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Effects of Plant Growth Promoting Bacteria on Canopy Cover of Rangelands in Eastern Region of Turkey

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ABSTRACT

This study was conducted on high-elevation rangelands of Erzurum, Turkey, between 2011 and 2014 for four year. The aim was to determine the effects of Plant Growth Promoting Bacteria (PGPBs) applications on rangeland canopy cover ratio. PGPBs strains (*Pseudomonas fluorescens* T26, *Pantoea agglomerans* 16B, *Paenibacillus polymyxa* TV-12E, *Bacillus cereus* TV-30D, and *Bacillus megatherium* TV-3D) used in this study were obtained from the culture collection unit in the Department of Plant Protection, Faculty of Agriculture at Atatürk University, Erzurum, Turkey. Four study year results showed that PGPBs treatments had significant effects on the canopy cover of rangelands. The highest canopy cover ratio occupied was in treatment T21 (50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*B. megatherium*. TV-3D). There was a significant difference between treatments T21 and the other treatments, while the lowest was in treatment T14 (50 kg N ha^{-1}+*B. cereus* TV-30D). In plots of *P. polymyxa* TV-12E, *P. fluorescens* T26, and *B. megatherium* TV-3D bacteria strains plus half of N+P fertilizer, the canopy cover was higher than that of the other treatments.

Keywords: Bacteria treatment, Environmentally friendly, Natural rangelands, Plant cover, Rangeland fertilization.

INTRODUCTION

Fertilization is important to achieve high yields in crop production. The way to provide sustainable productivity with fertilizer applications is to apply the appropriate fertilizer type at the suitable dose and time. The importance of environmentally friendly practices such as Plant Growth Promoting Bacteria (PGPBs) treatments is increasing day by day in terms of protecting the natural dynamism of the ecological balance and sustainable use and management of resources (Vessey *et al*., 2003; Atieno *et al*., 2020). Especially in the rangeland, vegetation, which are the most important link of the nutrient cycle, applications of Plant Growth Promoting Bacteria (PGPBs) are more important in terms of realizing a healthy and efficient sustainable production extending over a laonger period of time.

Rangelands are vitally important for the national economy and development of the animal's products industry in Turkey, especially in the eastern Anatolia regions of Turkey where animal husbandry mainly depends on rangelands (Gokkus and Koc, 1996; Koc *et al*., 2015). These regions are characterized by low precipitation, short plant growth, long snowy winter (Koc, 2001); and cool-season plants such as sheep fescue (*Festuca ovina* L.) are dominant in the native vegetation of the region (Serin and Tan, 2001). The most limiting factor for plant production in these areas is low precipitation (Herbel and Pieper, 1991; Holechek *et al*. 2011) and it is important to increase the efficiency of rainfall. In addition to the amount, irregular distribution of precipitation causes fluctuations in the yield of rangelands (Herbel and Pieper, 1991).

Due to the effects of mismanagement practices such as excessive grazing, most rangelands have lost their canopy coverage. Proper management of rangelands is the most

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effective way to maintain adequate levels of canopy cover to decrease water and soil run-off (Anonymous, 2005) because proper canopy cover protects the soil against erosion resulting from the impact of raindrops (Ritter, 2012). As the plant cover decrease infiltration rate decreases because canopy cover provides raindrops keeping in soil longer and be available for plant.

Grazing management is one of the most effective practices for the health and restoration of rangelands, but in some conditions, other improvement practices are necessary to increase yield (Holechek *et al*., 2011). Chemical fertilizers have been widely applied to increase rangeland productivity. However, in recent years, some objections have been raised about use of chemical fertilizers, concerns with environmental pollution, production costs, and healthy animal production. Increasing demand for healthy food production and improving environmental quality have focused on the importance of environmentally friendly fertilizers, such as plant growth promotion bacteria (PGPBs), which are rhizosphere bacteria that can increase plant growth by different mechanisms like phosphate solubilization, biological nitrogen fixation, and offer alternatives to chemical fertilizers in agriculture (Bhattacharyya and Jha, 2011; Shrivatava *et al*., 2015).

