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## Application of Some Organic Farming Methods to Enhancement The Growth and Production of Green Onion

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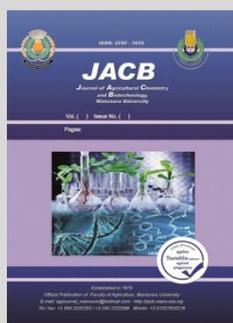


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### ABSTRACT

*In vitro* antagonistic effect of various PGPR namely *A. chroococcum*, *B. megaterium*, *B. circulans*, *B. subtilis*, *Ps. fluorescens* and *T. viride* against *F. solani* or *R. solani* was determined. Results observed that using all PGPR strains suppressed *F. solani* and *R. solani*. The inhibition zone increased by increasing incubation time. Under greenhouse condition, onion inoculation with investigated PGPR strains reduced disease severity of *F. solani* by rate ranging between (79.4-93.5%). However, they reduce disease severity of *R. solani* by rate ranging between (84.5-93.6%). Accordingly, the rate of disease incidence reduced by the range between (68.4-81.8%) and (65.5-81.8%) in case of infested with *F. solani* and *R. solani* respectively. Moreover, they had positive effect on plant defense enzymes, nutrients uptake and growth parameters. In farm of the Central Laboratory of Organic Agriculture, Giza station, ARC Giza, Egypt, the open field experiment was carried out to study the integrated effect of biofertilizing-PGPR and biocontrol agents with compost to improve green onion growth characteristics and yield under the organic agriculture system. During the experiment the soil microbial enzymes activity, total NPK and plant uptake, defense enzymes activity, growth characteristics and vegetative yield were estimated. Data obtained showed that onion treated with compost + biofertilizing-PGPR + *B. subtilis* + *T. viride* (T7) gave the highest values of in all estimated parameters except defense enzymes activity. So, it can be recommended as integrated fertilizing program to promote green onion growth, increase crop production, and decrease production costs.

**Keywords:** Organic farming, green onion, plant growth-promoting rhizobacteria, soil-borne fungi



### INTRODUCTION

Onion (*Allium cepa* L.) is a very important and widespread vegetable crop grown all around the world (Sidhu *et al.*, 2019). It is one of the oldest bulb vegetables, which its bulb has been found in ancient Egypt (Abd Alla, 2015). Regarding its global production, over 3.6 million hectares of onions are grown annually around the world and about 170 countries cultivate this crop for domestic use. Egyptian onion crop is famous all over the world due to its superior quality and early appearance in European markets (Hussein *et al.*, 2007).

Conventional agriculture may provide short-term gains in production, but in most cases, it is not sustainable in the long term, undermines the viability of small farm units, and does not guarantee safe foods as well as cause great environmental harms (Migliorini and Wezel 2017).

The change from conventional to organic farming is accompanied by changes in soil chemical and biological properties and processes that affect soil fertility. Fundamental differences, both qualitative and quantitative, in the flow and processing of nutrients result from soil amendment, plant community structure, tillage, and elimination of synthetic fertilizers and pesticides (Lee, 2010). Generally, organic agricultural practices aim to enhance biodiversity, biological cycles,

and soil biological activity to achieve an optimal system that is socially, ecologically, and economically sustainable (Nejadkoorki, 2012 and Migliorini and Wezel 2017 and Riaz *et al.*, 2021). Many plant growth-promoting rhizobacteria strains (PGPRs) that have been identified have seen a great boost, mainly because the role of the rhizosphere as an ecological unit has gained importance in the functioning of the biosphere and also because mechanisms of action of PGPR have been deeply studied. PGPR shows an important role in the sustainable agriculture industry. The increasing demand for crop production with a significant reduction of synthetic chemical fertilizers and pesticides use is a big challenge nowadays (Pravin *et al.*, 2016).

In general soil microbial communities are often difficult to characterize, mainly because of their immense phenotypic and genotypic diversity. The rhizosphere is the soil nearest area to the root system that inhabits the microorganisms and capable to colonize very well to the roots. These microorganisms are stated as plant growth-promoting rhizobacteria (PGPR). Plants depend upon valuable interactions between these microbes and roots for growth promotion, nutrient availability, and disease suppression and they fulfill vital functions for plant growth (Khan *et al.*, 2019).

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Root-rot diseases caused by soil-borne fungi are the most important diseases of many crops. (Bodah, 2017). In onion (*Allium cepa* L.), *Fusarium* root rot caused by *Fusarium solani* f. sp. *phaseoli* disease severity is increased by environmental factors that stress the plant (Karen et al, 2007). Also, Rhizoctonia root rot caused by *Rhizoctonia solani* is a frequent disease of many crops such as bean (Schroeder and Paulitz 2012). This study aimed to investigate the effects of investigated PGPR strains and organic fertilizer on onion during organic production as well as their effects on enhancing the nutrient efficiency and root rot diseases caused by soil-borne fungi.

## MATERIALS AND METHODS

### PGPR strains

The strains namely, nitrogen fixing bacteria *Azotobacter chroococcum* CLOA A27 (Azoto). *Bacillus megaterium* var. *phosphaticum* CLOA Bm 3 (Bm) was used as phosphate dissolving bacteria (PDB). *Bacillus circulans* CLOA Bc 4(Bc) was used as potassium solubilization bacteria (PSB). *Bacillus subtilis* CLOA Bs 12 (Bs), *Pseudomonas fluorescens* CLOA Pf 5 (Pf) and *Trichoderma viride* CLOA Tri 24 (Tv) were used as biocontrol agents. were kindly obtained from Central Lab. of Organic Agriculture, Agricultural Research Center, Giza, Egypt, they were purified and stored at 4°C. *Fusarium solani* CLOA Fs 10 and *Rhizoctonia solani* CLOA Rhizo 6 were kindly obtained from Central Lab. Of Organic Agriculture, Agricultural Research Center, Giza, Egypt, were purified and maintained on potato dextrose agar medium (Ricker and Ricker, 1936).

