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The response of yield and related traits of pinto-bean cultivars towards the use of nitrogen chemical and biological fertilizers

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Key words: Chemical and Biofertilizer, Sustainable Agriculture, Pinto bean.

Abstract

By replacing the chemical fertilizers through biological fertilizers, we can increase the yield of organic agricultural products and decrease production costs, pollution of environment and at least to reach a lasting yield in agriculture. For this purpose, effect of chemical and nitrogenous biological fertilizers has done on some traits of different cultivars of pinto-beans an experiment was conducted in the split plot factorial based on completely randomized block design with three replications during growing seasons of 2013-2014. In this experiment, The studied factors were: 6 cultivar of pinto bean such as (Talash, local Aghgouneh from Sarab, line cos16, Ghermez ghuneh From Sarab, Sadri and Khomein) as the main plot, nitrogen fertilizer in three levels as second factor: (0-50 and 100% recommended amount), biologic fertilizer in two levels: (inoculation with super plus rhizobium and without inoculation) is sub second factor. The effect of two fertilizers were significant on all the traits and the use of nitrogen fertilizer and inoculation of seeds by Rhizobium caused increasing of amount of all traits rather than traits of the controll, as the amount of the grain per pod, grain number and weight were more in all levels which were treated by biologic fertilizer rather than without biological fertilizer. In the situation of use and disuse of biologic fertilizer, the high degree in use of nitrogen fertilizer of treatment attendance up to 100% in all levels and in both situation of use and disuse of biologic fertilizer led the grain to be filled very quickly. Using the biologic fertilizer with nitrogen fertilizer caused the decrease of filling of the line cos16 and Talash. The effects of three factors were significant on the 100 kernel weight. The highest and the least 100 kernel weight in local Aqgune and line cos16 were 21.51 and 27.82 grams. In this research the seed yield which was product by biologic fertilizer with 50% chemical fertilizers more than of 100% chemical fertilizer, so far reached to needed yield, we can decrease use of chemical nitrogen fertilizer and use it as strategy for getting the sustainable agriculture.

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Introduction

The stabilization of biologic nitrogen is one of the most important aspects in sustainable agriculture and its safe according the biologic environment and supports the production power of agricultural plants for a long period (Caranca *et al*., 2009). Despite the importance of the chemical fertilizers in increasing the yield and fields fertility this compounds are one of the environmental pollutions specially, in aqueous environment. The extreme use and absorption of these fertilizers which leads to poisoning will increase density of these elements in plant tissue and imbalance of elements. Other than this, chemical fertilizers will decrease the absorption of copper, iron and some other micronutrients by root. Easy accessibility and low price of chemical fertilizers has caused disuse of biologic fertilizers and have been forgotten for a long time. By revealing the negative effects of chemical fertilizers consumption and its increasing price has occurred to use the biologic fertilizers in agriculture in recent years again (Singh and Kappor, 1998). Extreme use of nitrogenous chemical fertilizers caused the environmental pollution which has negative effects on soil quality (Gheysari *et al*., 2009). Stable amount of nitrogen is being controlled by 4 factors: 1) efficiency of Rhizobioms coexistence. 2) Storage power efficiency of host plant for amount of collected nitrogen. 3) Amount of nitrogen in soil. 4) Effective environmental pressures on stabilizing nitrogen in soil (Kesel and Hartley, 2000). Agriculture always depends on large amount of stabilized biological nitrogen for producing. Recognizing the fact that the annual part of stabilized nitrogen by plants is 4 or 5 times more than all nitrogen fertilizers which are produced via industries, the importance of biologic nitrogen can be revealed. Everyday growing of the world population and their growing supply for food will add to the importance of the biologic nitrogen (Lemke *et al*., 2009). Research about recognition of the effects of the chemical and biological fertilizers on quality and quantity yield of bean plants is one of the researching fields in betterment of the quality and increasing of its products. Results of research on vetch showed that the inoculation with Rhizobium bacteria caused the significant increase its dry aerial organs weight rather than controlled treatment. Bambara *et al.,* (2010) showed that inoculation with Rhizobium bacteria increased dry weight of plant and number of pods rather than control. Single and twofold inoculation of Rhizobium bacteria and solvent phosphate bacteria increased the root and shoot dry weight, plant height, spike height, seed yield, seed protein content and leaf protein of wheat (Saharan *et al*., 2011). Reports indicate that the Rhizobium bacteria caused increase in leaf dry weight (Franzini *et al*., 2009). Rodriguez-Navarro *et al.,* (2000) observed mutual effects among different degrees of bacteria and number of number of increased nodulation and growing of plant, biomass and mixture of sap of xylem. Namvar *et al.,* (2012) to evaluate the effects of chemical nitrogen fertilizer and biologic fertilizer on yield and yield components of bean and sunflower oil announced that the most plant height and anthodium diameter, number of seeds per anthodium and 100 kernel weight are achieved from high level inoculation of nitrogen fertilizer and biologic fertilizers. Mehrpouyan *et al*., (2009) reported that inoculation of different sort of bean with biological fertilizer has caused some permanent features which are in quality and quantity rather than without inoculation form or even rather than 100 kg of pure nitrogen fertilizer. According to research in Egypt by EL-Zeiny (2007), compounding of been seeds with two kinds of biologic fertilizers (Phosphorine and Microbine) has increased growth rate, also it was more with inoculation with Phosphorine rather than control. Bhattarai *et al.,* (2011) studied 4 treatments: Rhizobium mycorrhizal, Diamonium phosphate fertilizer in amount of 100 kg per hectare and control was on red bean. They observed that treatment inoculated with Rhizobium has significant at 5% on some traits like dry weight, root height and shoot, number of pod and the number of knots rather than the treatment. The grain yield between treatment with Rhizobium and with fertilizer was not very different. Yazdani *et al.,* (2009) declared that inoculation with phosphate bacteria and

