

Full Length Research Paper

The effect of *Bacillus* sp. OSU-142 inoculation at various levels of nitrogen fertilization on growth, tuber distribution and yield of potato (*Solanum tuberosum* L.)

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The objective of this study was to evaluate the yield response of potato to inoculation with *Bacillus* sp. OSU-142 at three levels of N fertilization (0, 120, and 240 kg ha⁻¹) under field conditions in Ahlat District of Eastern Anatolia Region of Turkey in 2006 and 2007. Tuber inoculation with *Bacillus* sp. OSU-142 significantly affected yield and yield components in both years. Tuber yields and yield components were higher at all levels of nitrogen fertilizer in the inoculated plots as compared to the control. However, beneficial effect of *Bacillus* sp. OSU-142 on tuber yield was noted at 120 kg N ha⁻¹, possibly indicating either more effective of inoculation in the low-N input agriculture or an interaction of *Bacillus* sp.OSU-142 with higher yielding seasonal conditions. In general, more response to inoculation was observed in the absence of major crop growth limitations, suggesting the complementary contribution of the *Bacillus* sp.OSU-142 treatment to more efficiently developing higher yielding potato.

Key words: *Bacillus* sp., PGPR, N fertilizer, potato, yield.

INTRODUCTION

Potato (*Solanum tuberosum* L.) is an annual herbaceous plant that produces a tuber rich in starch. It is the world's fourth most important food crop, after maize, wheat and rice, with annual production approaching 321 million tone from a production area of 19 million ha (FAO, 2008). Potato cultivation has been officially encouraged in Turkey since 1872, and today the country is the Middle East's biggest producer after Iran, with production in 2007 of almost 4.3 million tones from 158.500 ha as average yield of 27 t ha⁻¹ (FAO, 2008). Although the potato production has mainly been performed in Central Anatolian region, accounting for nearly half of the national potato area, the crop is well adapted to the different climatic zones in Turkey, including Eastern Anatolia region. The aim of potato producers has got higher tuber yields. There are many strategies to get higher tuber yields in potato. One of them is nitrogen fertilizers. The role of N fertilization for tuber growth, development, yield and quality of potato is well known (Morena et al., 1994; Joern and Vitosh, 1995; Kumar et al., 2007). Inadequate N

fertilization leads to poor crop growth and yield, while excessive application leads to delayed maturity, poor tuber quality, excessive nitrate leaching and occasionally reduction in tuber yield (Harris, 1992; Lauer, 1986; Westermann et al., 1988). For this purpose, the efficiency of N fertilizer must be known to successfully apply N fertilizer according to crop growth needs. Due to the consumers concern of excessive use of chemical fertilizers for avoiding deleterious effects on the environment, there is increasing interest in the use of biofertilizer such as Plant Growth-Promoting Rhizo-bacteria (PGPR) because of their positive effects on soil conditions, nutrient availability, crop growth and yields for sustainable farming (Glick, 1995; Döbereiner, 1997; Vance, 1997).

PGPR are microorganisms living in the rhizosphere of cultivated crops with known plant growth promoting effects. Their introduction into cropping systems could contribute to better crop productivity. The previous studies indicated that PGPR could stimulate growth and increase yield in sugar beet and barley (Çakmakçı et al., 2001, 2006), soybean (Zhang et al., 1997), maize (Wu et al., 2005; Shaharoon et al., 2006), rice (Cong et al., 2009), peanut (Dey et al., 2004), and wheat (Zorita and

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Table 1. Monthly meteorological parameters during the crop seasons (growing periods: May to September).

Month	2006					2007				
	Monthly mean air temperature (°C)			Monthly rainfall (mm)	Monthly BSS ^a (day)	Monthly mean air temperature (°C)			Monthly rainfall (mm)	Monthly BSS ^a (day)
	Max.	Min.	Mean			Max.	Min.	Mean		
May	16.8	6.7	11.8	56.5	14	18.2	9.4	13.8	30.6	18
June	26.4	12.2	19.3	-	23	23.1	12.1	17.6	35.3	24
July	27.4	15.2	21.3	29.1	26	25.7	15.0	20.4	20.7	27
August	29.9	16.8	23.4	3.2	24	26.8	15.4	21.1	8.3	22
September	24.0	11.6	17.8	3.0	27	25.0	12.6	18.8	14.1	27
Total/mean			18.7	91.8				18.3	109.0	

Source: Experimental meteorological station near the research farm. ^a BSS : Bright Sun Shine days.