### Antagonistic activity of PGPRs against soil-borne fungi

According to (Berg et al, 2005). PGPR strains was inoculated on 9-cm Petri dish containing specific media for different tested PGPR in circular line using 4-cm diameter Petri dish by dipping in 24-h-old PGPR cultures and incubated for 24-h at 30 °C. Then, a five-mm<sup>2</sup> disk of a pure seven days culture from either *F. solani* or *R. solani* was placed at the central of circular line and incubated ageing at 25±2 °C for 3, 5, and 7 days. At the end of the incubated period, the growth (fungal growth) was measured and compared to control (un-inoculated with PGPRs). Three Petri dishes were used as replicates for each treatment. Obtained results were expressed as means of radii of fungal growth (mm) and inhibition percentage was calculated according to (Tariq et al, 2010). Concerning the estimation of antagonism among *Trichoderma viride* and pathogenic soil-borne fungi, a five-mm<sup>2</sup> disk of 7-days old culture from *Trichoderma viride* was placed on the edge of Petri dishes while, either *F. solani* or *R. solani* were placed in the other edge of Petri dishes. Three Petri dishes were used as replicates for each treatment. Percentages of reduction in pathogenic fungal mycelial growth were calculated (Gebily, 2015)

### Preparation of PGPR and pathogens inocula

PGPR inocula were prepared using specific media for each bacterium (PGPR). While, the pathogenic fungus strain inoculum of either *F. solani* or

*R. solani* was prepared on Corn sand meal medium according to (Abd-El-Moity, 1985).

### Effect of PGPRs on onion production under greenhouse condition

A greenhouse experiment was layout at the greenhouse of Central Lab. of Organic Agriculture, Agricultural Research Center, Giza, Egypt during the two successive winter seasons, (2017–2018) to determine the most efficient PGPR as biocontrol for soil-borne pathogenic fungi *F. solani* or *R. solani* on onion growth performance and production. Sterilized plastic pots (30 cm diameter) were filled with sterilized soil (mixing of silt clay, peat moss, and compost at the rate of 200:200:1). The analysis of soil, peat moss, and plant-animal compost presented in Table 1, 2 and 3.

**Table 1. Soil particles size distribution and chemical analysis of the used soil.**

Property	Values
Soil particles size distribution	
Sand %	9.8
Silt %	59.1
Clay %	31.1
Texture	Silt clay
Chemical analysis	
pH (in soil paste)	7.5
E.C (dsm <sup>2</sup> )	3.1
Saturation percentage (%)	53
CEC emol/kg soil	38.17
CaCO <sub>3</sub> %	0.70
Organic matter %	1.70
Available N ppm	17
Available P ppm	10.4
Available K ppm	182
Available Fe ppm	4.1
Available Zn ppm	0.5
Available Mn ppm	0.1
Available Cu ppm	0.2
Soluble cations and anions (meqL <sup>-1</sup> )	
Ca <sup>++</sup>	10.3
Mg <sup>++</sup>	8.7
Na <sup>+</sup>	18.3
K <sup>+</sup>	1.4
CO <sub>3</sub> <sup>-</sup>	8.8
Cl <sup>-</sup>	14.2
SO <sub>4</sub> <sup>-</sup>	15.7

**Table 2. Chemical analysis of the used peat moss.**

Property	Values
Moisture content (%)	16.9
Water holding capacity (%)	468.8
pH	4.50
E C (dsm <sup>2</sup> )	0.56
Organic carbon (%)	46.68
Organic matter (%)	80.72
Total N (%)	1.24
C/N ratio	37.64

The soil was pre-mixed with *F. solani* or *R. solani* inoculum at a rate of 4%. Plastic pots were sterilized by immersing in 5 % formalin solution (38 %) for 15 minutes and covered overnight with plastic sheets, then left to dry in the open air. Sterilization of experimental soil was carried out using H<sub>2</sub>O<sub>2</sub> (30 %). Infested pots were irrigated for seven days before transplanting. Four transplants of onion “Giza red” were transplanted in each pot and five replicate pots were specified for each treatment in a completely randomized

experimental design. Before cultivation, all transplants, except control, were soaked on cell suspension of each PGPRs strain at a rate 108.ml<sup>-1</sup>. Regarding *T. viride* the spore's suspension was used at a rate 104.ml<sup>-1</sup>. The boost prepared inocula were added after 21 and 55 days after transplanting (DAT) at a rate of 10 ml pot<sup>-1</sup>. Pots were kept under greenhouse conditions until the end of the experiment.

The experiment included the following treatments: infestation with either *F. solani* or *R. solani*, inoculation with *A. chroococcum* + compost + *F. solani* or *R. solani*, inoculation with *B. megaterium* + compost + *F. solani* or *R. solani*, inoculation with *B. circulans* + compost + *F. solani* or *R. solani*, inoculation with *B. subtilis* + compost + *F. solani* or *R. solani*, inoculation with *P. fluorescens* + compost + *F. solani* or *R. solani*, and inoculation with *T. viride* + compost + *F. solani* or *R. solani*. At the end of the experiment (90 DAT), the following assessments were done.

**Table 3. Physical and chemical properties of used compost in this study.**

Parameter	value
Bulk density (kg M <sup>3</sup> )	670
Moisture content%	29
pH	7.4
EC (dsm <sup>2</sup> )	5.6
Organic matter %	47.7
Organic carbon %	27.7
Ash %	71
Total nitrogen %	1.5
C/N ratio	18.4:1
Total P	0.7
Total K	0.8
Fe ppm	1400
Zn ppm	1.4
Mn ppm	300
Cu ppm	80
Nematoda	Nil
Seed weed	Nil
<i>E. coli</i>	Nil

**Diseases assessment**

- **Disease scale:** Depending on visible symptoms, a scale from zero (healthy) to 5 (infected) was recorded to measure the diseased areas on all treatments in the greenhouse.
- **Disease incidence:** After 60 days of transplanting, onion plants were rated for disease incidence as a present by methods described by (Liu *et al*, 1995).