The aim of this study was to evaluate the effects of different PGPBs, commercial organic fertilizer and chemical fertilizer treatments on canopy cover of rangeland in Erzurum, Turkey, and provide a recommendation regarding the effectiveness of PGPBs on canopy coverage for rangeland users and scientists.

MATERIALS AND METHODS

Bacterial Strains Used in This Study

PGPBs strains (*Pseudomonas fluorescens* T26, *Pantoea agglomerans* 16B, *Paenibacillus polymyxa* TV-12E, *Bacillus cereus* TV-30D and *Bacillus megatherium* TV-3D) used in this study were obtained from the culture collection unit in the Department of Plant Protection, Faculty of Agriculture at Atatürk University, Erzurum, Turkey. These non-pathogenic bacterial strains had been isolated from the *rhizosphere and phyllosphere* of *wild* and traditionally cultivated plants growing in the Eastern Anatolia region of Turkey (Kotan *et al.* 2005; Erman *et al*. 2010). The identity of the bacteria using Sherlock Microbial Identification System was confirmed according to Fatty Acid Methyl Esters (FAME) analysis (Microbial ID, Newark, DE, USA) (Miller, 1982). In the previous studies, all strains were determined that they showed the capacity to grow in N-free conditions, for hormones production (IAA, GA_3) and to solubilize phosphate (Ekinci *et al*. 2014; Turan *et al*. 2014). In the study, bacterial cultures maintained in Nutrient Broth (NB) and 15% glycerol at -80°C were grown on Nutrient Agar (NA). The bacterial isolates and their origin used in our study is presented in Table 1.

Production of Starter Culture for Liquid Inoculants in Fermenters

Frozen bacterial cultures were streaked on Tryptic Soy Agar (TSA, Oxoid) plates. The cultures were individually incubated in TSA at 27°C for 24 hours. After the incubation period, a single colony was transferred to 1,000-mL flasks containing Tryptic Soy Broth (TSB, Oxoid), and grown aerobically in the flasks on a rotating shaker (150 rpm) for 48 hours at 27°C (Merck, Germany). These cultures were used as liquid starter culture for broth culture production into a glass vessel containing 10 mL of TSB medium in an industrial scale in fermenters.

Production of Broth Culture in Fermenters

TSB medium used in the production of broth cultures in fermenters were previously prepared in the glass vessel and sterilized by

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Table 1. Bacterial strains used in this study, their isolated host, nitrogen fixation and phosphate-solubilising activity.

Bacterial strain	Sources	N_2-fixation	P-solubilization
Pantoea agglomerans 16B	*Thymus* sp.	+	+
Bacillus megaterium TV-3D	*Secale* sp.	+	+
Pseudomonas flourescens T26	*Wild raspberries*	S^+	+
Paenibacillus polymyxa TV-12E	*Wheat*	S^+	+
B Bacillus cereus TV-30D	*Wild beet*	+	-

-: Negative reaction, +: Positive reaction, S+: Strong positive reaction.

autoclaving for 20 minutes at 121°C. Then, they were cooled by using chilled water cooling systems to 27°C. The liquid starter cultures were transferred to the glass vessel with the ration of 1-10 % (v/v) of media. Fermentations were carried out in 15 L fermenters (BIOFLO III, New Brunswick Scientific) for 72 hours at 27°C and 700 rpm. For sterilizing the inlet air, a compressor pump with sterile filters was used. The broth cultures were used as starter culture for production of industrial-scale bacterial bioformulation in a bioreactor.

Production of Sterile Liquid Carrier

For the production of industrial-scale bacterial bioformulation in bioreactor, we used the liquid carrier material as growth medium, which provided suitable conditions for them to grow as quickly as possible. Compositions of the Modified Pikovskaya's Medium (Pikovskaya, 1948) as liquid carrier material were as follows (per liter): 1.5 g $Ca_3(PO_4)_2$, 3 g Sucrose, 150 mg $(NH_4)_2$ SO_4, 60 mg NaCl, 30 mg $MgSO_4$ $7H_2O$, 60 mg KCL, 0.4 mg $MnSO_4$, 0.4 mg $FeSO_4$, 150 mg yeast extract, 10 gr *Carboxymethyl cellulose*, 20 mL glycerine, 75 mL whey powder (93% water), 20 mL vinas, 40 mL, liquid seaweed extract and 695 mL distilled H_2O. The pH of the carrier material was adjusted to 7 by HCL. The mixture was homogenized and sterilized with water vapour at 110°C and 2 psi for 30 minutes. After cooling down to 27°C by using chilled water cooling systems, the starter culture was added.

