sup>-2</sup> (Table 3). The least seed operation belongs to Azetobacter with only 196.55 g m<sup>-2</sup>. Differences in other treatments were not meaningful (Table 3).

Table 1: Mean squares of the experimental treatments on studied traits in the factorial test

SOV	df	Mean squares					
		1000 kernel weight	Grain yield	Plant height	No. of grain per cluster	Harvest index	Nitrogen content of grain
Replication	2	0.016	127.189	30.774	5.652	11.158	9.622
Azetobacter (A)	1	0.082	1176.000	2.100	185.927	21.751	25.890
Azospirillum (B)	1	1.402*	444.792	34.320	39.527	6.078	3.897
A×B	1	0.107	0.073	12.760	737.400**	0.210	0.023
Azorhizobioum (C)	1	1.042*	1075.217	4.594	7.042	0.045	0.041
A×C	1	0.240	2453.899	131.100*	368.167*	8.260	22.820
B×C	1	0.167	2889.060	6.304	238.140	5.612	41.584*
A×B×C	1	0.375	8970.666*	0.070	20.535	49.421*	106.248**
Error	14	0.180	1811.025	26.358	78.355	8.430	8.58
C.V	-	1.680	17.660	4.800	10.100	10.710	13.51

\*\* and \*significant at 1 and 5% probability levels, respectively

Table 2: Mean comparison of two some effects experimental treatments on studied traits in the factorial test

Treatment	Nitrogen content of grain (g m <sup>-2</sup> )	Number of grains per cluster	Plant height (cm)
Azetobacter (Azospirillum)	22.34	80.61	104.71
Azetobacter (without Azospirillum)	23.08	89.13	108.56
without Azetobacter (Azospirillum)	20.20	97.26	106.76
without Azetobacter (without Azospirillum)	21.07	83.61	107.70
Q	6.28	10.84	3.58
Azetobacter (Azorhizobioum)	109.41	89.33	23.65
Azetobacter (without Azorhizobioum)	103.86	80.41	21.78
without Azetobacter (Azorhizobioum)	105.33	87.06	19.62
without Azetobacter (without Azorhizobioum)	109.13	93.81	21.65
Q	6.28	10.84	3.58
Azospirillum (Azorhizobioum)	105.66	86.33	19.91
Azospirillum (without Azorhizobioum)	105.81	91.55	22.63
Without Azospirillum (Azorhizobioum)	109.08	90.06	23.35
Without Azospirillum (without Azorhizobioum)	107.18	82.68	20.80
Q	6.28	10.84	3.58

Table 3: Mean comparison of triple effects experimental treatments on studied traits in the factorial test

Treatment	Grain yield (g m <sup>-2</sup> )	Harvest index (%)	Nitrogen content of grain (g m <sup>-2</sup> )
Azetobacter, Azospirillum, Azorhizobioum	238.66	26.28	19.85
Azetobacter, Azospirillum, without Azorhizobioum	265.66	29.03	24.83
Azetobacter, without Azospirillum, Azorhizobioum	290.77	30.94	27.44
Azetobacter, without Azospirillum, without Azorhizobioum	196.55	26.02	18.73
Without Azetobacter, Azospirillum, Azorhizobioum	243.21	25.89	19.97
Without Azetobacter, Azospirillum, without Azorhizobioum	233.33	25.25	20.43
Without Azetobacter, without Azospirillum, Azorhizobioum	217.77	25.18	19.26
Without Azetobacter, without Azospirillum, without Azorhizobioum	241.33	28.34	22.87
Q	73.70	5.02	5.07

Ebrahimi showed that using nitrogen fertilizer and Azospirillum meaningfully increase grain yield of rice genotype in comparison with control treatment (nonconsumption of nitrogen fertilizer and Azospirillum). Malik *et al.* (2002) suggested yield increase of 18-32% because of fertilization of rice seed with Azospirillum. A research was done in India about the effect of Azetobacter fertilization on rice grain which resulted in increased rice yield (Subba Rao, 1988).

**Number of grain per cluster:** Reciprocal effect of Azetobacter and Azospirillum on the number of grains per cluster in the probability level of 1% was meaningful (Table 1) in which the most amount of grain per cluster belong to Azospirillum treatment 97.2 and the least amount was achieved from Azetobacter-Azospirillum and control, respectively 80.617 and 83.617 (Table 2). In the situation when fertilizer treatment of Azetobacter and Azospirillum was used, Azetobacter fertilizer resulted in the increase of grain number per cluster but Azospirillum resulted in the decrease of grain number per cluster which shows that Azospirillum has a positive effect on the increase of rice yield in the situation of consolidating Azetobacter and Azospirillum.

Reciprocal effect of Azetobacter and Azorhizobioum on the number of grain per cluster is meaningful in 5% probability level (Table 1) in which the most amount of grain was observed in control treatment (93.8). The least

amount of grain per cluster was observed in Azetobacter treatment (80.4) and does not have any meaningful difference with Azetobacter-Azorhizobioum treatments (Table 2). When Azetobacter fertilizer was used, Azorhizobioum fertilizer usage resulted in the increase of grain number per cluster but Azetobacter did not result in increase which shows that consolidating these two bacteria with each other does not have any effect on the increase of grain number per cluster.