**Field experiment**

Two successive field experiments were carried out at a farm of the Central Laboratory of Organic Agriculture, Giza station, ARC Giza, Egypt, during the first week of February (2017–2018). This experiment was designed to obtain the superior combination of biofertilizing-PGPR and biocontrol agent with compost to improve onion growth characteristics and yield in the open field under the organic agriculture system. After preparing the soil for cultivation, the treatments were distributed in a randomized complete block design with three replicates. The plot area was 10.5 m<sup>2</sup> (3 x 3.5 m). The experiment included the following treatments: compost (recommended dose), compost + *A. chroococcum* + *B. megaterium* + *B. circulans*, Compost + *A. chroococcum* + *B. megaterium* + *B. circulans* + *B.*

*subtilis*, compost + *A. chroococcum* + *B. megaterium* + *B. circulans* + *P. fluorescens*, compost + *A. chroococcum* + *B. megaterium* + *B. circulans* + *T. viride*, compost + *A. chroococcum* + *B. megaterium* + *B. circulans* + *B. subtilis* + *P. fluorescens*, compost + *A. chroococcum* + *B. megaterium* + *B. circulans* + *B. subtilis* + *T. viride*, compost + *A. chroococcum* + *B. megaterium* + *B. circulans* + *P. fluorescens* + *T. viride* and compost + *A. chroococcum* + *B. megaterium* + *B. circulans* + *B. subtilis* + *P. fluorescens* + *T. viride*.

**Cultivation process**

Compost was added to the soil at a rate of recommended full dose (5 ton/fed) before transplanting. Before cultivation, onion transplants were soaked with various PGPRs inocula for 30 seconds before planting. During irrigation, biofertilizers and biocontrol inocula were added at the rate of 10L.fed<sup>-1</sup>. The boost addition of the previous inocula was repeated after 21 and 55 DAT. All agricultural operations related to onion production were applied according to the recommendations of the Ministry of Agriculture and Land Reclamation.

**Microbial enzymes activity**

Soil samples were taken from the onion rhizosphere for measuring the microbial enzymatic activities at 30, 60, and 90 DAT. The activities of dehydrogenase (DH) and alkaline phosphatase (Alp) were measured using a spectrophotometer (SCO-Tech, SPUV-19, Germany) at 464 and 400 nm, respectively, as described by Schinner *et al*, (1996). However, nitrogenase (N<sub>2</sub>-ase) activity was measured as previously mentioned with some modification by Okafor and MacRae (1973).

**Defense enzyme activities in plants:**

Peroxidase activity in plants was determined according to the method described by Allam and Hollis (1972). polyphenol oxidase activity in plants was determined according to the method described by Matta and Dimond (1963).

**Plant chemical analyses:**

Nitrogen content was determined according to the method described by A.O.A.C. (1980). Phosphorus content was determined according to A.PH.A. (1992). Potassium content was determined according to the method described by Dewis and Freitas (1970).

**Plant growth characteristics and yield**

Samples were taken after 60 days of planting to determine the plant height (cm), leaves number, dry weights of the plant (g/plant), total soluble solids, and total chlorophylls were measured according to (A.O.C.A. 2003). Finally, the biological yield of onion (ton/fed.) was determined.

**Statistical analysis**

The data were statistically analyzed according to the procedures outlined by Gomez and Gomez (1984). For comparison between means, Duncan's multiple range test was used (Duncan, 1955). Means followed by the same alphabetical letters were not significantly different at 5% level of significance.

**RESULTS AND DISCUSSIONS**

**Antagonistic effect of PGPR on soil-borne disease fungi**

Antagonistic effect of various PGPRs namely *A. chroococcum*, *B. megaterium*, *B. circulans*, *B. subtilis*, *Ps.*

*fluorescens* and *T. viride* against *F. solani* or *R. solani* was done. Results shown in Fig 1 indicated that using all PGPR strains suppressed *F. solani* and *R. solani*. The inhibition zone increased by increasing incubation time (3, 5 and 7 days). These results may be due to high amounts of antibiotic or antagonistic substances produced by PGPR strains. These results are in agreement with (Jan *et al*, 2011, Chen *et al*, 2012, Lukkani and Reddy 2014, and Susilowati and Syekhfani 2014) who found that the main compounds produced by PGPRs are siderophores, cyanogenic substances, antibiotics, and some weakly organic acids. These substances reduced the mycelium formation, spore germination and caused hyphal lysis of *F. solani* and *R. solani*. Moreover, both *B. subtilis* and *Ps. fluorescens* recorded higher values of inhibition zone followed by *B. megaterium* and *B. circulans*. While the

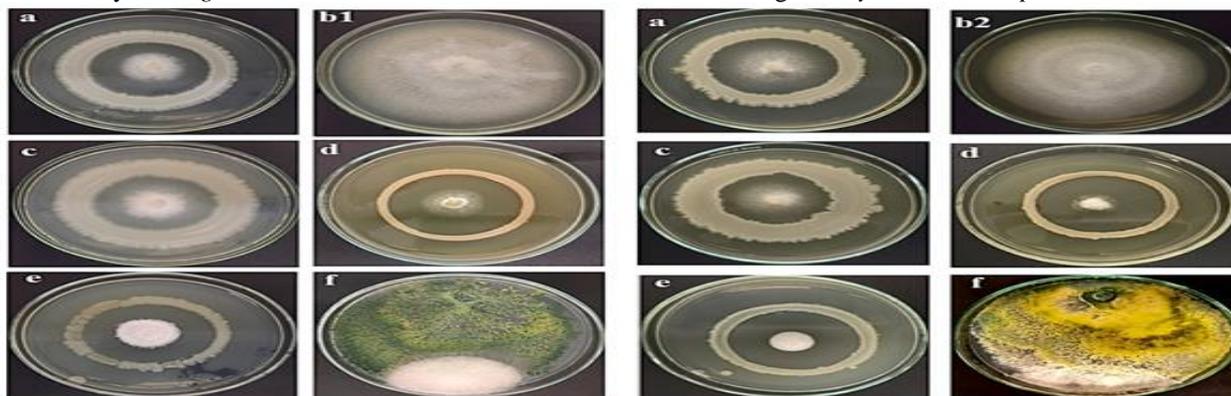


Fig. 1. Effect of PGPR strains against pathogenic fungi

a: *B. megaterium*; b1: *F. solani*; b2: *R. solani*; c: *B. circulans*; d: *P. fluorescens*; e: *B. subtilis*; f: *Trichoderma viride*.