Canigia, 2009). The effects of PGPR on plant growth and productivity are either direct (biological N₂ fixation, P solubilization, increasing nutrient availability, etc.) or indirect catalytic actions. Biological N₂ fixation provides a major source of nitrogen for plants as part of environmental friendly agricultural practices. Biofertilizers including microorganisms may add nitrogen to the soil by symbiotic or asymbiotic N₂ fixation. The symbiotic fixation of N₂ through inoculation of legume crops with effective rhizobia is well known (Döbereiner, 1997; Vance, 1997). However, asymbiotic N₂-fixing bacteria, which live in the rhizosphere (Döbereiner, 1997) and/or endophytically (Hecht-Buchholz, 1998) is important for plant nutrition by increasing N uptake by the plants, and playing a significant role as PGPR in the bio-fertilization of crops (Çakmakçı et al., 2006). *Bacillus* OSU-142 is able to fix N₂ asymbiotically and growth promoting agent providing varied contributions to the enhancement of growth and productivity in many agricultural crop species (Çakmakçı et al., 2001, 2006; Eşitken et al., 2002; Orhan et al., 2006). However, information about the effects of *Bacillus* sp. OSU-142 inoculation on the growth and yield of potato has not yet been reported.

The aim of this study was to determine the effect of inoculation with *Bacillus* sp. OSU-142 on growth, tuber distribution and yield of potato at various levels of nitrogen fertilization under field conditions, and to test the hypothesis if *Bacillus* sp. OSU-142 can use an important potential as biofertilizer for potato production.

MATERIALS AND METHODS

Field experiments were conducted on a farm located in Ahlat District (38° 46'N and 42°30'E with an altitude of 1722 m), Eastern Anatolia region of Turkey in 2006 and 2007.

The experimental soil was a silt-clay-loam with organic matter content of 2.4 and 2.9% and with 4.5 and 4.3% lime content (pH = 7.7 and 7.6) in 2006 and 2007, respectively. Available P₂O₅ (Spectrophotometrically), K₂O (ammonium acetate method) and total nitrogen (Kjeldahl method) contents were 8.12 and 10.20 ppm, 2076.71 and 2057.61 ppm and 0.13 and 0.17% in 2006 and 2007, respectively. In 2006-2007 and 2007-2008 total rainfall during growing season (from May to September) was 91.8 and 109.0 mm,

respectively, and the average temperature for the same period was 18.7 and 18.3 °C, respectively (Table 1). These values indicated that hot and dry conditions prevailed in 2006.

The bacterial strain used in the study was *Bacillus* OSU-142 (N₂-fixing bacterium) that was kindly obtained from Atatürk University, Department of Plant Protection. *Bacillus* OSU-142 was reported as plant growth promoting bacteria, and it may well be suited alone or in combination to achieve sustainable and ecological agricultural production (Çakmakçı et al., 2001; Orhan et al., 2006). The bacterial strain was maintained for long-term storage in nutrient broth (NB) with 15% glycerol at -80°C. For this study, the bacterial strain was incubated on nutrient agar. A single colony was transferred to 1000 ml flask containing NB, and incubated aerobically in flasks, on a rotating shaker (150 rpm) overnight at 27°C. The bacterial suspension was then diluted with sterile distilled water to a final concentration of 10⁹ CFU ml⁻¹. Seed potato tubers were treated with the bacterial suspensions for 30 min.

In the study, "Melody" variety of potato, which had intermediate maturity, high marketable yields, oval tuber shape with light yellow flesh and excellent long term storability, was used as plant material. The experiments were carried out in a factorial design, using two different inoculations (control, *Bacillus* sp. OSU-142) and three levels of N fertilization (0, 120, 240 kg ha⁻¹) with three replicates. Potato plots were prepared in spring of each year when the soil was plowed to a depth of approximately 25 cm, disked to 15 cm depth, and harrowed to 10 cm depth. Half of the N (as per treatment), full P (100 kg P₂O₅ ha⁻¹) and K (80 kg K₂O ha⁻¹) were applied at planting. The remaining half of the N was applied at the time of hilling when the plants were about 15-25 cm high. N was applied through ammonium sulfate at planting and through urea at hilling. P and K were applied through triple super phosphate and potassium phosphate, respectively. The experimental crop was planted on 25 May, 2006 and 15 May, 2007. Well-sprouted tubers (50-60 g seed weight) were hand planted in furrows 10 cm depth at 70 cm inter row spacing and 35 cm intra row spacing in four rows. The experiment was conducted in irrigated conditions and it carried out a sprinkler irrigation system as required. All plots were ridged three times at an interval of 15 to 20 days for removing the weeds and covering the growing tubers. Plots were harvested by hand middle two rows excluding side rows and 1 m from each end of plots on 28 September, 2006 and 1 October, 2007. The investigated parameters were recorded from ten randomly selected potato plants from each plot. The observations on growth parameters such as plant height and stem number were also recorded at 50-55 days after planting. Tubers were weighed, counted and graded to assess tuber yield relevant for processing. Size distribution was determined to tuber diameter. Undersized tubers were determined in less than 35 mm diameter (<35 mm), and small-sized tubers were determined as between 35 and 50 mm diameter (35-50 mm), large-