mycorrhizal Arboskular in mushroom decreased use of Phosphorus fertilizer up to 50% without decreasing of corn yield. According to studies of Mishra *et al.,* (2010) using chemical and biological fertilizers will do betterment in growth and yield of peas. Noticing the negative effects of chemical fertilizers on human health and decrease of quality of agriculture productions, the importance of the high need of plants to nitrogen and role of elements in growth and role of it in growing of plants, the current using the different levels of starter nitrogen fertilizer and biologic fertilizer for preparing the feeding elements will be revealed.

In this research, evaluate effects of chemical and biological fertilizers on grain production and grain related traits of pinto bean cultivars.

Materials and methods

This experiment has done in 2012 in Marand city, located at 60 km of south west of Tabriz, Iran. This area has geographical longitude of 45 ˚20' and 38 ˚20' northern with average height of 2200 meters from sea with semi-arid climate. The Temperature in the most hot and cold months of the year are July - August with 33.6 ℃ and December -5.3 ℃ and the average annual heat is 8.10 ℃ with average humidity of 65.9%. This experiment was conducted in the split plot factorial based on completely randomized block design with three replications during growing seasons of 2013-2014. The studied factors were: 6 cultivar of pinto bean such as (Talash, local Aghgouneh from Sarab, line cos16, Ghermez ghuneh From Sarab, Sadri and Khomein) as the main plot, nitrogen fertilizer in three levels as second factor: (0-50 and 100% recommended amount), biologic fertilizer in two levels: (inoculation with super plus rhizobium and without inoculation) is sub second factor. In this research dimension of each main plot is 20 × 3 meters. Distance of planting between rows is 50 cm and on the rows 5 cm and after thinning it reached to 10 cm. depth of the planting of seed is 7-10 cm and distance between replication is 1.5 meter. Before planting, the soil of field was sampling for physico-chemical characters recognizing. The results of soil analysis are given at the table 1.

Table 1. Soil properties measured prior to the initiation of the experiment.

Depth (cm)	Phosphor (ppm)	Nitrogen (%)	Potassium (ppm)	OM (%)
0-30	20	0.046	169	0.52

According to results of analyze of soil, amount of urea fertilizers with 100 kg/ha. The amount of rhizobium super plus biological fertilizer was 2 kg/ha which seeds for sub secondary plots that should have inoculated with bacteria, mixed with %20 water and sugar and got wet and mixed with rhizobium super plus fertilizer in shade and then they dried. The measured traits were seed number per pod, seed number per plant, 100 kernel weight, seed weight per plant, grain filling rate and effective filling period.