Production of Industrial-Scale Bacterial Bioformulation in Bioreactor

The broth bacterial cultures grown in fermenters were transferred into an industrial scale bioreactor containing 500 L of the liquid carrier solution. Fermentations were carried out in 1000 L bioreactor for 72 hours at 27°C and 200 rpm until reaching maximum cell concentrations of 10^9 cells mL^{-1}. For sterilizing the inlet air, a compressor pump with a 0.45 µm sterile filters was used. After about 72 hours fermentation period, the broth was packed in in sterilized polyethylene bags (1 L/pack) by using an automatic dispensing machine (auto syringe). They were stored at 4°C for long-term storage before use. These bacterial bioformulations were used as biofertilizer for microbial inoculation of plant roots. One of the stored broth cultures was analyzed for pH and viable cell population at monthly intervals up to 12 months.

Application Pocedure of Bacterial Bioformulation

Bacteria application was carried out using the spray method. Approximately, 0.2 g of sucrose (10 mg mL^{-1}) was added to each clear spray bottle containing 500 mL of the bacterial bioformulation ($1x10^7$ CFU mL^{-1}). After shaking, the suspension was sprayed on plants. Additional applications were started 15 days after the first application.

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Research Area and Treatments

This study was carried out on rangeland in 37S 0677633E- 4420815N and 37S 0677625E-4420790 N with 2010 m altitude in Erzurum, Turkey, for 4 years. The pasture where the study was carried out was exposed to grazing for a long time. The study area was delineated and fenced in the year 2010. to protect animal grazing. In this study, 22 treatments including: (T1) Control (No bacteria and fertilizer), (T2) N+P (100 kg N ha^{-1} + 50 kg P_2O_5 ha^{-1}), (T3) N (100 kg N ha^{-1}), (T4) P (50 kg P_2O_5 ha^{-1}), (T5) *P. fluorescens* T26, (T6) *P. agglomerans* 16B, (T7) *P. polymyxa* TV-12E, (T8) *B. cereus* TV-30D, (T9) *B. megatherium* TV-3D, (T10) Commercial organic fertilizer, (T11) 50 kg N ha^{-1}+*P. fluorescens* T26, (T12) 50 kg N ha^{-1}+*P. agglomerans* 16B, (T13) 50 kg N ha^{-1}+*P. polymyxa* TV-12E, (T14) 50 kg N ha^{-1}+*B. cereus* TV-30D, (T15) 50 kg N ha^{-1}+*B. megatherium* TV-3D, (T16) 50 kg N ha^{-1}+commercial organic fertilizer, (T17) 50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*P. fluorescens* T26, (T18) 50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*P. agglomerans* 16B, (T19) 50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*P. polymyxa* TV-12E, (T20) 50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*B. cereus* TV-30D, (T21) 50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*B. megatherium*. TV-3D, (T22) 50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+commercial organic fertilizer were applied. Fertilizers used were ammonium nitrate (33.5 percent nitrogen) and triple superphosphate (44 percent available P_2O_5). The study was designed in a randomized complete block design with three replications. The size of treatment plots was 6 m^2 and the total plot number was 66. There was 3 m distance between blocks, and 2 m between plots to prevent treatment contaminating each other.

In research area, Sheep fescue, Smooth brome, and Blue grass were common grasses, and Clover, Sainfoin, and Alfalfa, which have nitrogen-fixing properties, were common legumes. Also, Thyme, Veronica, Alssum were common weed species.