Table 2. Growth and tuber characters of potato in response to inoculation with *Bacillus* sp. OSU-142 at various levels of N fertilization (2006 and 2007¹).

Variable	Plant height (cm)		Stems per plant		Tuber number per plant		Av. tuber weight (g)	
	2006	2007	2006	2007	2006	2007	2006	2007
Bacterium (B)								
Control	53.4	57.5 b	4.4	5.2 b	17.3 b	18.0	91.7	101.7 b
<i>Bacillus</i>	53.8	59.3 a	4.5	6.4 a	19.5 a	18.5	92.1	111.7 a
Nitrogen levels (N)								
0 kg N ha ⁻¹	39.6 c	50.0 b	3.2 b	5.5 b	15.4 c	14.8 c	85.5 b	104.6 b
120 kg N ha ⁻¹	58.7 b	61.6 a	4.9 a	5.6 b	18.0 b	19.2 b	102.6 a	111.9 a
240 kg N ha ⁻¹	62.6 a	63.6 a	5.3 a	6.3 a	21.7 a	20.8 a	88.1 b	103.6 b
Bacterium (B) X Nitrogen levels (N)								
Control + N ₀	35.6 d	43.7 e	2.6 d	3.9 d	12.2 e	14.8	91.0 abc	89.7 e
Control + N ₁₂₀	60.6 a	63.7 ab	5.1 ab	5.1 c	16.9 d	18.5	101.0 a	110.5 bc
Control + N ₂₄₀	64.0 a	65.1 a	5.5 a	6.5 ab	22.8 a	20.7	83.1 bc	104.8 cd
<i>Bacillus</i> + N ₀	43.5 c	56.3 d	3.9 c	7.1 a	18.6 c	14.9	80.1 c	119.5 a
<i>Bacillus</i> + N ₁₂₀	56.7 b	59.6 c	4.6 b	6.1 b	19.1 c	19.8	103.2 a	113.3 ab
<i>Bacillus</i> + N ₂₄₀	61.2 a	62.0 b	5.1 ab	6.1 b	20.7 b	20.8	93.1 ab	102.3 d
CV ² (%)	3.41	2.87	8.77	8.42	3.01	4.43	7.17	3.39
F values								
B	0.22	12.38**	0.52	29.19**	69.69**	1.64	0.02	34.55**
N	272.72**	269.34**	46.47**	4.77*	198.57**	84.86**	10.95**	9.52**
B x N	19.16**	110.19**	9.52**	19.33**	89.20**	1.32	3.85*	34.47**

¹Means followed by different letter within each column are significantly different. F values marked with *and** are significant at 0.05 and 0.01 levels, respectively.

²Coefficient of variation.

sized tubers were determined with at least a 50 mm diameter (>50mm). Data were evaluated by analysis of variance using SAS software (SAS Institute, Cary, NC, USA). When "year x treatment" interactions were significant, data were evaluated separately according to years, and means were presented. Duncan's Multiple Range Test was used to determine the differences among the "bacterium x nitrogen levels" combinations (p<0.05) using MSTAT-C software.

RESULTS AND DISCUSSION

Analysis of variance showed statistically significant differences between years for the characteristics investigated (Tables 2, 3 and 4). More favorable climatic conditions increased growth and yield of potato in 2007. Especially, a higher number of sun shine days during 2007 season coincided with the tuber initiation and the establishment of the crop canopy. Growth of canopy is known to be hastened by a high amount of irradiance which is primarily due to the effects on axillary branch formation (Salisbury and Ross, 1992; Wheeler, 2006). Thus, favorable conditions during haulm growth in 2007-2008 resulted in a large haulm surface (higher stem number and higher plant height), and maintained tuber growth at a higher rate for a longer time period leading to higher yield (Salisbury and Ross, 1992; Kumar et al., 2007).

