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Efficacy of Bacilli Strains in Growth Promotion and Biological Control of Soilborne *Rhizoctonia* and *Fusarium* on Alfalfa (*Medicago sativa* L.) and Potato (*Solanum tuberosum* L.).

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ABSTRACT

Rhizoctonia solani and *Fusarium solani* are among the most important soilborne root pathogens affecting production of alfalfa (*Medicago sativa* L.) and potato (*Solanum tuberosum* L.) in Qassim region, Saudi Arabia. This study aimed to test 5 strains of *Bacillus* spp., isolated from Qassim soil for controlling *R. solani* and *F. solani* in greenhouse experiments. The efficacy of the 5 plant growth-promoting Rhizobacteria (PGPR), (*Bacillus amyloliquefaciens* sub sp. *plantarum* ME 8, *Paenibacillus polymyxa* ME6, *Bacillus amyloliquefaciens* sub sp. *plantarum* ME 106, *Bacillus subtilis* ME 105, and *Bacillus amyloliquefaciens* subsp. *plantarum* ME 3), on controlling *R. solani* and *F. solani* on alfalfa and potato were tested. Seeds of alfalfa, California-1 cultivar and Sponta potato variety microtubers were treated with PGPR powder and sown in *R. solani* or *F. solani* artificially infected soil in a greenhouse. Seed treatments with PGPR powders induced significant changes in percentage of seed germination and/or plant fresh and dry weights. *R. solani* was more virulent on alfalfa than *F. solani*. Fresh weights and dry weights of alfalfa seedlings increased by up to 21% in response to seed treatments with *Paenibacillus polymyxa* ME6 or *B. amyloliquefacaciens* subsp. *plantarum* ME 106 when compared to seeds treated with the fungicide Rizolex or non-treated check. Similar results were found with Sponta potato variety. However the effect of PGPR isolates on the interactions between *R. solani* and *F. Salami* and potato were not highly distinctive as found with the alfalfa.

Key words: Alfalfa, *Bacillus* spp., *Fusarium solani*, PGPR, Potato, *Rhizoctonia solani*.

INTRODUCTION

Alfalfa (*Medicago sativa* L.) and potato (*Solanum tuberosum* L.) are the most common field crops cultivated in Al-Qassim region and Saudi Arabia, where 14,785 and 2,500 hectares are planted with these crops, respectively (Anonymous, 2010). Pathogenic microorganisms affecting plant health are a major and chronic threat to food production and ecosystem stability worldwide. As agricultural production intensified over the past few decades, producers became more dependent on agrochemicals as a reliable method for crop protection. However, increasing usage of chemical expressed several negative impacts, *e.g*., development of chemical resistance pathogenic races and environmental determination (Gerhardson, 2002). Furthermore, the growing cost of pesticides, particularly in less-affluent regions of the world and consumer demand for pesticide-free food has led to a search for substitutes to these products. There are also a number of fastidious diseases for which chemical solutions are few, ineffective, or nonexistent (Gerhardson, 2002). Biological control has thus been considered as an alternative or a supplemental way of reducing the use of chemicals in agriculture (Welbaum *et al.*, 2004). Agriculture production in Saudi Arabia in general and in particular in Al Qassim region is mostly dependent on the use of chemicals as fertilizers and for pest control.

There has been a large body of literature describing potential uses of plant-associated bacteria as agents stimulating plant growth and managing soil and plant health (Sturz *et al.*, 2000 and Welbaum *et al.*, 2004). PGPB are associated with many, if not all, plant species and are commonly present in many environments (Bashan and Holguin, 1998). The most widely studied group of PGPB is plant growth-promoting rhizobacteria (PGPR) (Kloepper and Schroth, 1978), which colonizing the root surfaces and the closely adhering soil interface the rhizosphere (Kloepper *et al.*, 1999). Corn, sorghum and wheat grains treated with certain *Pseudomonas* spp. were protected against root pathogens and significant grain yields were reported (El-Meleigi, 1989). As reviewed by Kloepper *et al.*, 1999 some of these PGPR can also enter root interior and establish endophytic populations. Many of them are able to transcend the endodermis barrier, crossing from the root cortex to the vascular system and subsequently thrive as endophytes in stems, leaves, tubers and other organs (Haas *et al.*, 2000 and Bloemberg and Lugtenberg, 2001). The extent of endophytic colonization of host plant organs and tissues reflects the ability of bacteria to selectively adapt to these specific ecological niches (Haas *et al.*, 2000). Consequently, intimate associations between bacteria and host plants can be formed without harming the plant (Leeman *et al.*, 1995; Lodewyckx *et al*., 2002 and Compant *et al*., 2005). Despite their different ecological niches, free-living rhizobacteria and endophytic bacteria use

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some of the same mechanisms to promote plant growth and control phytopathogens (Sturz *et al.*, 2000; Lodewyckx *et al.*, 2002 and Gray and Smith, 2005).

