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Evaluation Use of Growth-Promoting Bacteria and Manure on Some Properties of Canola (*Brassica napus*)

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Abstract To study effect of bio fertilizers and manure on yield and yield components of Canola (*Brassica napus*)an experiment was conducted at research farm of University of Zabol, Iran in 2012 cropping season. Experimental treatments were arranged in a factorial design in randomized complete block design with three replications. Treatments included; three manure levels; (control, 20 and 30 t/ha) and tree levels of bio fertilizers, (control, *Azospirillum*, *Azotobacter* and *Azospyrilium+Azotobacter*). Based on the results of the different levels of manure had significant effect only on biological yield and other traits were not significant. Effect of bio fertilizers on plant height, number of pods per plant, seeds per pod, economic and biological yield was significant. Interaction among factors showed the highest plant height (154/3 cm) treated 20 t/ha and *Azospirillum* bio-fertilizer was mixed. The highest number of pods per plant was obtained of plus *Azotobacter* and *Azospirillum* bio-fertilizers mixed use of control manure (1280 pcs). The study results showed that the combined use of bio-fertilizers and livestock than individual consumers can increase their yield and yield components of canola.

Keywords Yield, Height Plant, Bio-fertilizer

1. Introduction

One of the problems of Iranian society today, the excessive increase of population. Regardless of population control measures, discussed increasing agricultural production should be a priority. Due to limited soil and water resources used in agriculture should be provided effective strategies to increase production per unit area. This is what happens in our country and the unbalanced use of chemical fertilizers and pesticides is uncommon[2]. Not produce an increase in the cost of environmental degradation and waste of resources such as soil and groundwater is over, it proved that conserve natural resources and the environment has increased production more economical. The climate, the soil is considered the most important component of the human environment. But unlike these two, the chemical composition of soil contamination is not easily measurable, unsuitable soil contamination, a phenomenon that ultimately a threat to human life cast[16].

The use of mineral fertilizers fastest way to supply nutrients needed by plants, but the high cost of chemical fertilizers, pollution, environmental degradation and soil is concerned. while the use of plant and animal resources development of renewable sources of biological and chemical resources can play an important role in Fertility and preserve biological activity, organic farm soil, ecological health system and raising the quality of agricultural products[27]. Bio-fertilizers contain a substance called micro organism that when the seed, root and soil used to stimulate and enhance plant growth[24]. It can be the most bacteria *Azospirillum, Azotobacter, Closridium*, Bacillus noted. Among these bacteria, *Azospirillum* and *Azotobacter* by the ability to communicate important crop plants such as maize, sorghum and wheat have attracted more attention.[14, 17]. Several mechanisms to explain how the bacteria growth increased growth and development plants known that the mechanisms to be basically consists of two groups of direct and indirect[26].

In direct mode, the mechanisms of PGPR nitrogen fixation, increased absorption and availability of nutrients or dissolved to produce plant growth hormones, enzymes modulate the synthesis of plant growth and development, manufacture vitamins and soluble phosphate to stimulate and increase the plant are[21]. In indirect mode, using different agonistic mechanisms to neutralize or mitigate harmful effects on plant disease and are thereby enhance plant growth. Competition to attract and occupy a position suitable material for pathogens, production of antibiotics, enzymes and reverse the hydrogen cyanide production (HCN) are the most important mechanisms used in this way. organic materials due to the effects of the constructive specifications physical and biological soil as one of the pillars of a power plant and soil fertility and organic fertilizers, known as the most important factor availability organic plant in the rhizo sphere[7, 12, 22]. Use compost and manure to increase soil

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organic matter, inorganic nutrients, improve soil structure and function leads[3]. Thus the use of organic fertilizers such as manure and compost improves soil physical properties and bulk density is reduced[9].

Yasari and Patwardhan (2007) Significant increase in total dry weight of the plant at different growth stages (rosette stage to maturity) in the inoculated treatments with growth-promoting bacteria strains (*Azotobacter and Azospirillium*) compared with non-inoculated reported[25]. Results of seed dry weight values obtained during the investigation showed that there were significant effects of inoculation with *Azospirillium brasilense*. Results of seed dry weight values obtained during the investigation showed that there were significant effects of inoculation with *Azospirillium.* The expression Shehata and El-Khawas (2003) application of bio-fertilizers increased growth yield and quality traits in sunflower compared with the control (non-inoculated) improved. So that the increase in seed yield, seed protein and oil content[20]. Mishra et al (1998) reported that the *Azospirillium* inoculation on corn yield and grain dry weight increased[14]. Shaalan (2005) showed that black seed inoculation with *Azotobacter* and *Azospirillium* and *Pseudomonas* biological fertilizers such as improved growth characteristics such as plant height, which is the main reason for the increased uptake of nutrients by plants[18]. Microbial inoculants also improve soil properties such as organic matter content and soil available nitrogen content is increased. with regard to the need for more research to be carried out in response to the application of the cultivated plants bacteria driver of growth and organic materials, as well as the possibility of replacing the chemical fertilizers, with the compounds in the experiment of bacteria. Vegetation stimulant properties of animal and plant canola and reviewed.