Growth and tuber characters

Two years of trials under field conditions showed that treatments including bacterial inoculation and N fertilization levels significantly affected the parameters investigated depending on the year. *Bacillus* sp. OSU-142 applications significantly increased plant height, stem number per plant and average tuber weight in 2007, although the effect of bacterial inoculation on total tuber number per plant and tuber distribution was not significant (Tables 2 and 3).

Plant growth parameters were higher and more responsive to inoculation with *Bacillus* sp. OSU-142 in 2007 than in 2006 (Table 2). The lower positive effects of bacterial inoculation on growth parameters may be associated with seasonal influences such as weather conditions and soil fertility. Positive responses to inoculation on crop productivity depend on soil type and environmental conditions (Çakmakçı et al., 2001, 2006). The maximum temperature was considerably higher in 2006 than in 2007 (Table 1). A high day temperature is usually associated with a high rate of respiration and transpiration, leading to moisture stress even when soil moisture content is high. Rate of net photosynthesis has been reported to decrease when day temperature is higher than 25°C (Horton, 1987; Salisbury and Ross, 1992). The plant photosynthesis has been also reported to

Table 3. Tuber number per plant by size class of potato in response to inoculation with *Bacillus* sp. OSU-142 at various levels of N fertilization (2006 and 2007¹).

Variable	Tuber number per plant					
	Undersized (<35 mm)		Small-sized (35-50 mm)		Large-sized (> 50 mm)	
	2006	2007	2006	2007	2006	2007
Bacterium (B)						
Control	3.2 b	6.4	6.5 b	4.9	7.5	6.8
<i>Bacillus</i>	4.1 a	6.8	7.5 a	5.0	7.9	6.8
Nitrogen levels (N)						
0 kg N ha ⁻¹	3.4 b	4.5 c	6.0 b	4.3 b	6.0 b	6.1 c
120 kg N ha ⁻¹	3.0 b	6.8 b	6.5 b	5.0 ab	8.5 a	7.5 a
240 kg N ha ⁻¹	4.5 a	8.5 a	8.6 a	5. a	8.7 a	6.8 b
Bacterium (B) X Nitrogen levels (N)						
Control + N ₀	2.3 c	4.8 d	5.1 c	4.1 c	4.9 c	5.9
Control + N ₁₂₀	2.7 bc	6.3 c	5.8 c	4.8 bc	8.3 a	7.5
Control + N ₂₄₀	4.7 a	8.0 b	8.7 a	5.8 a	9.3 a	6.9
<i>Bacillus</i> + N ₀	4.6 a	4.1 d	6.9 b	4.6 bc	7.1 b	6.3
<i>Bacillus</i> + N ₁₂₀	3.3 b	7.3 b	7.2 b	5.2 ab	8.6 a	7.5
<i>Bacillus</i> + N ₂₄₀	4.3 a	9.0 a	8.4 a	5.2 ab	8.1 ab	6.6
CV ² (%)	13.57	7.82	8.71	7.61	8.53	5.36
F values						
B	12.35**	3.69	11.49**	0.25	1.84	0.06
N	14.00**	92.64**	29.68**	15.35**	32.18**	22.85**
B x N	12.16**	6.10*	5.38*	4.18*	10.04**	1.26

¹Means followed by different letter within each column are significantly different. F values marked with * and ** are significant at 0.05 and 0.01 levels, respectively.

²Coefficient of variation.

to increase by PGPR application over a wide range of plant growth stages (Zhang et al., 1997). The growth promoting effects of PGPR strains are usually affected by temperature (Ahmed et al., 2008; Shahab and Ahmed, 2008), and increased plant leaf development and dry matter accumulation at 16°C compared to 26°C in loamy sand in semi-continental climate (Egamberdiyeva and Höflich, 2003; Zhang et al., 1997).