Saturation extract electrical conductivity for 25°C was measured with a Schott Instruments Lab 960 brand EC meter (Rhoades *et al*., 1990; Ayyıldız, 1990). Potassium (K) concentrations in the soil was measured by the method determined in the ICP-AES (Inductively Coupled Plasma, Varian Vista Pro, Austria) device (Knudsen *et al*., 1982; Thomas, 1996). While determining the bulk weight of the soils in the research area, the mass/volume relationship was used. (Grossman and Reinsch, 2002). Soil textures were determined by Bouyoucus Hydrometer method (Gee and Bauder, 1986), lime contents were determined by Scheibler calcimeter by volumetric (Nelson, 1982), organic matter contents were determined by Smith-Weldon method (Nelson and Sommers, 1982), phosphorus contents were determined by spectrophotometer using molybdophosphoric blue color method. (Olsen and Sommers, 1982), pH and electrical conductivity the pH in the extract obtained from the saturation paste was determined by the electrical conductivity device (Thomas, 1996). Some chemical and physical properties of the soils taken from study area are shown in Table 2. The climatic data of the experiment areas for the study years are shown in Table 3.

Botanical composition of the rangeland sites were determined by the line intercept method developed by Canfield (1941); calculations were made by dividing the total plant species ratio by the total plant species number and multiplying by 100, according to Holechek *et al*. (2011) suggestions for each year in July. In the study, SAS (2002) GLM was used to evaluate the data.

RESULTS

Four study years results showed that Plant Growth Promoting Bacteria (PGPBs) treatments had significant effects on canopy cover of rangelands. The highest canopy cover ratio was in treatment T21 (50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*B. megatherium*. TV-

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Table 2. Results of physical and chemical soil analysis in treatment plots.

Saturation (%)	pH	EC (dS m^{-1})	P_2O_5 (kg/da)	Organic matter (%)	Lime (%)	K_2O (kg/da)	Volume weight (g cm^{-3})	Structure (%)		
								Clay	Loam	Sand
50	7.03	1.81	4.82	2.77	0.33	98	1.2	41.8	20.72	37.48

Table 3. Climatic data and soil temperature of study area.

		Autumn	Winter	Spring	Summer	Total/Mean
2011	Total precipit. (mm)	73.3	59.3	262.6	102.8	498.0
	Average temp. (°C)	5.12	-4.60	2.41	15.97	4.71
	Average soil temp. (5 cm) (°C)	7.71	1.15	8.53	22.42	9.95
2012	Total precipit. (mm)	43.7	66.0	121.2	25.5	256.4
	Average temp. (°C)	7.59	-11.55	3.94	17.89	4.47
	Average soil remp.(5 cm) (°C)	12.21	-3.92	9.61	21.50	9.85
2013	Total precipit. (mm)	94.0	82.2	86.1	41.2	303.5
	Average temp. (°C)	8.88	-7.34	6.14	17.69	6.34
	Average soil temp.(5 cm) (°C)	13.11	-0.04	10.70	25.37	12.28
2014	Total precipit. (mm)	114.0	50.8	174.5	23.2	362.5
	Average temp. (°C)	7.58	-5.16	-4.76	15.28	3.24
	Average soil temp. (5 cm) (°C)	12.21	0.42	2.98	18.18	8.45

3D), and there was significant difference between T21 and the other treatments, while the lowest was in T14 (50 kg N ha^{-1}+*B. cereus* TV-30D) (Table 4).

Canopy cover ratios, determined in T7-*P. polymyxa* TV-12E, T5-*P. fluorescens* T26 bacteria strains alone, and T2- 100% N+P fertilizer treatment plots, and in T6-*P. agglomerans* 16B, T7- *P. polymyxa* TV-12E, T11- *P. fluorescens* T26, commercial organic fertilizer plus 50% N treatment plots, canopy coverage ratios were significantly ($P< 0.01$) different from control plots (Table 4). In addition, except for T8-*B. cereus* TV-30D strains, all bacterial inoculation with half of N+P fertilizers significantly increased canopy coverage ratios.

Canopy cover, obtained from some of the treatments with bacteria strains plus 50% N alone or 50% N+50% P fertilizers, were higher than the treatments with bacteria strains without fertilizers. Although T2-100% N+ P without bacteria strains had higher canopy cover than T1- check plots and bacteria alone (T5, T6, T7, T8, T9), there was no significant difference between this treatment with T5- *P. fluorescens* T26 and T7- *P. polymyxa* TV-12E bacteria strains alone.