Statistical analysis

For analyzing data MSTATC software, for recognizing correlation SPSS software, for comparing average Duncan test in %5 and for drawing Fig.s Excel program were used.

Results and discussion

The result among different cultivars showed that use of chemical and biological fertilizers has significant difference with observed traits. The interactions of observed factors were significant in most traits, also (table 2).

Seed number per pod

Mean comparison showed that use of biological and chemical fertilizer increased seed number per pod. Seed number in all nitrogenous levels with biologic fertilizer was more than without biologic fertilizer. In use and non use conditions, biologic fertilizer increased the seed number per pod. This increasing in using biologic fertilizer treatment 50% and 100% recommended amount of nitrogen were 17.84% and 27.36% and in without using of biologic fertilizer they were 39.35% and 45.43%, respectively (Fig. 1). For every unit increase of nitrogen with inoculation of

seeds with rhizobium super plus, increase seed number 0.63 unit and in without inoculation condition it will increase seed number 0.83 unit. The highest seed number achieved in Sadri cultivar with average of 5.38. Aqgune and Ghermezgune and linecos16 cultivars had the lowest seed number which did not have statistically significant different with Khomein cultivar (Fig. 2). Yadegari (2013) reported that inoculation of bean seeds with rhizobium increase seed number per pod. Sharifi and Haghnia (2010) declared that according to a research on effect of biological nitroxin fertilizer on the yield and yield components of wheat, effect of this biological fertilizer on average of seed number per spike were not significant. Khalequz Zaman and Hossain (2007) reported that in their experiments, inoculation of bean seeds with Rhizobium and biological fertilizers had not significant effect on seed number per pod. Radwan and Awad (2002) reported that presence of Azotobacter biological fertilizers and Azosperilium biological fertilizers with organic remains in soil did not have significant effect on seed number per pod in peanut. Noruzi *et al.*, (2010) reported that among bacteria treatments, effect of using rhizobium bacteria in pea showed that treatments with inoculation with bacteria in comparison with treatments without inoculation, has increased seed number per pod. Safapour *et al.*, (2010) reported that in different genotypes of bean with different growth type had significant differences in the seed number per pod. Mohammadi *et al.*, (2011) reported that there was significant difference in compare of inoculation and not inoculation of seeds in %1 in number of seeds, but Yadegari *et al.*, (2003) in research on two kinds of soybean, inoculated with Rhizobium japonicum bacteria, interaction of Strain and cultivar had not significant difference in seed number per pod.

Table 2. Analysis of variance for studied traits in 6 varieties of pinto bean under use of chemical and biological fertilizers.

S.O.V.	df	M.S.					
		Seeds Number per pod	seed Number per plant	seeds Weight per plant	100-seed weight	Seed filling rate	Effective seed filling period
R	2	0.454^{*}	4462.954^{**}	487.917^{*}	5.373^{**}	$4/634^{ns}$	0.495^{**}
Cultivar A	5	0.770^{**}	28434.881^{**}	4626.339^{**}	1703.932^{**}	$6782/222^{**}$	27.485^{**}
Error	10	0.076	380.587	79.289	329.672	3/797	2.098
nitrogen B	2	21.454^{**}	136582.704^{**}	20883.355^{**}	596.952^{**}	$7926/240^{**}$	4356.54^{**}
A×B	10	0.076^{ns}	3508.737^{**}	285.847^{**}	28.573^{ns}	$171/582^{**}$	29.372^{**}
Error	24	0.194	438.676	38.409	25.573	3/534	0.861
Biofertilizer C	1	12.000^{**}	52448.148^{**}	9935.240^{**}	195.267^{**}	$1967/608^{**}$	47.730^{**}
A×C	5	0.533^{ns}	989.504^{ns}	89.217^{ns}	16.505^{ns}	$23/590^{**}$	6.730^{**}
B×C	2	0.861^{*}	2176.259^{*}	541.749^{**}	28.518^{ns}	$81/678^{**}$	7.847^{**}
A×B×C	10	0.261^{ns}	396.981^{ns}	73.668^{ns}	31.412^{ns}	$22/078^{**}$	9.583^{**}
Error	36	0.250	477.500	39.059	21.285	2/395	0.802
CV		9.93	14.70	10.29	11.26	1.88	1.87

ns: non significant, * and ** significant at 5% &1% respectively.