2. Materials and Methods

In this study, conducted in the Institute of Agriculture, University of Zabol, Zabol, run 35 km southeast of the city in 2012. Elevation of 481 meters above sea level in the region of 61 degrees 41 minutes' east longitude and 30 degrees 54 minutes north latitude located. Based on weather data, greatly warm, dry area with average annual rainfall of 63 mm and the average maximum temperature is about 23℃ and 49℃ for at least - 7℃. Before planting to determine the chemical and physical properties of soil samples were 0- 30 cm bottom and determine the sandy loam soil texture and pH is of the 8.1. Experiment factorial experiment in a randomized complete block design with three replications, was conducted on the plant canola. The first factor of manure at three levels: no fertilizer, 20 and 30 t/ha livestock and the second factor consisted of four levels bio-fertilizers includes control application (C), *Azospirillium* (Azo), *Azotobacter* (Aze) and mixed *Azospirillium* and *Azotobacter* (Azo + Aze) were considered. Include land preparation operations plow, Disc, create plots of length 2/5 and a width of 2 meters, the addition of nitrogen-based fertilizers, and manure phosphorus and potassium ratio in October was 50:100:100. Sowing seeds by hand and then seed inoculation with bio-fertilizers with a density of 33 p/m^2 (distance between rows and between plants 10 to 30 cm) were planted and watered immediately. Characteristics studied in this research, including plant height, number of branches, number of pods per plant, seeds per pod, seed weight, biological yield and harvest index and economic yield. Rape traits measured at the end of the growing season in May when the seeds were in maturity stage. So the middle two rows of each plot was sampled and then eliminate the effects of a wide margin m^2 done bush at the height of the collar end to stem the most with meters. Height of the crown to the ends of the stems with was measured. Continue working on the ground and cut bushes were coded and then transferred to the laboratory. After drying in an oven at 74℃ for 48 hr and dry weight was measured.

2.1. Height Plant

In the present experiment, meaning plant biomass was affected by bio fertilizer treatment (Table 1). Also significant interaction between treatment Bio-fertilizers and animal manure was observed (Table 1).

The use of bio-fertilizers increased plant height and maximum height plant (150/1 cm) in *Azospirillum* treatment, which was significantly different from other treatments (Table 2). plant growth by stimulating growth (PGPR) with molecular nitrogen fixation weather control activities pathogenic fungi produce and secrete growth regulators, producing vitamins help to release phosphorus, potassium, nitrogen and micronutrients consumption soil done. Comparison showed that the treatment effect of manure and bio fertilizers plus manure 20 t/ha *Azospirillum* maximum plant height (154.3 cm) and the highest (Table 3). the reasons of can be used for fertilizers at the height of the increasing influence of biological bushes counted on the use of fertilizers led to an increase in the between the nodes that this could be related to the plant hormones produced by the fertilizers[8]. Shaalan (2005) also showed that black seed inoculation with bio fertilizers like *Azotobacter*, *Azospirillum* and Pseudomonas improved growth characteristics such as plant height, which is the main reason for the increased uptake of nutrients by plants[19].

Table 1. Analysis of variance for economical yield and agronomy index of Canola (*Brassica napus*) in manure and bio-fertilizers treatments

		MS							
S.O.V	df	plant High	Number of branches	Number of pods per plant	Number of pods per plant	Weight of 1000seed	Economical yield	Biological yield	Harvest index
Replication	2	148.3^{*}	1^{ns}	2965.4^{ns}	27.7^{*}	0.027^{ns}	0.04^{ns}	4.03^{ns}	33.29^{ns}
Manure (A)	2	34.3^{ns}	0.08^{ns}	29888.6^{ns}	19.0^{ns}	0.58^{ns}	0.24^{ns}	11.01^{**}	40.9^{ns}
Bio fertilizers (B)	3	1333.2^{**}	0.07^{ns}	1202701.8^{**}	31.6^{*}	0.07^{ns}	2.07^{**}	26.06^{**}	5.7^{ns}
(A×B)	6	298.6^{**}	0.49^{ns}	101737.1^{**}	8.1^{ns}	0.49^{ns}	0.30^{ns}	0.92^{ns}	24.6^{ns}
Error	22	42.17	0.93	15013.9	7.14	0.93	0.17	1.41	18.84
C.V (%)	-	4.6	18.1	19.5	10.4	14.5	11.2	8.66	16.1

ns: Non significant

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

Table 2. Means comparison of economical yield and agronomical index of Canola (*Brassica napus)* in manure and bio-fertilizers treatments