The most significant canopy cover increases were determined in T7- *P. polymyxa* TV-12E, T5- *P. fluorescens* T26, and T21- *B. megatherium* TV-3D bacteria strains plus half of N+P fertilizer treatments. After these treatments, T16- 50 kg N ha^{-1}+commercial organic fertilizer, and T11- 50 kg N ha^{-1}+*P. fluorescens* T26 treatments had significantly higher canopy cover than T2-100% N alone, 100% N+P fertilizer and most of the other treatments (Table 4).

In the control plots, canopy coverage ratio was higher than in the other treatment in the year (2010) before the study. In treatments T3 (100 kg N ha^{-1}) and T4 (50 kg P_2O_5 ha^{-1}) plots of the canopy coverage ratios were similar to the control plots and in the other treatments, canopy cover was slightly higher than T1-control plots (Figure 1).

There was a significant difference between study years based on average canopy cover: the highest canopy cover was determined in the last study year and it was the lowest in the first study year (Figure 2). In the first study year, while the precipitation value was the highest and canopy cover ratio was

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Table 4. Changes in average canopy cover (%) in different PGPBs treatment plots.

	Treatments	Average canopy cover (%)
T1	Control (No bacteria and fertilizer)	42.08 G
T2	N + P (100 kg N ha^{-1}+50 kg P_2O_5 ha^{-1})	46.75 D
T3	N (100 kg N ha^{-1})	37.42 I
T4	P (50 kg P_2O_5 ha^{-1})	37.92 I
T5	*P. fluorescens* T26	46.50 D
T6	*P. agglomerans* 16B	42.83 G
T7	*P. polymyxa* TV-12E	46.25 DE
T8	*B. cereus* TV-30D	38.08 I
T9	*B. megaterium* TV-3D	37.92 I
T10	Commercial organic fertilizer	41.00 H
T11	50 kg N ha^{-1}+*P. fluorescens* T26	48.00 C
T12	50 kg N ha^{-1}+*P. agglomerans* 16B	45.17 EF
T13	50 kg N ha^{-1}+*P. polymyxa* TV-12E	44.25 F
T14	50 kg N ha^{-1}+*B. cereus* TV-30D	36.33 J
T15	50 kg N ha^{-1}+*B. megaterium* TV-3D	40.92 H
T16	50 kg N ha^{-1}+Commercial organic fertilizer	48.58 C
T17	50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*P. fluorescens* T26	47.92 C
T18	50 kg N ha^{-1} + 25 kg P_2O_5 ha^{-1}+*P. agglomerans* 16B	45.58 DE
T19	50 kg N ha^{-1} +25 kg P_2O_5 ha^{-1}+*P. polymyxa* TV-12E	50.33 B
T20	50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*B. cereus* TV-30D	43.08 G
T21	50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*B. megaterium*. TV-3D	52.00 A
T22	50 kg N ha^{-1} +25 kg P_2O_5 ha^{-1}+Commercial organic fertilizer	46.50 D

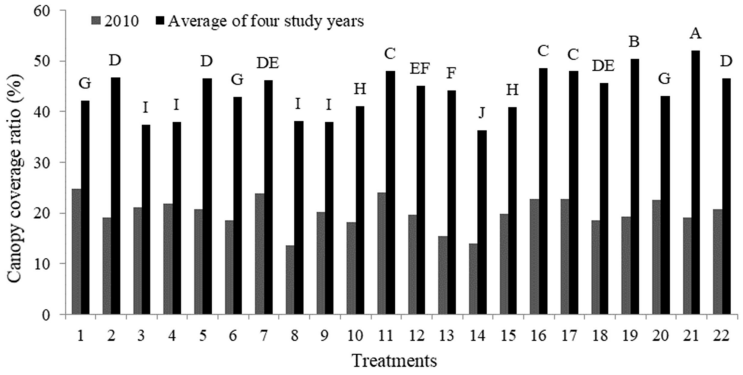

Figure 1. Average canopy cover ratios in study years and the year (2010) before study.