In 2007, inoculation with *Bacillus* sp. OSU-142 increased plant height by 3.1%, stem number per plant by 23.1% and average tuber weight by 9.8%, compared to the Control+N₀ application (Tables 2 and 3). However, the significant effect of inoculation with *Bacillus* sp. OSU-142 was found for total tuber number per plant and tuber distribution (except large-sized tuber number per plant) in 2006 only. In the growth season of 2006 significant increases were obtained from total tuber number, tuber numbers of undersized and small-sized per plant by 12.7, 28.1 and 15.4%, respectively, compared to the control. However, the no significant effect of inoculation with *Bacillus* sp. OSU-142 was found for large-sized tuber number per plant in both years. It may be related to the higher vine growth than in tuber set of potato with inoculated *Bacillus* sp. OSU-142 (Table 2). The beneficial effects of PGPR on growth of different crops such as maize, sugar beet and barley were also observed in

previous studies, and similar results have been reported in both greenhouse and field conditions (Çakmakçı et al., 2006; Wu et al., 2005; Shaharoon et al., 2006). The results are in agreement with those researchers and Çakmakçı et al. (2001) who reported that the bacterial inoculation could be increased marginally when tested under ideal climatic situations.

Analysis of variance showed that growth and tuber parameters were significantly influenced by N levels (Tables 2 and 3). In the study, the highest plant height and stem number per plant were obtained with the highest N level, i.e. 240 kg N ha⁻¹ in both years. The growth parameters of potato were significantly affected by various levels of nitrogen, and the highest values under the highest level of N were obtained due to the fact that plants received more nitrogen, which might have encouraged more vegetative growth as previously reported by Singh and Raghar (2000), Shakh et al. (2001), Kushwah (1989), Rai et al. (2002), Kumar et al. (2007) and Sincik et al. (2008). Nitrogen fertilization also increased total tuber number per plant (Shakh et al., 2001; Sincik et al., 2008). However, both positive and negative effects of N application on tuber number per plant have been reported (Morena et al., 1994, Belanger et al., 2002; Love et al., 2005; Kumar et al., 2007). Nitrogen fertilization increased average tuber weight

significantly over control at 120 kg N ha⁻¹ whereas tuber weight decreased in 240 kg N ha⁻¹ in both years. Zvomuya et al. (2002), Knowles and Knowles (2006) and Kumar et al. (2007) reported that vine growth was favored over the tuber growth at high N level, and average tuber weight varied with tuber number per plant. These data were confirmed by present results showing that there was a trend for reduced average tuber weight due to higher amounts of undersized tuber number per plant at 240 kg N ha⁻¹ (Tables 2 and 3).

The interaction between bacterial inoculation and nitrogen fertilization was significant for growth and tuber parameters (except total and large tuber number per plant in 2007) investigated in both years. When *Bacillus* sp. OSU-142 and N fertilization levels were combined, their effect on parameters was generally higher than those of N fertilizer. Nitrogen fertilization increased plant height significantly at all levels of N with or without *Bacillus* sp. OSU-142, while the tallest plants were recorded for Control+N₂₄₀ combined plot in both years. In the study, stem number per plant was higher at all levels of N in the inoculated plots with *Bacillus* sp. OSU-142 compared with N fertilization, while the highest stem number per plant was recorded for *Bacillus* sp. OSU-142+N₀ combined plot in 2007 (Table 2). In the absence of any fertilizer application, significant effect of *Bacillus* sp. OSU-142 on stem number was attributed to well soil fertility status of experimental site which had the higher organic matter content (2.9%) and total N content (0.17%) as well as favorable climatic conditions. In general, it is known that the number, diversity and activity of soil organism are influenced by soil organic matter properties (Kobabe et al., 2004). Also the growth and metabolic activity of soil microorganisms are limited by the availability of nutrients (Welbaum et al., 2004) and mineral nutritional factors can affect the number of bacteria in the rhizosphere (Marschner et al., 1999). Similarly, Çakmakçı et al. (2006) reported that the growth promotion of sugar beet with tested PGPR strains strongly depends on soil organic matter content and the sugar beet from the high-OM soil had significantly higher yields. Also, Çakmakçı et al. (2001) reported the N₂-fixing bacteria in the absence of any fertilizer application achieved root yields equal to the nitrogen treatment at the level of 100 kg N ha⁻¹ in sugar beet. The results are in agreement with those researchers. In the study, the interaction of bacterial inoculation and N fertilization level was significant on total tuber number per plant in 2006 only. The inoculation with *Bacillus* sp. OSU-142 significantly increased total tuber number per plant compared with N fertilization alone, while the highest total tuber number per plant 22.8 was recorded for Control+N₂₄₀ combined plots (Table 2). In 2006, the highest average tuber weight was obtained from *Bacillus* sp. OSU-142+N₁₂₀ combined plots, followed by Control+N₁₂₀ combined plots, while the highest values were recorded for *Bacillus* sp. OSU-142+N₀ combined

plots in 2007 (Table 2). At 0 and 120 kg N ha⁻¹ levels, the response of average tuber weight of potato to bacterial inoculation in general was higher than with 240 kg N ha⁻¹ application. As mentioned above, these increases could be related to the higher vine growth and the lower tuber number per plant at 0 and 120 kg N ha⁻¹ levels.