Seed number per plant

In this research with high use of nitrogen fertilizer seeds number per plant increased. The highest increasing seeds number per plant was for Talash cultivar with 86.08 units for every unit increase of nitrogen. The local Ghermezgune with 75.83 units increase had lowest increase for every unit increase of nitrogen (Fig. 3). Seeds number per plant in all levels of nitrogen with treatment of biologic fertilizer was more than without using of biologic fertilizer. Increase of using nitrogen was leading to increase seed number per plant from 50% to 100%. This increase in use of biologic fertilizer following the increase of supplied nitrogen is from %50 to %100 which equals to 19.53 % and in the situation of without using biologic fertilizer it was 25.36%, but in

the situation of inoculation with rhizobium super plus the increase of nitrogen from 0-50% did not have any effect on increasing this feature. Increase of per unit of nitrogen in the condition of inoculation with bacteria is 18.55 units and without inoculation increased 29.3 units in seed number per plant (Fig. 4). In research of Upadhyay *et al.,* (1999) treatment inoculation caused significant increase in seed number per plant. Yazdani *et al*., (2009) showed that corn weight, number of rows and seed number per row by use of stimulus bacteria for growth and phosphate solubilizing as the chemical fertilizer complimentary increased. Houshmand (2006) reported that use of Thiobacillus along with phosphate solubilizing has decreased the seed number to 8% in corn. Yadegari (2013) reported that use of rhizobium increased seed number per plant.

Seed weight per plant

With increasing use of nitrogen, seed weight per plant increased. The most seed weight per plant was in Sadri and local Khomein (113.12 g. and 87.64 g. respectively) which obtained with use of 100% recommended fertilizer. This amount of seed yield per plant recommended with 50% supply of fertilizer and without using of it was 24.44 and 130.9% in Sadri and 22.66 and 156.09% more in local Khomein, respectively. The most increase was in Sadri. Increase of nitrogen per unit in Sadri, increased 32.07 units in seed weight. The least increase was about line cos16 with 15.5 units. By increase of nitrogen per unit, seed weight per plant was 25.96 units by presence of biologic fertilizer and 21.2 units by not being of bacteria has increased (Fig. 5). Seed weight per plant in all levels of use of nitrogen in effect of treatment with biologic fertilizer was more than without using of biologic fertilizer. In the situations of use and without using of biologic fertilizer, the increase in use of nitrogen increased the seed weight per plant. This increase in the situation of use of biologic fertilizer following increase of consumable nitrogen from 0-50 and 100% were in order of 90.9 and 15.37 and in the situation of without using of biological fertilizer were 84.32 and 39.97%, respectively (Fig. 6). This difference shows that the existence of nitrogen leads the increase of yield, also Rhizobium super plus which contains nitrogen stabilizer for bean, could cover a better situation rather than the other treatments. So by use of consolidated chemical and biological fertilizers can reach better yield in this plant. Rather than bacteria important role in fixing nitrogen, they can cause an improvement in growth of plants by their effect on better absorption of other necessary elements (Mehrpouyan *et al*., 2010). Yadegari *et al*., (2004) reported that inoculation of red bean seeds with different ways of bacteria cause an increase in seed weight per plant.