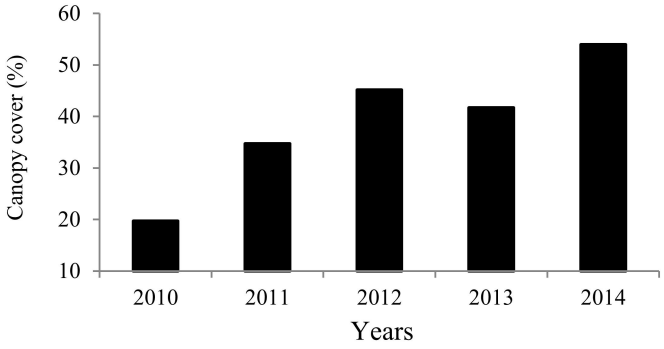

Figure 2. Changes in average canopy cover ratios in study years (%).

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significantly lower than that in the other years. These results may be due to irregular precipitation distribution in the growing seasons. In the second study year, annual total precipitation value was lower than the other years, but canopy cover showed increasing trend, because of the effects of higher spring precipitation in this year.

In the third study year, canopy cover ratio decreased because total precipitation and seasonal precipitation, especially in spring period, was lower than the other years. Because of the increase in total and seasonal precipitation in the last study year, canopy cover showed increasing trend (Table 4 and Figure 3). Canopy coverage ratios significantly increased in all treatments and the control plots, compared to the year (2010), before the study years (Figure 1). Similar to the treatment plots, canopy cover ratios in all study years were higher than that in 2010 year, just before the study.

DISCUSSION

There was a significant interaction between years and treatments most probably due to the diversity in climatic condition, recorded in the study years (Table 3). In study years, the reasons for the canopy coverage ratios being higher than 2010 is probably the effects of the enclosure to animal grazing and treatments (Dasci *et al*., 2010). The reason for the difference between study years is most probably related to the diversity in climatic conditions. In some years, total precipitation was higher than the others, and higher level of soil moisture might increase the efficiency of fertilizers and activity of bacteria, because in low moisture conditions both fertilizer efficiency (Stark and Firestone, 1995; Su-me *et al*., 2020) and the survival and activity of microorganisms may change (Orchard and Cook, 1983). Greater performance of the bacteria tested under relatively wetter conditions prevailed in the second and fourth year and this, probably, substantiate the suggestion that the N_2-fixing and P-solubilizing activity of free-living bacteria may strongly depend on favorable moisture, pH, temperature and climatic conditions of the soil (Sahin *et al*. 2004; Sharma *et al*. 2013). The beneficial effects of the bacteria on canopy cover of rangelands varied significantly depending on bacterial strains and, especially, precipitation during the growing season, years, and soil conditions. As reported previously, the effect of PGPBs is a complex process and depends on the bacterial strains and populations, the plant-bacterial strains combination, the growth parameters, environmental conditions, years, plants and soil types (Sahin *et al*., 2004; Cakmakci *et al*., 2006, 2009, 2014;

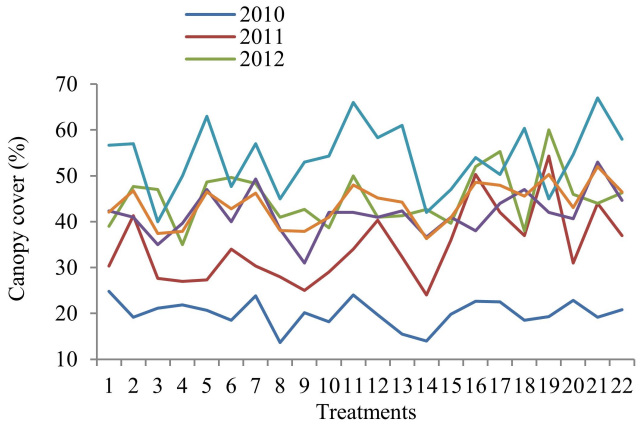

Figure 3. The interaction effects of year and treatment on canopy cover ratios (%).