**Effect of PGPRs on pathogenic fungi under greenhouse condition**

This experiment was carried out during the two successive winter seasons, (2017 – 2018) on green onion to evaluate the efficiency of PGPR strains on the causal agent of root disease fungi.

**Effects on disease severity and disease incidence**

Data in Table (4) indicated that significant decreases were found in disease severity and disease incidence of onion that inoculated with various PGPR strains and infested with *F. solani* or *R. solani* compared with uninoculated one. Moreover, a treatment that non-infested with *F. solani* or *R. solani* gave lower percentage values compared to infested one.

The inoculation with *B. subtilis*, *P. fluorescens* and *B. megaterium* scored a lower percentage of disease severity of onion compared to other tested PGPR strains either soil infested with *F. solani* or *R. solani*. Regarding disease incidence, the treatments inoculated with *B. megaterium* and *P. fluorescens* gave a lower percentage of disease incidence compared to other tested PGPR strains in the soil infested with *F. solani*. While, the inoculation with *B. subtilis*, *P. fluorescens* and *B. megaterium* gave lower values in case of infested with *R. solani*. The lowest percentage of disease severity and disease incidence of onion was recorded in the case of inoculation with *B. subtilis*. While the highest percentage for disease severity and disease incidence of onion were found in infested soil with pathogenic fungi solely

lowest values were showed in the case of *T. viride* against *F. solani* and *R. solani*. The lower values of inhibition zone in case of *T. viride* against *F. solani* and *R. solani* may be due to parasitism, this process including four steps namely; chemotropism, recognition, attachment, cell wall degradation, and penetration are the steps involved in mycoparasitism. For example, *Trichoderma* spp. parasitize *Rhizoctonia* spp., the soil and the root pathogen (Manoharachary *et al*, 2020)

*In vitro* antagonism of *T. viride* was confirmed against *Cylindrocladium parvum* infecting Eucalyptus. Mycoparasitic interaction was assessed by following Bell’s ranking (Bell *et al*. 1982). It was detected that after 5 days the pathogen was significantly suppressed recording to Bell’s ranking No.4. Results also indicated the antagonism of *T. viride* against *Cylindrocladium parvum*.

Table 4. Effect of PGPR inoculation on disease severity and disease incidence of onion in soil infested with *F. solani* and *R. solani*.

	Disease severity (%)		Disease incidence (%)	
	S <sub>1</sub>	S <sub>2</sub>	S <sub>1</sub>	S <sub>2</sub>
Non-infested soil	6.67 <sup>d</sup>	5.00 <sup>e</sup>	16.67 <sup>c</sup>	12.50 <sup>d</sup>
Soil infested with <i>F. solani</i>	90.00 <sup>a</sup>	89.17 <sup>a</sup>	70.83 <sup>a</sup>	66.67 <sup>a</sup>
<i>F. solani</i> + Azoto	7.50 <sup>d</sup>	9.17 <sup>d</sup>	20.83 <sup>b</sup>	16.67 <sup>c</sup>
<i>F. solani</i> + Bm	6.67 <sup>d</sup>	8.33 <sup>d</sup>	16.67 <sup>c</sup>	16.67 <sup>c</sup>
<i>F. solani</i> + Bc	10.83 <sup>c</sup>	12.50 <sup>c</sup>	16.67 <sup>c</sup>	16.67 <sup>c</sup>
<i>F. solani</i> + Bs	5.83 <sup>d</sup>	6.67 <sup>de</sup>	12.50 <sup>d</sup>	12.50 <sup>d</sup>
<i>F. solani</i> + Pf	6.67 <sup>d</sup>	8.33 <sup>d</sup>	16.67 <sup>c</sup>	12.50 <sup>d</sup>
<i>F. solani</i> + Tv	15.83 <sup>b</sup>	18.33 <sup>b</sup>	22.50 <sup>d</sup>	20.83 <sup>d</sup>
Non-infested soil	5.83 <sup>d</sup>	5.83 <sup>c</sup>	12.50 <sup>d</sup>	12.50 <sup>f</sup>
Soil infested with <i>R. solani</i>	92.50 <sup>a</sup>	91.67 <sup>a</sup>	75.00 <sup>a</sup>	62.50 <sup>a</sup>
<i>R. solani</i> + Azoto	7.60 <sup>cd</sup>	11.67 <sup>b</sup>	16.67 <sup>c</sup>	25.00 <sup>c</sup>
<i>R. solani</i> + Bm	10.83 <sup>b</sup>	6.67 <sup>c</sup>	16.67 <sup>c</sup>	20.83 <sup>d</sup>
<i>R. solani</i> + Bc	9.17 <sup>bc</sup>	14.17 <sup>b</sup>	16.67 <sup>c</sup>	29.17 <sup>b</sup>
<i>R. solani</i> + Bs	7.50 <sup>cd</sup>	5.83 <sup>c</sup>	12.50 <sup>d</sup>	12.50 <sup>f</sup>
<i>R. solani</i> + Pf	9.17 <sup>bc</sup>	8.33 <sup>c</sup>	16.67 <sup>c</sup>	16.67 <sup>e</sup>
<i>R. solani</i> + Tv	11.33 <sup>b</sup>	11.83 <sup>b</sup>	26.67 <sup>b</sup>	20.83 <sup>d</sup>