The interaction between nitrogen fertilization and bacterial inoculation was significant for numbers of tuber per plant in different size classes (except large tuber number per plant in 2007) investigated in both years (Table 3). *Bacillus* sp. OSU-142 and nitrogen combinations increased numbers of undersized and small per plant tuber significantly as compared to Control+N₀ application. The highest numbers of undersized and small per plant were obtained from the *Bacillus* sp. OSU-142+N₂₄₀ and Control+N₂₄₀ combinations depending on the year.

Total and graded tuber yields

Two years of trials under field conditions showed that bacterial inoculation significantly affected total and graded tuber yields in potato (Table 4). Bacterial inoculation mainly affected marketable small tuber yield (tubers between 35 and 50 mm diameter). Inoculation with *Bacillus* sp. OSU-142 significantly increased small tuber yield by 34.7 and 11.8% in 2006 and 2007, respectively, compared with control, while its effect on unmarketable (tubers less than 35 mm diameter) and large tuber yield (tubers larger than 50 mm diameter) varied according to years. In the study, the results showed that the inoculation with *Bacillus* sp. OSU-142 had the capacity to increase total tuber yield (+17.8 and +21.3 t ha⁻¹ in 2006 and 2007, respectively) as compared to the Control+N₀ combined plot. This is the first study to demonstrate that *Bacillus* sp. OSU-142 inoculation can increase yield and growth of potato. Similar findings were reported in the previous studies showing that inoculation with *Bacillus* sp. OSU-142 and other bacterial strains can stimulate yield and growth parameters in sugar beet and barley (Çakmakçı et al., 2001, 2006), wheat (Zorita and Canigia, 2009), sweet potato (Yasmin et al., 2007), maize (Chela et al., 1993; Wu et al., 2005), tomato (Mena-Violante et al., 2007), apple (Karlıdağ et al., 2007) and rice (Cong et al., 2009).

Analysis of variance showed that total and graded tuber yields were significantly influenced by N levels in both years (Table 4). The highest yields of unmarketable and marketable small tubers were obtained with the highest N level, that is, 240 kg N ha⁻¹. In the study, no significant differences were determined between 120 and 240 kg N ha⁻¹ applications for large tuber and total tuber yields. Similar increases in yields of marketable and unmarketable tuber with increasing N levels have been reported in previous studies (Singh et al., 1997; Kumar et al., 2007) with a non-significant effect on the yield of large tubers

Table 4. Total tuber yield and yields by size class of potato in response to inoculation with *Bacillus* sp. OSU-142 at various levels of N fertilization (2006 and 2007¹).