Treatments	High plant (cm)	Number of branches	Number of pods per plant	Number of pods per plant	Weight of 1000 seed	economical yield (th^{-1})	Biological yield	Harvest index
Organic Manure								
Control	140.4a	5.41a	683.2a	25.1ab	3.75a	3.53a	14.17a	25.2a
20 ton/h^{-1}	137.6a	5.25a	589.0a	24.5b	3.47a	3.81a	14.41a	26.5a
30 ton/h^{-1}	137.3a	5.33a	607.6a	27.0a	3.31a	3.63a	12.6b	28.9a
$LSD_{0.05}$	1.20	0.57	0.14	1.720	4.23	78.09	153.3	0.14
Bio-fertilizer								
Control	121.3c	5.4a	189.2d	23.0b	3.25b	3.13c	11.48c	27.4a
Azt	141.4b	5.3a	468.5c	25.4a	3.44ab	3.54bc	13.53b	26.0a
Azo	150.1a	5.3a	864.1b	26.6a	3.55ab	3.79b	14.47ab	26.3a
Azo+Azt	141.0b	5.2a	984.6a	27.2a	3.8a	4.26a	15.4a	27.7a
$LSD_{0.05}$	6.3	0.94	119.8	2.61	0.49	0.40	1.16	4.24

Means in each column follow by similar letter(s) are not significantly different at 5% probability level, using Duncan s Multiple Range Test

Table 3. Means comparison of economical yield and agronomical index of Canola (*Brassica napus*) in interaction effects of manure and bio-fertilizers treatments

Manure	Bio-fertilizer	High plant (cm)	Number of pods per plant
0 ton/h^{-1}	**0**	109f	67h
	Azt	145ab	386efg
	Azo	154a	1000b
	Azo+Azt	153.6a	1280a
20 ton/h^{-1}	**0**	125.6e	191.6gh
	Azt	139.6bcd	425.6ef
	Azo	154.3a	852.6bc
	Azo+Azt	131cde	886bc
30 ton/h^{-1}	**0**	129.3de	309fg
	Azt	139.6bcd	594de
	Azo	142bc	739.6cd
	Azo+Azt	138.3bcd	788bcd

Means in each column follow by similar letter(s) are not significantly different at 5% probability level, using Duncan's Multiple Range Test. Azo: Azospirillium, Azt: Azotobacter, Azo+Azt: Azospirillium, + Azotobacter

2.2. Number of Pods Per Plant

Bio fertilizer had a significant effect on number of pods per plant (Table 1). *Azospirillium* and *Azotobacter* treatments using the most The Pod per plant (984.6) was obtained (Table 2). Interaction of different treatments on the number of pods per plant, manure and bio fertilizers, means was significant (Table 1) and the highest number of pods per plant, combined with the use of biofertilizers and lack of manure *Azospirillium* and *Azotobacter* (1280) that significant differences were obtained with the other treatments (Table 3). Number of pods per plant and seed yield of canola is dependent on the post-anthesis, with a reduction in leaf area per plant, pods have a role in plant photosynthesis. Elikaee and Emam (2003) high influence on canola can help light the positive relationship between the light received per flower and turn it into a pod, number of pods per plant is increased, and number of pods per plant is increased[6]. Shakeri et al (2012) stated that application of biological fertilizer significant increase in the level of one percent of the number of capsules in sesame[19].

2.3. Number of Seeds in the Pod

Analysis of variance indicated that the bio-fertilizer had no significant effect on the number of seeds in the pod (Table 1). Comparison of means showed that the levels of animal manure, the highest number of seeds per pod (27/0) were obtained from 30 t/ha (Table 2). Application of bio fertilizers showed that the highest number of seeds per pod (27/2) in the combined treatment of *Azospirillium* and *Azotobacter* obtained (Table 2). Interaction between manure and bio fertilizers on seed was not significant (Table 1).

2.4. Weight of 1000 Seeds

Analysis of variance indicated that the effect of bio fertilizers, manure and interaction on grain weight did not show significant differences (Table 1). Comparison showed that the levels of animal manure, the highest value (3/75 g) was obtained without the use of manure and other treatments showed no significant difference (Table 2). Bio fertilizer application on mean grain weight showed the highest value (3/8 g) was treated with a combination *Azospirillium* and *Azotobacter* (Table 2). Have reported a significant increase in grain weight (2.92%) in addition to *Azotobacter* and *Azospirillum* bio fertilizer treatments, compared to control.