In attempts to minimize dependence on chemical fungicides for controlling root diseases in Central Saudi Arabia fields, *Bacillus polymyxa* strain was isolated from wheat roots in Al-Qassim region and used successfully in the field for control of wheat root rot (El-Meleigi *et al.*, 2007) and cucurbits gummy stem (black rot) (El-Meleigi and Al-Rehiani, 2004). Rhizoctonia and root rot diseases caused by *Rhizoctonia solani and Fusarium solani* are among the most common diseases of alfalfa and potato in Al-Qassim region (Al-Kherb *et al*., 1996 and El-Meleigi, 2008).

Therefore, the main goal of this study was to offer an alternative biologically safe tool for control of *Fusarium* and *Rhizoctonia* diseases in Al-Qassim region, Saudi Arabia, using native isolates of rhizobacteria.

MATERIALS AND METHODS

Source of PGPR isolates

The PGPR isolates were tested for their antibiosis efficacy against *Rhizoctonia solani* and *Fusarium solani* and secretion of Indole Acetic Acid (IAA) as described by (El-Meleigi *et al.*, 2014). The efficacy of *Bacillus amyloliquefaciens* subsp. *plantarum* ME 8, *Paenibacillus polymyxa* ME6, *Bacillus amyloliquefaciens* subsp. *plantarum* ME 106, *Bacillus subtilis* ME 105, and *Bacillus amyloliquefaciens* subsp. *plantarum* ME 3 on control of *R. solani* and *F. solani* on alfalfa and potato were tested.

Formulation of bacteria for use in biological control

The PGPR isolates were prepared in the form of powder for seed treatments and field applications as described by El-Meleigi (1989). PGPR isolates were grown on PDA media plates for 48 h at 28°C. Sterile cornstarch, mixed in sterile 0.1 M Mg SO4 (10% w/v.) with the pH adjusted to 7.2 by adding CaCO3. Bacterial growth was scraped from the agar surfaces with a sterile glass rod and mixed with the starch suspension (four plates/250 ml). Bacteria were suspended in sterilized corn starch and dried in a stream of sterile air in a laminar flow hood for 2 h. PGPR powders were placed in sterile plastic bags and stored at -5ºC until used for alfalfa seed or potato tuber treatments. The effect PGPR isolates and formulations on germination of alfalfa seeds (Calfornia1 variety) and growth potato microtubers (Sponta potato) were tested. Micro tubers of potato, (var. Sponta) were collected from the field and stored at 5ºC for two months, then left at room temperature until germinated.

Both, alfalfa seeds and potato microtubers were washed with tap water; surface sterilized by dipping in 10% Clorox (*a.i*. 0.5% NaOCl) for 30 sec., washed with sterilized distilled water and blotted dry on sterile filter paper. PGPR formulated powder of each isolate was suspended in sterilized distilled water (10g/100 ml). Alfalfa seeds and potato micro tubers were dipped in the PGPR suspension for 30 sec. and left to dry in a sterile air stream in a the Laminar flow hood for 1 hr. Plastic jars (12 and 16 cm in diameter with 4 small holes in the bottom) were filled each with sterilized soil mix (120-160 g), respectively for sowing alfalfa seeds or potato tubers.