Ozturk *et al.* (2003) reported that fertilizing wheat seeds with Azospirillum meaningfully results in the increase of the number of panicle in m<sup>2</sup>, the number of grain per panicle, seed yield and protein content. Barik and Goswami (2003) stated that fertilizing wheat seeds with Azetobacter and Azospirillum with a positive effect on the number and yield of grain can finally reduce the use of chemical fertilizers for producing wheat. Results of the study by Yasari and Patwardhan (2007) indicated that using Azetobacter and Azospirillum in comparison with control (nonusage of bacteria), increases sesame yield to 21.7 and has a positive and meaningful effect on the number of capsule per bush, the number of subordinate cluster and weight of a thousand grains but on the contrary, decreases the number of grains per capsule. In another study on the effect of using biologic fertilizer, Zn sulfate and nitrogen fertilizer on wheat qualitative and quantitative operation, the most value of grain was achieved from the treatment related to

Azetobacter fertilizer, Zn usage and second level factor of nitrogen fertilizer division (Khassehesirjani *et al.*, 2011).

**Plant height:** Reciprocal effect of Azetobacter and Azorhizobioum showed that compound Azetobacter and Azorhizobioum bacteria with 109 cm height had a higher stalk than Azetobacter treatment, 103 cm (Table 2).

Zahir *et al.* (2000) also reported height increase of corn bush because of fertilizing its grains with Azetobacter bacterium. Other researchers also believe that the increase of rice height is completely dependent on absorbable nitrogen of soil (Liagas *et al.*, 1987; Islam *et al.*, 2008; Rezaei *et al.*, 2009; Bindra *et al.*, 2000; Sharief *et al.*, 2006; Navin *et al.*, 1996; Hussain *et al.*, 2006).

**Harvest index:** Triple reciprocal effect of bacteria on rice harvest index is meaningful in 5% level (Table 1). The highest cultivation index is related to using Azetobacter and Azorhizobioum, 3.09% and the lowest harvest index was observed in Azorhizobioum treatment (25.18) (Table 3). As soil nitrogen increases growing growth more than productive growth, it would be expected that increasing soil nitrogen decreases harvest index. Khorshidi and Ardakani (2011) reported the increase of rice harvest index because of using Azospirillum and Pseudomonas bacteria while fertilizing grains of fennel with Azetobacter did not have any effect on harvest index (Tehlan *et al.*, 2004).

**Grain nitrogen content:** Double reciprocal effect of Azospirillum and Azorhizobioum on rice grain nitrogen content is meaningful in 5% level (Table 1). The most content of grain nitrogen was related to Azorhizobioum treatment (23.35) and the least content of nitrogen grain was related to Azospirillum and Azorhizobioum (19.91) which indicates a positive effect of Azorhizobioum in the consolidation situation of the two bacteria, Azospirillum and Azorhizobioum (Table 2).

Triple reciprocal effect of Azetobacter, Azospirillum and Azorhizobioum on the nitrogen content of rice grain is meaningful in 1% level (Table 1). The most nitrogen content is related to Azetobacter-Azorhizobioum treatment (27.442) and the least is related to Azetobacter treatment (18.734) which indicates the positive effect of consolidation Azetobacter and Azorhizobioum in increasing grain nitrogen content (Table 3). Pandey *et al.* (1998) also observed a considerable increase of nitrogen and phosphorus of different parts of bush and the increase of corn yield because of the effect of grain fertilization with Brasilense Azospirillum bacterium. Stancheva and Dinev (2003) stated that interaction between corn stem

system and Brasilense Azospirillum bacterium increases biomass and nitrogen amount of the whole bush. De Freitas (2000) reported that fertilizing wheat seeds with Azospirillum resulted in the increase of nitrogen amount in stalk texture of this plant. The effect of Azetobacter bacterium on growth features, yield and yield components of corn was studied in a research. Biari showed that fertilizing corn with Azetobacter caused the grain weight per bush, total weight of bush and the amount of nitrogen and zinc per seed be meaningfully affected in comparison with control. Zahir *et al.* (2000) reported the increase of grain nitrogen density because of simultaneous fertilization of corn grain with Pseudomonas and Azetobacter. Kumar and Ahlawat (2006) also observed positive effects of Azetobacter on growth improvement, yield, yield components and nitrogen absorption in the wheat (*Triticum aestivum* L.) in comparison with control treatment. Narula *et al.* (2000) reported that some races of Azetobacter in greenhouse situation resulted in the increase of producing stem mass and also nitrogen, phosphorus and potassium absorption in wheat. Hajji-Boland *et al.* (2004) reported that fertilizing wheat with Azetobacter in comparison with using nitrogen fertilizer, simultaneous usage of nitrogen fertilizer and Azetobacter and control, increased nitrogen density in aeral organ and plant stem. They have also stated that nitrogen absorption and transformation to wheat aeral organ in the given attendance was more than other treatments.

## CONCLUSION

In general, results showed that using biologic fertilizers for rice results in the increase of the power of midtillering stem bulk, diameter and length of stalks and number of grains per cluster. Azetobacter fertilization has shown a positive effect. Azospirillum biologic fertilizer usage increases the weight of a 1000 kernel weight and the number of grains per rice cluster. Simultaneous usage of Azetobacter and Azorhizobioum biologic fertilizers increases rice grain yield.

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