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Olanrewaju *et al*., 2017)

The reason of canopy cover increase in control plots in study years is most probably the positive effects of the enclosure to animal grazing, because the rangeland where the study was carried out was exposed to grazing for a long time, which reduced the canopy cover. On the other hand, high canopy coverage ratio in some bacteria strain plus half of N + P treatments probably resulted from the combined effects of bacteria strains and N + P fertilizers. It is expressed that plant growth promoting rhizobacteria facilitates uptake (Verma *et al*., 2010) and enhances the availability of certain nutrients (Holguin *et al*., 1999; Richardson, 2001; Cakmakci *et al*., 2009, Erturk *et al*., 2011) and the use of chemical fertilizers may decrease due to the application of rhizobacteria that promote plant growth (Adesemoye *et al*., 2009; Civelek and Yildirim, 2022). PGPBs are also considered as inexhaustible sources of nitrogen and phosphorous that helps in providing these nutrients to plants by different mechanisms (Cakmakci *et al*. 2006, 2009). The increasing effects of PGPBs on the availability of applied fertilizer or soil organic nutrients may change plant community (Walter *et al*., 2016) and the changing of plant community enhances the canopy cover (Khumalo *et al*. 2007).

Canopy cover showed increasing trend in the study years, due to the positive effects of the enclosure to animal grazing and treatments. These results are supported by the results of a research that reported perennial grass cover was better maintained in reducing grazing pressure conditions (Khumalo *et al*., 2007) and it may be affected by climatic conditions (Tieszen *et al*., 1979).

CONCLUSIONS

The aim of the study was to determine the effects of plant growth-promoting bacteria (PGPBs) on rangeland canopy cover ratio. PGPBs strains (*Pseudomonas fluorescens* T26, *Pantoea agglomerans* 16B, *Paenibacillus polymyxa* TV-12E, *Bacillus cereus* TV-30D and *Bacillus megatherium* TV-3D) were used in this study. Also some commercial chemical fertilizers were applied. Results of four years study showed that PGPBs treatments had significant effects on canopy cover of rangelands. The highest canopy cover ratio was in treatment T21 (50 kg N ha^{-1}+25 kg P_2O_5 ha^{-1}+*B. megatherium*. TV-3D). Furthermore, results indicated that application of PGPR with half dose of the recommended N fertilizer may increase canopy cover of rangelands and reduce the need for applying additional fertilizer.

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اثرات باکتری های محرک رشد گیاه بر تاج پوشش مراتع منطقه شرق ترکیه

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چکیده

این پژوهش در مراتع مرتفع ارزروم (Erzurum) ترکیه بین سالهای ۲۰۱۱ تا ۲۰۱٤ به مدت چهار سال انجام شد. هدف پژوهش تعیین اثرهای کاربرد باکتری‌های محرک رشد گیاه (PGPBs) روی نسبت پوشش تاج‌مرتع بود. سویه‌های PGPBs استفاده شده در این مطالعه (Pseudomonas fluorescens T26، Pantoea agglomerans 16B، Paenibacillus polymyxa TV-12E، Bacillus cereus TV-30D، و Bacillus megatherium TV-3D) از واحد جمع‌آوری کشت در بخش حفظ نباتات دانشکده کشاورزی در دانشگاه آتاتورک، ارزروم، ترکیه به دست آمد.نتایج چهار سال مطالعه نشان داد که تیمارهای PGPBs اثرات قابل توجهی بر پوشش تاج مراتع داشتند. بیشترین نسبت پوشش تاج پوشش اشغال شده در تیمارT21 (۵۰ کیلوگرم نیتروژن در هکتار + ۲۵ کیلوگرم P_2O_5 در هکتار+ B. megatherium. TV-3D بود. بین تیمارهای T21 و سایر تیمارها تفاوت معنی‌داری وجود داشت، در حالی که کمترین آن در تیمارT14 (۵۰کیلوگرم نیتروژن در هکتار (B. cereus TV-30D +) بود. در کرت های سویه های باکتریP. polymyxa TV-12E ، P. fluorescens T26، و B. megatherium TV-3D به علاوه نیمی از کودN + P ، پوشش تاج پوشش بالاتر از تیمارها ی دیگر بود.

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