Isolation and inoculation of pathogenic isolates

Inocula of *R. solani* and *F. solani*, isolated from infected alfalfa and potato plants, were prepared by inoculating sterile wheat grains and incubated at 25ºC in a dark for 3 weeks. Each soil jar was inoculated with 2 g of pathogen colonized wheat grains. Check treatments were inoculated with 2 g of sterilized non-inoculated wheat grains, according to El-Meleigi *et al.* (2014). Seed treatments included 5 PGPR isolates and Rizolex fungicide. Jars were placed in a greenhouse and wetted for establishing fungal inoculum for one week before seeding. The temperature was maintained in the greenhouse during the experiments at 25±4ºC. Alfalfa plants were grown in the greenhouse for 4 weeks, while potato was grown for 6 weeks before recording the results. Experiments were set as a Randomized Complete Block design (RCBD). The fungicide Rizolex (Tolclofos-methyl) was applied to seeds, as recommended for protecting crop seedlings against *Rhizoctonia* and *Fusarium* at the rate of 50 lb/A (EPA Reg. No. 59639-178). Alfalfa seeds and potato tubers were dusted with Rizolex 50% WP as a control treatment in comparison to other treatments in greenhouse experiments.

Statistical analysis

Data were statistically analyzed according to the technique of analysis of variance (ANOVA) for two factors randomized complete block design (RCBD). All analyses of variance were computed using the MSTATC microcomputer program (Mstatc, 1990).

RESULTS AND DISCUSSION

The interactions among all the experimental factors involved in testing *Bacillus* strains on alfalfa, fresh weight, dry weight and number of surviving

plants in soil infested with either *R. solani* or *F. solani* are presented in table (1). The results showed that *R. solani* caused significant losses in alfalfa, fresh weight and number of surviving plants compared to the *F. solani*. The interactions of all the other treatments including soilborne pathogen, seed treatments with the fungicide Rizolex or *Bacillus* spp. influenced the results significantly. According to fresh weights, dry weights and number of surviving alfalfa plants, PGPR *P. polymyxa* ME6 and *B. amyloliquefaciens* subsp. *plantarum* ME B106 were generally the most effective in protecting plants from disease, regardless of the soilborne pathogen. *B. subtilis ME* 105 and *B. amyloliquefaciens* subsp. *plantarum* ME B3 were ineffective against *R. solani* and *F. solani*. Results in table (1) showed also that the interaction between the pathogens and seed treatments was also significant. When the soil was infected with *R. solani*, the highest values of germination percentages (70.5-86%), fresh weights (5.6- 6.8g) and dry weights were associated with treating alfalfa seeds with *P. polymyxa* ME6 and BAP ME106 when compared to non-treated seeds sown in pathogens' infected soils. The same trend was also observed with BAP ME106. The other 3 PGPR isolates had no significant effects on alfalfa seedlings growth and did not offer significant protection against soilborne fungi compared to Rizolex fungicide, PP ME6 or BAP ME106 seed treatments (Fig. 1).

Concerning the virulence of *R. solani* and *F. solani*, the results showed that *R. solani* was significantly more virulent on alfalfa than *F. solani* (Table 2), as the infecting soil with *R. solani* reduced significantly fresh weights, dry weights and percentage of alfalfa germination when compared to *F. solani*. On the other hand, BAP ME8, BAP ME 3 and BS ME 105 isolates were insignificantly effective against *R. solani* as shown by seedlings, fresh weight, dry weight and percent of germs.

The results of the interactions of various treatments on fresh weight, dry weight and plant height of 6 weeks old potato plants grown in the greenhouse are presented in table (3). *R. solani* and *F. solani* differed significantly in plant dry weighs, where *F. solani* was more repressive to plant dry weight than *R. solani*. Results also showed that the 4 PGPR isolates (*B. amyloliquefaciens* subsp. *plantarum* ME B8, *P. polymyxa* ME6, *B. amyloliquefaciens* subsp. *Planetarium* ME B106 and *B. subtilis* ME 105) increased plant dry weights significantly over fungal infection, regardless of soilborne fungal pathogen. On the other hand, insignificant differences were found in potato, fresh weights or plant heights, related to these 4 PGPR isolates. Best growth parameters were found in check microtubers (Table 3). The effect of interactions between fungal pathogens and other treatments had a significant effect on fresh weights and plant heights and highly significant effect on potato dry weights. The effect of various treatments on potato growth was insignificant in most cases; however *B. subtilis* ME 105 increased fresh weight significantly over *R. solani* only infected plants, but was not as effective against *F. solani* (Table 4). *B. amyloliquefaciens* subsp. *plantarum* ME B106 and *B. subtilis* ME 105 increased potato dry weights significantly when compared to other PGPR potato microtubers treatments (Table 4 & Fig. 2). Rhizobacteria are an important functional group of beneficial bacteria used for plant growth promotion and control of soil borne pathogens (Hoflich *et al.*, 1994 and Rajkumar *et al.*, 2004). Screening studies resulted in the selection of 6 efficient strains out of approximately 35 strains isolated from the rhizosphere of alfalfa and potato (El-Meleigi *et al.*, 2014).