100 kernel weight

According to results of the compares of the highest and lowest average of 100 kernel weight in local Aqgune the average was 51.29 g. and in line cos16 the average was 27.82 g. (Fig. 7). Use of nitrogenous fertilizer had a significant effect on the 100 kernel weight (Fig.8). By use of 50 and 100% of recommended among of chemical fertilizer the amount of this traits increased in the order of 17.19 and 21.03% percent rather than without using of chemical fertilizers. Use of chemical fertilizers cause an increase in photosynthesis and produce of assimilate and at last increase of the seed weight. Sharif *et al.,* (2006) got the conclusion according to their experiments that the presence of biological Azospirilium fertilizer causes increase in the 100 kernel weight in rice. Also Mohammadi *et al*., (2011) reported similar results in their experiments. According to Fig. 9, use of biological fertilizer has caused increase in 100 kernel weight up to 7.52%. Dhillon *et al*., (1980) mentioned the increase of 100 kernel weight with presence of biological fertilizers. These researchers mentioned the increase of 100 kernel weight in corn with presence of biological fertilizers. According to the results of experiments of Eydizadeh *et al.,* (2011), 100 kernel weight in corn increased in effect of biological fertilizers. Adholeya and prakash (2004) said that the 100 kernel weight will better in presence of biological fertilizer and compost.

Grain filling rate

Growing of seed as an economical yield has 2 levels of filling rate and filling period. The seed filling rate shows the collection of dry weight in time and during the linear growth of seed, and the dry weight collection during some levels shows its growth and usually by omitting the nonlinear data which shows delay levels are prepared. (Kafi *et al.*, 2001). Use of nitrogenous fertilizer in controlled treatment recommended up to 100% in both condition of use and without using of biologic fertilizer will increase the seed filling rate, but this increase in local Aqgune with 100% nitrogenous fertilizer along with inoculation in compare with non-inoculation statistically did not have a significant difference. Seed filling rate in Talash and local Aqgune with 50% of recommended nitrogenous fertilizer along with inoculation was less than the speed of less inoculation of 100% nitrogenous and without inoculation. But in the other cultivars were not observed any significant difference (Fig. 10).

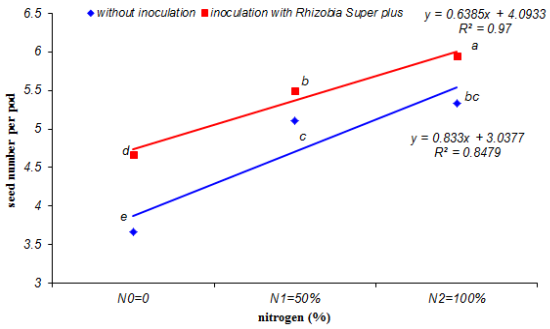

Fig. 1. The interaction between nitrogen fertilizer and organic manure on seed number per pod.

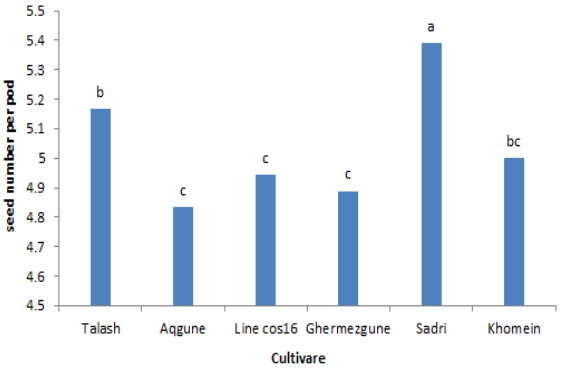

Fig. 2. The effect of cultivar of seed number per pod.

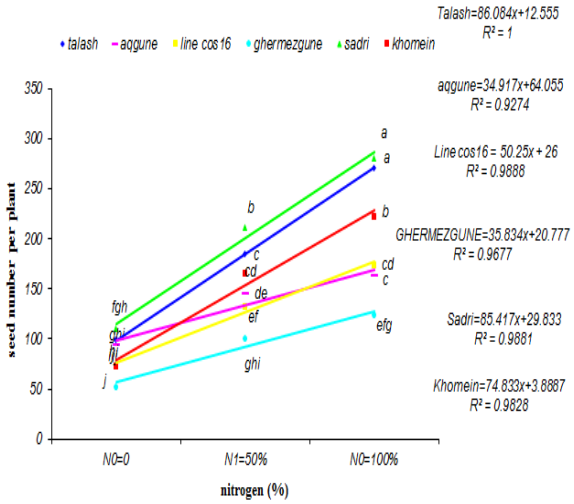

Fig. 3. The interaction between nitrogen fertilizer and cultivar on seed number per plant.