Bm: *B. megaterium*; Bc: *B. circulans*; Bs: *B. subtilis*; Pf: *P. fluorescens*; Tv: *T. viride*; S<sub>1</sub>: season one; S<sub>2</sub>: season two. A column followed by the same letter are not significantly different at P= 0.05 when compared by Duncan test

From obtained data, the inoculation with various PGPRs strain reduced disease severity of *F. solani* by rate ranging between (82.4-93.5%) and (79.4-92.5) in the first and second seasons respectively. While they reduce disease severity of *R. solani* by rate ranging between (87.8-91.9%) and (84.5-93.6%) in the first and second seasons respectively. Accordingly the rate of disease incidence reduced in the range between (68.4-81.8%) and (65.5-

81.8%) in case of infested with *F. solani* and *R. solani* respectively.

The decrement of disease severity and disease incidence in case of inoculation with PGPRs or biocontrol agent observed their role in suppressing the pathogen infection that may happen by several mechanisms. These results are in harmony with (Glic, 2012, Liu *et al*, 2017 and Karthika *et al*, 2020) who reported that PGPR act, as biocontrol agents and these is considered the major indirect mechanisms of plant growth promotion by rhizobacteria and they add PGPR major modes of biocontrol activity include competition for nutrients, niche exclusion, induced systemic resistance and antifungal metabolites

**Effects on defense enzymes activity**

Data tabulated in Table 5 showed that there are significant increases in the induction of defense enzymes (peroxidase, polyphenoloxidase) of onion that inoculated with various PGPR strains compared with uninoculated one. Moreover, there are no significant differences in plant enzymes content between a plant that is cultivated in infested or non-infested soil. Also, data revealed that the inoculation with *B. subtilis*, *P. fluorescens*, or *B. circulans* scored higher values of peroxidase activity of onion compared to other tested PGPR strains in case of infestation with *F. solani*

Concerning polyphenoloxidase activity data showed that treatments inoculated with *B. subtilis*, *P. fluorescens*, or *B. megaterium* gave higher values compared to other tested PGPR strains. The highest values of peroxidase and polyphenoloxidase activity of onion were recorded with plants inoculated with *B. subtilis*. These data are in harmony with those obtained by Elsharkawy and El-Khateeb (2019) who reported that plant enzyme activity was more activation and plant produce higher values of peroxidase and polyphenoloxidase by application different PGPR treatments.

**Table 6. Effect of PGPR inoculation on microbial enzymes activity of onion rhizosphere soil infested with *F. solani* and *R. solani*.**

Treatments	Dehydrogenase activity (µg TPF.g <sup>-1</sup> dry soil. 24 <sup>-1</sup> )					
	S1			S2		
	30	60	90 DAT	30	60	90 DAT
Non-infested soil with <i>F. solani</i>	2.6 <sup>e</sup>	17.86 <sup>f</sup>	14.52 <sup>l</sup>	11.23 <sup>f</sup>	19.5 <sup>g</sup>	18.54 <sup>e</sup>
Soil infested with <i>F. solani</i>	11.1 <sup>d</sup>	20.34 <sup>f</sup>	18.50 <sup>e</sup>	16.60 <sup>f</sup>	24.2 <sup>f</sup>	27.06 <sup>d</sup>
<i>F. solani</i> + Azoto	16.3 <sup>c</sup>	44.23 <sup>c</sup>	27.08 <sup>c</sup>	21.42 <sup>b</sup>	42.16 <sup>c</sup>	49.60 <sup>c</sup>
<i>F. solani</i> + Bm	19.1 <sup>b</sup>	49.16 <sup>b</sup>	27.94 <sup>c</sup>	18.90 <sup>c</sup>	47.46 <sup>b</sup>	51.73 <sup>bc</sup>
<i>F. solani</i> + Bc	15.1 <sup>c</sup>	35.94 <sup>d</sup>	32.81 <sup>b</sup>	12.68 <sup>bc</sup>	38.75 <sup>d</sup>	52.60 <sup>bc</sup>
<i>F. solani</i> + Bs	19.42 <sup>b</sup>	27.16 <sup>c</sup>	28.35 <sup>c</sup>	21.75 <sup>b</sup>	27.34 <sup>f</sup>	54.41 <sup>ab</sup>
<i>F. solani</i> + Pf	30.97 <sup>a</sup>	53.57 <sup>a</sup>	44.85 <sup>a</sup>	24.68 <sup>a</sup>	69.09 <sup>a</sup>	57.35 <sup>a</sup>
<i>F. solani</i> + Tv	11.49 <sup>d</sup>	35.64 <sup>d</sup>	22.91 <sup>d</sup>	14.90 <sup>e</sup>	30.72 <sup>e</sup>	49.98 <sup>c</sup>
Non-infested soil with <i>R. solani</i>	6.86 <sup>f</sup>	9.86 <sup>f</sup>	23.08 <sup>d</sup>	10.72 <sup>d</sup>	24.75 <sup>e</sup>	15.87 <sup>f</sup>
Soil infested with <i>R. solani</i>	11.0 <sup>e</sup>	20.16 <sup>e</sup>	15.83 <sup>e</sup>	8.46 <sup>e</sup>	25.90 <sup>e</sup>	16.81 <sup>f</sup>
<i>R. solani</i> + Azoto	12.34 <sup>cde</sup>	37.38 <sup>c</sup>	25.66 <sup>d</sup>	12.09 <sup>c</sup>	34.53 <sup>d</sup>	22.69 <sup>e</sup>
<i>R. solani</i> + Bm	12.53 <sup>cd</sup>	22.72 <sup>e</sup>	23.00 <sup>d</sup>	11.90 <sup>c</sup>	48.57 <sup>c</sup>	30.08 <sup>c</sup>
<i>R. solani</i> + Bc	12.23 <sup>de</sup>	31.31 <sup>d</sup>	24.56 <sup>d</sup>	10.75 <sup>d</sup>	52.16 <sup>bc</sup>	27.14 <sup>d</sup>
<i>R. solani</i> + Bs	19.97 <sup>b</sup>	40.6 <sup>b</sup>	42.94 <sup>b</sup>	15.83 <sup>b</sup>	53.27 <sup>b</sup>	34.35 <sup>b</sup>
<i>R. solani</i> + Pf	31.27 <sup>a</sup>	47.75 <sup>a</sup>	45.62 <sup>a</sup>	19.34 <sup>a</sup>	58.01 <sup>a</sup>	38.00 <sup>a</sup>
<i>R. solani</i> + Tv	13.75 <sup>c</sup>	31.97 <sup>d</sup>	37.37 <sup>c</sup>	12.90 <sup>c</sup>	50.83 <sup>bc</sup>	26.19 <sup>d</sup>