Regarding a selection of the most effective growth promoters of the 6 PGPR isolates, on alfalfa seed germination and growth test, *B. amyloliquefacaciens* subsp. *plantarum* ME 3 and *B. amyloliquefacaciens* subsp. *planetarium* ME 106 were the only isolates, which increased seedling growth over the non-treated seeds (5.71% of the tested isolates). The other isolates were either neutral 40% (14 isolates) or suppressive to alfalfa germination 54.3% (Table 1). Therefore, it is suggested that these 2 isolates were growth promoters to alfalfa as well as good antagonists to their root pathogens, *R. solani* and *F. solani*. This may suggest that fungistatic metabolites and antibiotics as well as growth promotion materials are secreted by certain PGPR isolates. Previous research indicated that PGPR isolates produced β 1, 3-glucanase, salicylic acid, and HCN when inhibiting the mycelial growth of *R. solani* (Nagarajkumar *et al.*, 2004). Suppression of hyphal growth of *R. solani* and *Fusarium* spp. by PGPR had also been reported (El-Meleigi, 1989).

In the present study, BAP ME 106 and PP ME 6 were the strong growth promoters to alfalfa and offered better protection against the soybean; *R. solani* and *F. solani*. As the host plant plays an important role in supporting the introduced antagonists in field conditions, a screening method involving the host plant, pathogen and the antagonist is expected to give a more realistic picture than the dual culture plate assay (Rajkumar *et al.*, 2004). Therefore, greenhouse experiments on PGPR isolates showed the best results obtained from the 3 screening methods described above. In greenhouse tests, *R. salami* was significantly more virulent on alfalfa than *F. solani*, regardless of seed treatments. The PP ME6 and BAP ME106 isolates were the most effective