2.5. Economic al Yield

The results of the variance showed that the effect of the various treatments animal manure not significant but bio fertilizer on economic yield was significant different, (Table 1). Maximum economic yield (3/81 t/ha) of treated manure application of 20 t/ha obtained showed no significant difference with other treatments. Lowest economic performance (3/53 t/ha) of the controls (Table 2). Research results[1] in the Effect of manure on yield and quality of cumin also showed that the number of umbels per plant, number of seeds per plant, biological yield and economic performance are significantly affected by manure livestock increased. There was also a significant difference between the levels of bio fertilizer treatment. Maximum economic yield (4/26 t/ha) from the treatments to *Azospirillium* and *Azotobacter* was obtained (Table 2). This relationship can be stated that the combination of the follow *Azospirillium* and *Azotobacter* bacteria produce vitamin stabilizer through nitrogenase, growth stimulators, root growth and nutrient absorption and increase speed improves overall economic performance. Jahan et al (2010) Stated that the application of manure and bio-fertilizers can improve the performance and quality characteristics of pumpkin skin paper[10]. Majidiyan et al (2008) Stated that the combined use of chemical fertilizers and manure, in addition to reducing the use of chemical fertilizers and improve soil physical and chemical properties, a better yield was obtained[13].

2.6. Biological Yield

The results of the analysis of variance showed significant effect treatments with manure and bio fertilizers on biological yield (Table 1). In this regard it can be said, perhaps adding manure to the soil by improving soil physical and vital processes, also creating a favorable environment for root growth and the availability of nutrients needed for the plant, and its subsequent enhance the growth of providing dry matter production of to. Also, comparing bio-fertilizer treatments showed no significant difference between them, so that in BM *Azospirillium* and *Azotobacter* inoculated seed composition (15/4) compared to control 34/1 per cent (Table 2). I *Azospirillium* and *Azotobacter* expected composition of the body to absorb more nitrogen increased vegetative growth and dry matter production per plant, grain yield was improved bio because it provides.

2.7. Harvest Index

The results of the analysis of variance showed no significant difference between treatments in terms of bio fertilizers, manure and interaction on harvest index (Table 1). The highest harvest index (28/9 percent) were treated with 30 t/ha. The highest harvest index in the application of bio-fertilizer treatment combination of *Azospirillum* and *Azotobacter* (27/7 per cent) was obtained. Since a significant positive correlation between the number of pods per plant, harvest index, number of branches, plant height and grain yield is thus increased retention of nitrogen fixation and nitrogen stabilization by bacteria (*Azotobacter* and *Azospirillum*) in rape increased the root growth and ultimately contribute to the production of reproductive organs and the yield is higher.

3. Discussion

Nutritional management is one of the most important factors in the successful cultivation of a plant. Because fertilizers can also affect plant performance characteristics of quantitative and qualitative performance characteristics. In

this study it was found that the bio-fertilizers, animal manures and observed a significant increase in plant height, number of pods per plant, seeds per pod in oilseed rape plants (Table 1). On the other hand, results have shown that bio-fertilizers significantly increased the economic and biological yield per unit area (Table 1). Bio fertilizers to improve access to food due to increased growth and yield of rapeseed plant, which is relative to the control and manure. Several studies have shown that the biological fertilizers, in addition to attract food elements in the plant, with biosynthesis plant hormones, plant pathogens, as well as some other mechanisms to improve the performance of the plant and growth[4]. Kalra (2003) is that this rhizo sphere bacteria through the fixation of atmospheric nitrogen, increasing the availability of food elements in the rhizosphere roots, raising the level of contact, growth and improve useful biological, with the host plant in various stages of increasing growth and development[11]. *Azotobacter* is able to produce antifungal compounds against plant diseases and encourage germination and seedling vigor and improved plant growth is the result[5]. Nitrogen- fixing *Azospirillum* addition, the production of growth-promoting substances that can improve plant growth and yield[23]. Organic matter due to the effects of physical and biological soil properties that are constructive, as a component of plant nutrition and soil fertility are known. Organic fertilizers are the most important factor is the availability of organic matter in the rhizo sphere[23]. Animal manure and compost addition to the role of nutrition, improve product quality, increase soil biological activity and physical properties have a significant effect. In addition to adding organic fertilizers supply nutrients and improve soil physical properties and provide favorable conditions for root growth[15] and provides adequate space for root development. However, individually or in combination with bio-fertilizers and animal manure on crop yield and quality of rapeseed oil had a significant positive impact. The substitution of bio-fertilizers instead of chemical fertilizers can reduce environmental pollution and take steps toward sustainable agriculture. So perhaps to achieve high performance in the area of seed inoculation with *Azospirillum* and *Azotobacter* be recommended before planting.

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