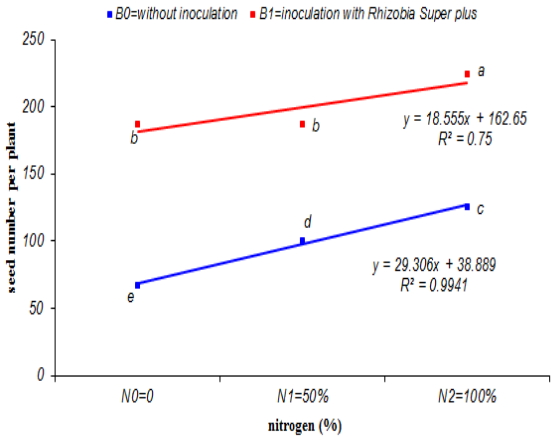

Fig. 4. The interaction between nitrogen fertilizer and organic manure on seed number per plant.

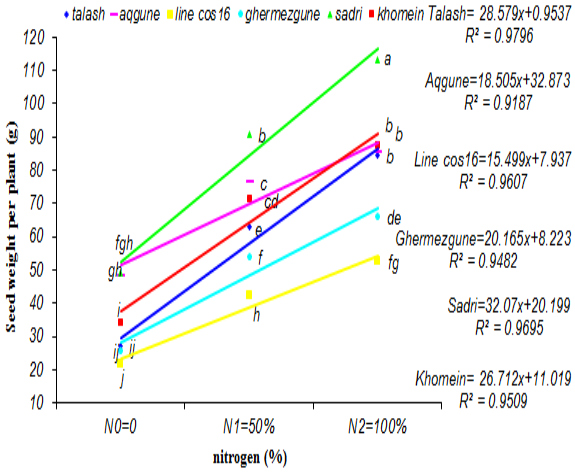

Fig. 5. The interaction between nitrogen fertilizer and cultivar on seed weight per plant.

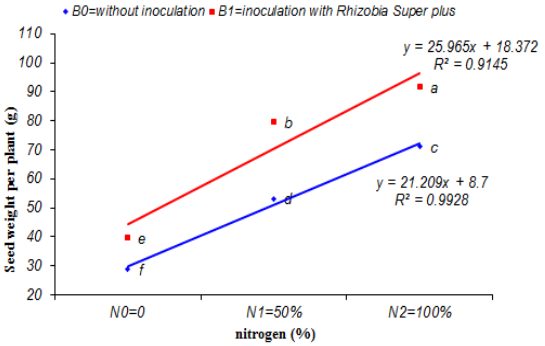

Fig. 6. The interaction between nitrogen fertilizer and organic manure on seed weight per plant.

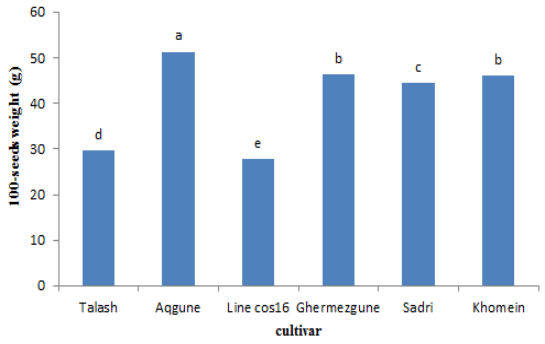

Fig. 7. The effect of cultivar on 100-seeds weight.

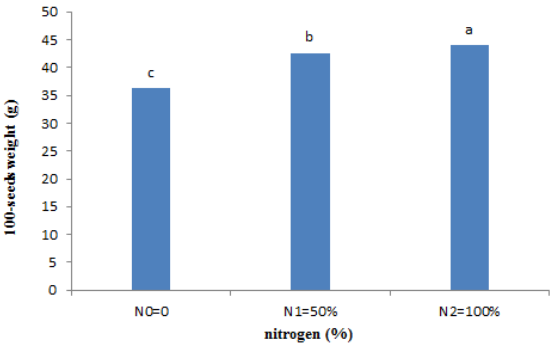

Fig. 8. The effect of nitrogen on 100-seeds weight.