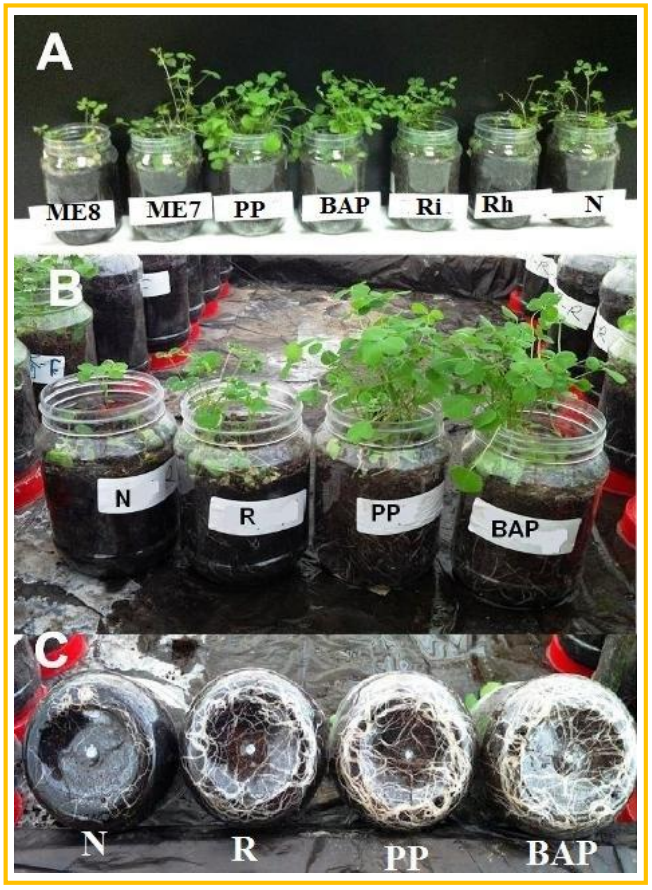

Fig. (1): Effect of PGPR isolates on germination, foliar and root growth of growth of Alfalfa, California1 variety, in greenhouse 4 weeks after sowing in soil infested with *Rhizoctonia solani*.: **A -** Effect of seed treatments with *Bacillus amyloliquefaciens* subsp. *plantarum* ME 8 (ME8), *Bacillus subtilis* ME 105(ME7), *Paenibacillus polymyxa* ME6 (PP, *Bacillus amyloliquefaciens* subsp. *plantarum* ME B106 (BAP)), Rizolex fungicide (Ri), no seed treatment (Rh) and no seed or soil treatments (N). B- Seeds not treated (N), seeds treated with Rizolex (R), seeds treated with *Paenibacillus polymyxa* ME6 (PP), seeds treated with *Bacillus amyloliquefaciens* subsp. *plantarum* ME 106 (BAP). **C**- Effect on growth of root system- Seeds not treated (N), Seeds treated with Rizolex (R), Seeds treated with *Paenibacillus polymyxa* ME6 (PP) and alfalfa seeds treated with *Bacillus amyloliquefaciens* subsp. *plantarum* ME 106 (BAP).

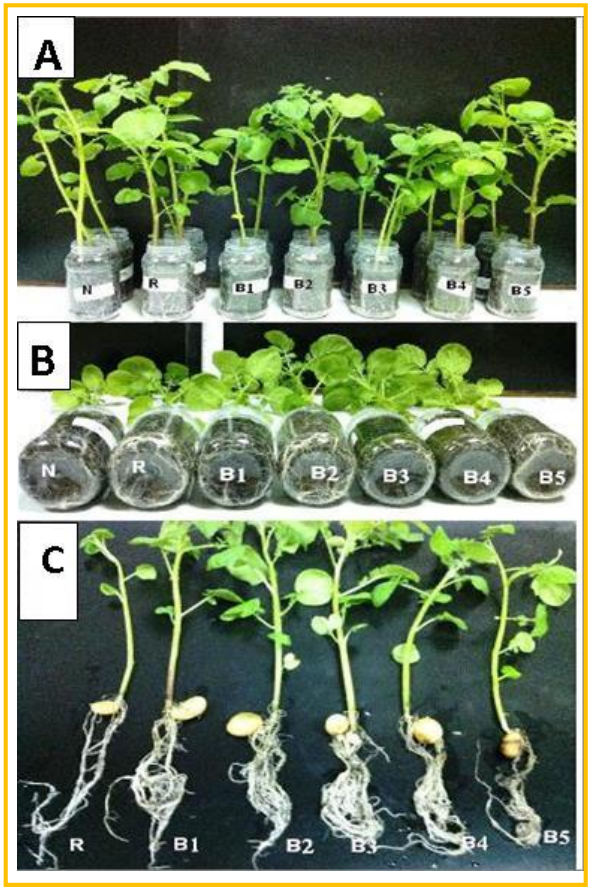

Fig. (2): Effect of PGPR isolates on plant height and root growth of 6 weeks old Sponta potato in the greenhouse, A &B: Comparison of foliar and root growth of potato plants non treated tubers and non-infested soil (N) or Rizolex treated tubers sown in *R. solani* infested soil (R) tubers treated with *Bacillus amyloliquefaciens* subsp. *plantarum* ME B8 (B1), *Paenibacillus polymyxa* ME6 (B2), *Bacillus amyloliquefaciens* subsp. *plantarum* ME B106 (B3) *Bacillus subtilis* ME 105 (B4) *Bacillus amyloliquefaciens* subsp. *plantarum* ME B3 (B5), C: comparison of the effect of the above treatments on potato roots.

Table (1): Interactions among *Rhizoctonia solani* and *Fusarium solani* soil treatments, *Bacillus* spp. and Rizolex fungicide seed treatments on fresh weight, dry weight and germination of 4 weeks old California 1 alfalfa variety in greenhouse

Treatments	Fresh Weight (g)	Dry Weight (g)	Germination (%)
Pathogenic fungi			
R. solani	4.78^{b}	0. 42^{b}	58.4^{b}
F. solani	6.03^{a}	0. 48^{a}	81.1^{a}
F-test	**	**	**
Seed and soil treatments	***R. solani/F. solani***		
None (Check)	6.54^{ab}	0.53^{ab}	77.0^{a}b
Fungi	4.27^{d}	0.37^{c}	66.0b^{c}
Rizolex	5.24^{cd}	0.35^{c}	75.5^{ab}
Bacillus amyloliquefaciens subsp. *plantarum* ME 8	4.61^{cd}	0.40^{c}	73.5^{b}
Paenibacillus polymyxa ME6	6.80^{a}	0.61^{a}	86.0^{a}
Bacillus amyloliquefaciens subsp. *plantarum* ME 106	5.60^{bc}	0.51^{b}	70.5^{b}
Bacillus subtilis ME 105	5.23^{cd}	0.40^{c}	52.0^{d}
Bacillus amyloliquefaciens subsp. *Plantarum* ME 3	4.96^{cd}	0.42^{c}	57.5^{cd}
F-test	**	**	**
Interaction between pathogens and seed treatments			
F-test	**	**	**