Abbreviations as those stated for Table (4)

Regarding soil infested with *R. solani*, the inoculation with *P. fluorescens* or *B. subtilis* had a great effect on improving the activity of dehydrogenase enzyme compared to another treatments. The highest values of DH were recorded with plants inoculated with *P. fluorescens*, *A. chroococcum*, or *B. megaterium* respectively. In all

**Table 5. Effect of PGPR inoculation on peroxidase and polyphenoloxidase activity of onion soil infested with *F. solani* and *R. solani* (as absorbance. g<sup>-1</sup> fresh leaves).**

Treatments	Peroxidase		Polyphenoloxidase	
	S <sub>1</sub>	S <sub>2</sub>	S <sub>1</sub>	S <sub>2</sub>
Non-infested soil with <i>F. solani</i>	0.434 <sup>e</sup>	0.483 <sup>d</sup>	0.085 <sup>e</sup>	0.088 <sup>f</sup>
Soil infested with <i>F. solani</i>	0.429 <sup>e</sup>	0.465 <sup>d</sup>	0.091 <sup>de</sup>	0.095 <sup>ef</sup>
<i>F. solani</i> + Azoto	0.708 <sup>bc</sup>	0.600 <sup>bc</sup>	0.105 <sup>c</sup>	0.118 <sup>c</sup>
<i>F. solani</i> + Bm	0.694 <sup>c</sup>	0.563 <sup>c</sup>	0.110 <sup>c</sup>	0.122 <sup>c</sup>
<i>F. solani</i> + Bc	0.763 <sup>b</sup>	0.685 <sup>a</sup>	0.100 <sup>cd</sup>	0.114 <sup>cd</sup>
<i>F. solani</i> + Bs	1.096 <sup>a</sup>	0.687 <sup>a</sup>	0.166 <sup>a</sup>	0.179 <sup>a</sup>
<i>F. solani</i> + Pf	1.045 <sup>a</sup>	0.708 <sup>a</sup>	0.133 <sup>b</sup>	0.143 <sup>b</sup>
<i>F. solani</i> + Tv	0.623 <sup>d</sup>	0.654 <sup>ab</sup>	0.103 <sup>cd</sup>	0.102 <sup>de</sup>
Non-infested soil with <i>R. solani</i>	0.437 <sup>d</sup>	0.436 <sup>d</sup>	0.099 <sup>c</sup>	0.099 <sup>cd</sup>
Soil infested with <i>R. solani</i>	0.424 <sup>d</sup>	0.493 <sup>d</sup>	0.086 <sup>c</sup>	0.092 <sup>d</sup>
<i>R. solani</i> + Azoto	0.778 <sup>b</sup>	0.659 <sup>c</sup>	0.139 <sup>b</sup>	0.085 <sup>d</sup>
<i>R. solani</i> + Bm	0.794 <sup>ab</sup>	0.745 <sup>b</sup>	0.147 <sup>ab</sup>	0.109 <sup>bc</sup>
<i>R. solani</i> + Bc	0.793 <sup>ab</sup>	0.739 <sup>b</sup>	0.128 <sup>c</sup>	0.106 <sup>bc</sup>
<i>R. solani</i> + Bs	0.642 <sup>c</sup>	0.688 <sup>bc</sup>	0.157 <sup>a</sup>	0.120 <sup>a</sup>
<i>R. solani</i> + Pf	0.851 <sup>a</sup>	0.922 <sup>a</sup>	0.148 <sup>ab</sup>	0.117 <sup>ab</sup>
<i>R. solani</i> + Tv	0.776 <sup>b</sup>	0.699 <sup>bc</sup>	0.145 <sup>ab</sup>	0.105 <sup>bc</sup>

Abbreviations as those stated for Table (4). Peroxidase activity was expressed as the increase in absorbance at 470 nm/g fresh weight/ min. Polyphenoloxidase activity was expressed as the increase in absorbance at 475 nm/g fresh weight/minute.

**Effect on dehydrogenase activity in the rhizosphere**

To evaluate the role of PGPR strains in plant protection and growth enhancement, their presence and activities should be determined. Data in Table (6) revealed that there are significant increases in DH activity of onion rhizosphere that inoculated with various PGPR strains compared with uninoculated one.

In infested soil with *F. solani*, the inoculation with *P. fluorescens* or *B. megaterium* scored higher values of DH activity compared to other tested PGPR strains in two seasons. The highest values of DH, were recorded with plants inoculated with *P. fluorescens*.

treatments, the activity of three estimated enzymes was increased gradually until 60 DAT and decreased again at 90 DAT. The increment of DH activity may be due to the boost addition of PGPR inocula.