Variable	Tuber yield (t ha ⁻¹)							
	Unmarketable (<35 mm)		Small (35-50 mm)		Large (> 50 mm)		Total	
	2006	2007	2006	2007	2006	2007	2006	2007
Bacterium (B)								
Control	2.4 b	4.8	17.6 b	14.4 b	53.7	67.4 b	73.6 b	86.6 b
<i>Bacillus</i>	3.7 a	4.7	23.7 a	16.1 a	57.0	75.4 a	84.3 a	96.3 a
Nitrogen levels (N)								
0 kg N ha ⁻¹	2.6 b	3.3 b	18.3 b	12.9 b	40.4 b	56.7 b	61.1 b	72.9 b
120 kg N ha ⁻¹	2.8 b	5.4 a	19.4 b	14.8 b	64.1 a	80.4 a	86.2 a	100.6 a
240 kg N ha ⁻¹	3.8 a	5.6 a	24.4 a	18.1 a	61.5 a	77.2 a	89.7 a	100.9 a
Bacterium (B) X Nitrogen levels (N)								
Control + N ₀	1.5 b	3.4	15.9 d	11.8	34.9 d	47.0 d	52.2 d	61.2 e
Control + N ₁₂₀	2.0 b	5.0	17.3 cd	13.2	60.5 ab	77.3 b	79.7 b	95.6 c
Control + N ₂₄₀	3.7 a	6.0	19.7 bc	18.1	65.6 ab	78.0 b	89.0 a	102.0 ab
<i>Bacillus</i> + N ₀	3.6 a	3.2	20.6 b	14.0	45.8 c	66.4 c	70.0 c	83.5 d
<i>Bacillus</i> + N ₁₂₀	3.6 a	5.7	21.4 b	16.4	67.7 a	83.5 a	92.7 a	105.6 a
<i>Bacillus</i> + N ₂₄₀	4.0 a	5.2	29.1 a	18.0	57.4 b	76.4 b	90.4 a	99.7 b
CV ² (%)	16.94	9.38	8.01	9.34	9.34	3.28	4.99	2.31
F values								
B	30.52 ^{**}	0.09	60.44 ^{**}	7.00 [*]	1.88	52.48 ^{**}	33.26 ^{**}	95.10 ^{**}
N	10.47 ^{**}	47.26 ^{**}	23.94 ^{**}	20.15 ^{**}	38.23 ^{**}	181.87 ^{**}	94.06 ^{**}	348.67 ^{**}
B x N	4.89 [*]	3.75	4.61 [*]	2.00	5.81 [*]	30.75 ^{**}	6.84 ^{**}	47.20 ^{**}

¹Means followed by different letter within each column are significantly different. F values marked with * and ** are significant at 0.05 and 0.01 levels, respectively.

²Coefficient of variation.

(Rykbost et al., 1993). Besides, significant increases in total tuber yield of potato as a result of N fertilization have been documented by Belanger et al. (2002), Kumar et al. (2007), Sincik et al. (2008) and Shillito et al. (2009). The results are in agreement with these researchers.

Interaction effect of bacterial inoculation and N level applications was significant on total tuber yield in both years (Table 4). In the absence of any fertilizer application, *Bacillus* sp. OSU-142 achieved comparable tuber yields to 120 kg N ha⁻¹ fertilization. The highest total tuber yields were obtained from *Bacillus* sp. OSU-142+N₁₂₀ combined plot, followed by *Bacillus* sp. OSU-142+N₂₄₀ combined plot in 2006, while the highest yield was recorded for *Bacillus* sp. OSU-142+N₁₂₀ combined plot in 2007. Significant effects of *Bacillus* sp. OSU-142 inoculation on large tuber yields were also obtained at 0 and 120 kg N ha⁻¹ fertilization, but not with 240 kg N ha⁻¹ fertilization. However, bacterial inoculation and N fertilization combinations increased unmarketable and small tuber yields significantly compared with control, while the highest yields were obtained from *Bacillus* sp. OSU-142+N₂₄₀ combined plot. The positive effects of *Bacillus* sp. OSU-142 on tuber yields in this study may suggest that potato can be more responded in N₂-fixing bacterial inoculation with N combinations than alone fertilizer usage under an N-deficient condition. Previous

studies indicated that inoculation with beneficial bacteria can increase the efficiency of fertilizer use at both high and low fertilization levels (Wu et al., 2005). Similarly, Shaharoon et al. (2006) reported that grain yields of maize were increased significantly by applying *P. fluorescens* biotype G (N₃); 19.4% in the presence and 25.6% in the absence of N fertilizer, and inoculation was effective even in the presence of high rates of N fertilizer. Cong et al. (2009) also reported that the maximum yield of rice possible with urea-N was achieved with about 60 kg less urea-N per ha when PGPR was inoculated. The results of the present study are also in agreement with those of Chela et al. (1993), Yasmin et al. (2007) and Cong et al. (2009), who reported that the PGPR and N fertilizer combinations produced significantly higher yields than those from fertilization alone under field condition.

Conclusion

This field study has demonstrated significant effects on potato with N₂-fixing *Bacillus* sp. OSU-142. The highest yield of potato possible with N fertilizer was achieved with about 120 kg N ha⁻¹ when N₂-fixing *Bacillus* sp. OSU-142 inoculated. The results of the present study suggested that *Bacillus* sp. OSU-142 alone or N fertilizer combinations have a great potential to increase yield and yield

components of potato and to reduce the need for chemical fertilizers as in many other crops previously tested. In conclusion, *Bacillus* sp.OSU-142 as plant growth-promoting bacteria can be suitable bio-fertilizer for potato in organic and low-N input agriculture.

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