Table (2): Effect of treating of alfalfa seeds, California 1variety, with Rizolex fungicide, five PGPR Isolates of *Bacillus* spp. and *Paenibacillus polymyxa* on protection against *Rhizoctonia solani* and *Fusarium solani*, as determined by fresh weight, dry weight and germination percentage of 4 weeks old plants in greenhouse

Treatments	Fresh Weight (g)		Dry Weight (g)		Germination (%)	
	R. solani	***F. solani***	***R. solani***	***F. solani***	***R. solani***	***F. solani***
Check	7.56^{ab}	5.52^{cde}	0.68^{a}	0.39^{cd}	70^{cd}	84^{abc}
Fungi only	3.02^{f}	5.52^{cde}	0.33^{d}	0.40^{bcd}	53^{e}f	79^{abc}
Rizolex	4.10^{ef}	6.38^{abcd}	0.38^{cd}	0.33^{d}	70^{cd}	81^{abc}
Bacillus amyloliquefaciens subsp. *plantarum* ME 8 (BAP ME8)	3.56^{f}	5.66^{cde}	0.41^{bcd}	0.39^{cd}	61de	86^{ab}
Paenibacillus polymyxa ME6 (PP ME6)	7.86^{a}	5.74^{cde}	0.68^{a}	0.54^{b}	81^{abc}	91^{a}
Bacillus amyloliquefaciens subsp. *plantarum* ME 106 (BAP ME106)	5.16^{de}	6.04^{bcd}	0.53^{bc}	0.49^{bc}	63de	78^{abc}
Bacillus subtilis ME 105 (BA ME105)	3.50^{f}	6.96^{ab}c	0.40^{bcd}	0.39^{bcd}	29^{g}	75^{bcd}
Bacillus amyloliquefaciens subsp. *plantarum* ME 3 (BAP ME3)	3.52^{f}	6.40^{abcd}	0.42^{bcd}	0.42^{bcd}	40fg	75^{bcd}

Table (3): Effect of the interactions among *Rhizoctonia solani* and *Fusarium solani* soil treatments, *Bacillus* spp. and Rizolex fungicide seed treatments on fresh weight, dry weight and plant height of 6 weeks old Potato microtubers variety Sponta in greenhouse

Treatments	Fresh Weight (g/plant)	Dry Weight (g/plant)	Plant Height (cm)
Pathogenic Fungi			
R. solani	21.17	1.98^{a}	33.30
F. solani	21.07	1.49^{b}	31.75
F-test	ns	**	ns
Seed and Soil Treatments		***R. solani/F. solani***	
Check	25.5^{a}	3.4^{a}	36.2^{a}
Fungi	21.0b	0.5^{d}	31.8^{ab}
Rizolex	22.5^{ab}	1.5^{bc}	35.5^{a}
Bacillus amyloliquefaciens subsp. *plantarum* ME B8	21.5^{b}	1.1^{c}	33.2^{ab}
Paenibacillus polymyxa ME6	21.5^{b}	1.5^{bc}	32.8^{ab}
Bacillus amyloliquefaciens subsp. *Plantarum* ME B106	22.7^{ab}	1.7^{b}	30.8^{a}b
Bacillus subtilis ME 105	22.1^{ab}	1.6^{bc}	32.6^{ab}
Bacillus amyloliquefaciens subsp. *Plantarum* ME B3	12.2^{c}	0.6^{d}	27.3^{b}
F-test	**	**	*
Interaction among Pathogens and other treatments			
F-test	**	**	*

**, * = Significant differences at confidence level 95% and 99%, respectively.

Averages that share the same character or characters in the same column for each factor are not significantly different according to the least significant difference test.