The enhancement of DH activity in onion rhizosphere during inoculation with PGPR are in

agreement with results obtained by Hui *et al.*, (2004), Li *et al.*, (2006), Mikanová *et al.*, (2009), Nosheen and Bano (2014) and Järvan *et al.*, (2014) who reported that application of various PGPR treatments can enhance and increase the activity of some soil enzymes as well as improve soil nutrition.

**Effect on onion growth parameters**

Data in Table (7) indicated that significant increases were found in plant height, leaf number, dry weight, and total chlorophyll of onion in all inoculated treatments with various PGPR strains compared with non-infested one. Furthermore, the treatment that non-infested with *F. solani* or *R. solani* gave higher values compared to infested one. Among The inoculated treatments, the inoculation with *A. chroococcum*, *B. megaterium* *B. circulans* or *B. subtilis* scored higher values in all vegetative growth parameters and total chlorophyll of onion compared to other tested

PGPR strains. The highest value of plant height, leaves number, dry weight, and total chlorophyll of onion were recorded with inoculated plants with *A. chroococcum* in both fungal infections. While the lowest values of all estimated parameters were found in onions cultivated in infested soil followed by non-infested one.

The significant increases in all growth parameters in inoculated plants with PGPRs reveal their importance not only in protecting the plants against pathogenic infection but also in enhancing plant growth. These results are agree with those obtained by (Abou Zaid *et al.*, 2011, Ahmed and El-Araby, 2012 and Li *et al.*, 2020) who reported that application of PGPRs with or without organic fertilizers led to score higher values of vegetative growth parameters for different vegetables and field crops as compared to untreated ones.

**Table 7. Effect of PGPRs inoculation on growth characters of onion in soil infested with *F. solani* and *R. solani*.**

Treatments	Plant height (cm/plant)		Leaves number.plant <sup>-1</sup>		Dry weight (g/plant)		Total Chlorophyll (mg/g)	
	S <sub>1</sub>	S <sub>2</sub>	S <sub>1</sub>	S <sub>2</sub>	S <sub>1</sub>	S <sub>2</sub>	S <sub>1</sub>	S <sub>2</sub>
Non-infested soil with <i>F. solani</i>	35.7 <sup>d</sup>	32.8 <sup>d</sup>	4.6 <sup>d</sup>	3.4 <sup>e</sup>	1.10 <sup>d</sup>	1.26 <sup>e</sup>	61.9 <sup>d</sup>	58.7 <sup>c</sup>
Soil infested with <i>F. solani</i>	33.1 <sup>d</sup>	33.9 <sup>d</sup>	3.6 <sup>e</sup>	2.9 <sup>e</sup>	0.74 <sup>e</sup>	1.27 <sup>e</sup>	47.4 <sup>e</sup>	49.5 <sup>d</sup>
<i>F. solani</i> + Azoto	49.3 <sup>a</sup>	47.8 <sup>a</sup>	9.0 <sup>a</sup>	10.1 <sup>a</sup>	3.79 <sup>a</sup>	3.80 <sup>a</sup>	75.7 <sup>a</sup>	73.4 <sup>a</sup>
<i>F. solani</i> + Bm	46.7 <sup>ab</sup>	47.2 <sup>a</sup>	8.9 <sup>a</sup>	8.8 <sup>b</sup>	3.80 <sup>a</sup>	3.97 <sup>a</sup>	68.7 <sup>b</sup>	69.1 <sup>ab</sup>
<i>F. solani</i> + Bc	46.7 <sup>ab</sup>	46.4 <sup>ab</sup>	7.4 <sup>b</sup>	7.4 <sup>c</sup>	3.26 <sup>b</sup>	3.22 <sup>bc</sup>	67.3 <sup>bcd</sup>	67.4 <sup>b</sup>
<i>F. solani</i> + Bs	49.2 <sup>a</sup>	43.3 <sup>b</sup>	8.8 <sup>a</sup>	7.9 <sup>c</sup>	3.23 <sup>b</sup>	3.43 <sup>b</sup>	68.2 <sup>bc</sup>	67.6 <sup>b</sup>
<i>F. solani</i> + Pf	40.9 <sup>c</sup>	39.5 <sup>c</sup>	6.7 <sup>c</sup>	6.7 <sup>d</sup>	2.38 <sup>c</sup>	3.04 <sup>cd</sup>	63.5 <sup>bcd</sup>	60.4 <sup>c</sup>
<i>F. solani</i> + Tv	43.0 <sup>bc</sup>	43.2 <sup>b</sup>	6.8 <sup>c</sup>	6.8 <sup>d</sup>	2.39 <sup>c</sup>	2.88 <sup>d</sup>	62.5 <sup>cd</sup>	60.8 <sup>c</sup>
Non-infested soil with <i>R. solani</i>	27.2 <sup>c</sup>	26.5 <sup>d</sup>	4.2 <sup>e</sup>	3.6 <sup>e</sup>	1.20 <sup>d</sup>	1.02 <sup>e</sup>	66.1 <sup>bc</sup>	57.6 <sup>e</sup>
Soil infested with <i>R. solani</i>	30.4 <sup>c</sup>	27.2 <sup>d</sup>	3.6 <sup>f</sup>	3.4 <sup>e</sup>	1.23 <sup>d</sup>	0.99 <sup>e</sup>	46.7 <sup>d</sup>	47.0 <sup>f</sup>
<i>R. solani</i> + Azoto	46.3 <sup>a</sup>	48.5 <sup>a</sup>	9.3 <sup>a</sup>	8.8 <sup>a</sup>	3.32 <sup>bc</sup>	3.84 <sup>a</sup>	73.7 <sup>a</sup>	72.8 <sup>a</sup>
<i>R. solani</i> + Bm	46.2 <sup>a</sup>	47.1 <sup>ab</sup>	8.8 <sup>a</sup>	8.2 <sup>b</sup>	3.61 <sup>a</sup>	3.73 <sup>a</sup>	70.6 <sup>ab</sup>	67.8 <sup>abc</sup>
<i>R. solani</i> + Bc	44.7 <sup>ab</sup>	44.1 <sup>bc</sup>	7.8 <sup>b</sup>	7.3 <sup>cd</sup>	3.46 <sup>ab</sup>	3.29 <sup>b</sup>	65.1 <sup>bc</sup>	61.9 <sup>de</sup>
<i>R. solani</i> + Bs	41.4 <sup>b</sup>	42.3 <sup>c</sup>	8.2 <sup>b</sup>	7.7 <sup>bc</sup>	3.60 <sup>a</sup>	3.67 <sup>a</sup>	68.3 <sup>abc</sup>	64.1 <sup>bcd</sup>
<i>R. solani</i> + Pf	41.1 <sup>b</sup>	40.5 <sup>c</sup>	6.6 <sup>d</sup>	7.2 <sup>cd</sup>	3.61 <sup>a</sup>	2.07 <sup>d</sup>	62.8 <sup>c</sup>	63.1 <sup>cd</sup>
<i>R. solani</i> + Tv	41.7 <sup>b</sup>	42.7 <sup>c</sup>	7.2 <sup>c</sup>	6.9 <sup>d</sup>	3.09 <sup>c</sup>	2.72 <sup>c</sup>	67.2 <sup>bc</sup>	68.8 <sup>ab</sup>