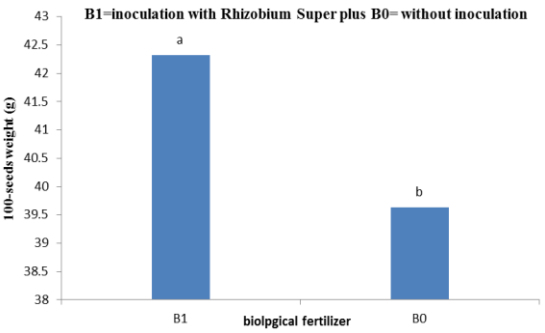

Fig. 9. The effect of biological fertilizer on 100-seeds weight.

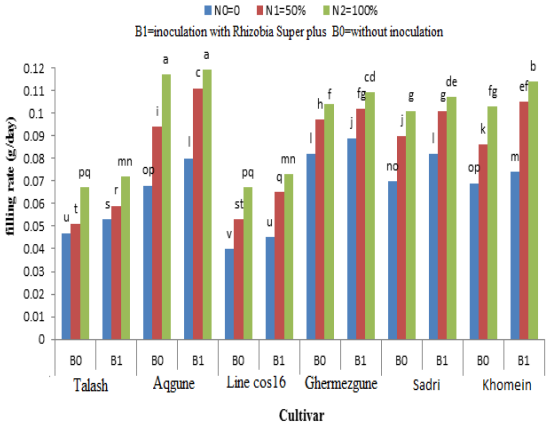

Fig. 10. The interaction of cultivar, nitrogen fertilizer and organic manure on filling rate.

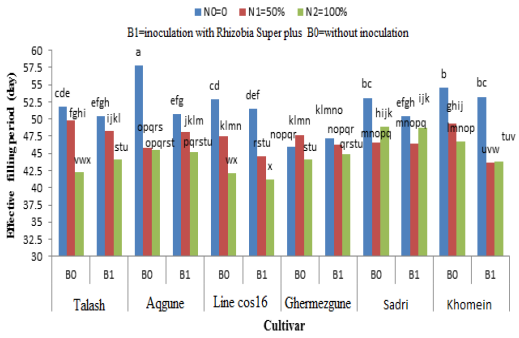

Fig. 11. The interaction of cultivar, nitrogen fertilizer and organic manure on effective filling period.

The effective filling period

The effective filling period is mostly used for evaluating of approximate length of filling period, and the growth of seed obtained through dividing the bean yield (kg/ha) to rate of the collected dry weight in seed (kg.day) via lineal step. Also about the single seed, we can calculate the effective filling period through dividing the weight of seed to its filling rate (Kafi *et al.*, 2001). Seed filling includes 3 steps: 1. slow growth (5 days after flowering) 2. Lineal increase (5-20 days after flowering) 3. Second slow growth (Chaubey and Singh, 1994). The results of mean comparison between the most effective period of filling in local Aqgune and khomein in condition of without inoculation and nitrogen fertilizer were 57.75 and 54.55 days. Use of biologic fertilizer with nitrogenous fertilizer has not significant effect in local khomein and Ghermez gune. Also use of nitrogenous fertilizer from controlled treatment was

recommended up to 100% in Talash and Line cos16 cultivars which led decrease of effective filling period. Use of biologic fertilizer with condition of not using nitrogenous fertilizer did not have significant effect on the filling period of Talash, local Ghermez gune, Line cos16 and khomein cultivars (Fig. 11).

Conclusion

The results showed that use of biological fertilizer increased the pinto bean grain production. In general we can say via use of biological fertilizers we can decrease the consume of chemical fertilizers, as the consumption of biological fertilizer along with half of the recommended chemical fertilizer did not have significant difference in the measured traits of 100% nitrogenous fertilizer with inoculation of Rhizobium. According to the results of experiments with consume of half of the recommended nitrogenous fertilizer along with the biological fertilizer, the seed weight is high with recommendation of 100% nitrogenous fertilizer, without use of biological fertilizer. so, use of biological fertilizer, not only decrease the use of chemical fertilizer and production costs and disadvantages for environment, but also can get a good yield in bean.

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