Table (4): Effect of seed treatments with Rizolex fungicide or *Bacillus* spp. and soil infestation with *R. solani* or *Fusarium solani*, on fresh weight, dry weight and plant height of 6 weeks old Potato variety Sponta in greenhouse

Treatments	Fresh Weight (g/plant)		Dry Weight (g/plant)		Plant Height (cm)	
	R. solani	***F. solani***	***R. solani***	***F. solani***	***R. solani***	***F. solani***
Check	24.0ab	26.9a	3.48a	3.22a	37.0a	35.4a
Fungi	22.0b	20.1bcd	0.46f	0.54ef	32.4ab	31.2ab
Rizolex	21.3bc	23.7ab	1.64c	1.42cd	35.6a	35.4a
Bacillus amyloliquefaciens subsp. *Plantarum* ME B8	22.1b	21.0bc	0.68ef	1.58c	36.6a	29.8ab
Paenibacillus polymyxa ME6	21.4bc	21.9bc	1.82c	1.18cde	33.2ab	32.4ab
Bacillus amyloliquefaciens subsp. *Plantarum* ME B106	23.3ab	22.0b	2.60b	0.70ef	32.4ab	29.2ab
Bacillus subtilis ME 105	27.1a	17.4cd	2.72b	0.42f	34.8a	30.4ab
Bacillus amyloliquefaciens subsp. *Plantarum* ME B3	8.2e	16.1d	0.40f	0.82def	24.4b	30.2ab

Averages that share the same character or characters in the same column for each factor are insignificantly different according to the least significant difference test.

protectants to alfalfa seeds against *R. solani* and *F. solani* causing significantly increase in germination (81and 63%), compared to 53% in non-treated seeds in soil infected with *R. solani* (Table 2). Fresh and dry weights of alfalfa seedlings were also increased significantly in response to seed treatments with *P. polymyxa* ME6 or *B. amyloliquefacaciens* subsp. *plantarum* ME 106 when compared to seeds treated with Rizolex or non-treated (Table 2). This suggests that treating alfalfa seeds with any of these *P. polymyxa* ME 6 or *B. amyloliquefacaciens* subsp. *plantarum* ME 106 PGPR isolates can achieve the same protection against *R. solani* and *F. solani* as the use of fungicide Rizolex. There were insignificant differences between *F. solani* and *R. solani* in pathogenicity tests as determined by fresh weight and percent of germination of microtubers (Table 3). Also, differences related to treatments of the tuber with PGPR isolate or the Rizolex were mostly insignificant. This is due to the nature of interactions between the potato plant and its pathogens. Developing of *Fusarium* or *Rhizoctonia* diseases requires long time compared to the time assigned to this experiment (6 weeks). Therefore, the differences among the various treatments in potato were not as obvious as that in alfalfa screening tests. However, BAP ME105 and BAP ME106 isolates gave the best protection against *R. solani* as determined by fresh weight (27.1 g and 23.3 g/stem, respectively) compared to other treatments (Table 4). The highest values of fresh weight, dry weight and plant height were associated with non-treated potato microtubers sown in sterile soil (Table 4). The widely recognized mechanisms of biocontrol mediated by PGPB are competing for an ecological niche or a substrate, production of inhibitory allelochemicals and induction of induced systemic resistance (ISR) in host plants to a broad spectrum of pathogens and/or abiotic stresses (Haas *et al.*, 2002; Ryu *et al*., 2004 and Gray and Smith, 2005). Although, the mechanisms by which PGPR promote plant growth, are not yet fully understood, many different traits of this bacteria are responsible for growth promoting activities (Cattelan *et al.*, 1999; Bakker *et al.*, 2003 and 2007). Rhizobacteria promoted plant growth by several mechanisms. These include: production or changing the concentration of indoleacetic acid (IAA), giberillic acid, fixes nitrogen, suppress the growth of deleterious organisms of production of siderophore, b-13-glucanase, chitinases, antibiosis, cyanide, and phosphate solubilization and other nutrients (Cattelan *et al*., 1999).

In conclusion, most of the effective PGPR isolates (5 isolates) were identified as *Bacillus* spp. *B. amyloliquefaciens* subsp. *plantarum* ME 106 and *P. polymyxa* ME6 were the most effective against *R. solani* and *F. solani* and significantly increased fresh and dry weights of treated plants in most of the experiments. Use of PGPR strains for control of *R. solani* and *F. solani* was more effective on alfalfa and *R. solani* when compared to potato and *F. solani*. The results of this study recommend the conducting further research on the formulation and application of the *B. amyloliquefaciens* subsp. *plantarum* ME 106 and *P. polymyxa* in the field for controlling root diseases in general and *R. solani* and *F. solani* in particular.

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