Abbreviations as those stated for Table (4)

**Effect on NPK uptake**

Data in Table (8) showed significant increases in nitrogen, phosphorus and potassium uptake of onion that inoculated with various PGPR strains in presence of *F.*

*solani* or *R. solani* compared with uninoculated one. Also, non-infested gave higher values of NPK uptake compared to infested one.

**Table 8. Effect of PGPR inoculation on NPK uptake of onion in soil infested with *F. solani* and *R. solani*.**

Treatments	Nitrogen (mg/plant)		Phosphorus (mg/plant)		Potassium (mg/plant)	
	S <sub>1</sub>	S <sub>2</sub>	S <sub>1</sub>	S <sub>2</sub>	S <sub>1</sub>	S <sub>2</sub>
Non-infested soil with <i>F. solani</i>	27.50 <sup>e</sup>	35.28 <sup>d</sup>	0.34 <sup>d</sup>	0.38 <sup>e</sup>	3.19 <sup>l</sup>	3.53 <sup>e</sup>
Soil infested with <i>F. solani</i>	18.50 <sup>f</sup>	33.02 <sup>d</sup>	0.27 <sup>d</sup>	0.39 <sup>e</sup>	2.07 <sup>g</sup>	3.56 <sup>e</sup>
<i>F. solani</i> + Azoto	150.46 <sup>a</sup>	151.24 <sup>a</sup>	1.25 <sup>b</sup>	1.33 <sup>b</sup>	11.75 <sup>c</sup>	11.40 <sup>c</sup>
<i>F. solani</i> + Bm	120.84 <sup>b</sup>	150.46 <sup>a</sup>	1.56 <sup>a</sup>	2.14 <sup>a</sup>	14.06 <sup>a</sup>	12.31 <sup>ab</sup>
<i>F. solani</i> + Bc	101.39 <sup>c</sup>	100.46 <sup>c</sup>	1.24 <sup>b</sup>	1.13 <sup>c</sup>	12.71 <sup>b</sup>	12.56 <sup>a</sup>
<i>F. solani</i> + Bs	127.91 <sup>b</sup>	132.40 <sup>b</sup>	1.20 <sup>b</sup>	1.23 <sup>c</sup>	9.69 <sup>d</sup>	11.66 <sup>bc</sup>
<i>F. solani</i> + Pf	74.49 <sup>d</sup>	101.23 <sup>c</sup>	0.76 <sup>c</sup>	1.16 <sup>c</sup>	8.09 <sup>e</sup>	9.42 <sup>d</sup>
<i>F. solani</i> + Tv	79.83 <sup>d</sup>	99.36 <sup>c</sup>	0.84 <sup>c</sup>	0.95 <sup>d</sup>	8.13 <sup>e</sup>	10.08 <sup>d</sup>
Non-infested soil with <i>R. solani</i>	32.40 <sup>d</sup>	28.56 <sup>f</sup>	0.36 <sup>d</sup>	0.31 <sup>f</sup>	3.24 <sup>d</sup>	2.45 <sup>d</sup>
Soil infested with <i>R. solani</i>	34.44 <sup>d</sup>	24.75 <sup>f</sup>	0.37 <sup>d</sup>	0.30 <sup>f</sup>	2.95 <sup>d</sup>	2.57 <sup>d</sup>
<i>R. solani</i> + Azoto	130.14 <sup>a</sup>	147.84 <sup>a</sup>	1.06 <sup>c</sup>	1.19 <sup>c</sup>	11.95 <sup>b</sup>	13.09 <sup>a</sup>
<i>R. solani</i> + Bm	138.62 <sup>a</sup>	138.38 <sup>b</sup>	1.73 <sup>a</sup>	1.83 <sup>a</sup>	13.36 <sup>a</sup>	13.50 <sup>a</sup>
<i>R. solani</i> + Bc	132.17 <sup>a</sup>	108.57 <sup>c</sup>	1.56 <sup>b</sup>	1.48 <sup>b</sup>	13.29 <sup>a</sup>	12.83 <sup>a</sup>
<i>R. solani</i> + Bs	113.04 <sup>b</sup>	114.87 <sup>c</sup>	1.48 <sup>b</sup>	1.50 <sup>b</sup>	13.32 <sup>a</sup>	13.62 <sup>a</sup>
<i>R. solani</i> + Pf	120.21 <sup>b</sup>	64.79 <sup>e</sup>	1.48 <sup>b</sup>	0.85 <sup>e</sup>	13.00 <sup>a</sup>	7.31 <sup>c</sup>
<i>R. solani</i> + Tv	97.03 <sup>c</sup>	85.68 <sup>d</sup>	1.11 <sup>c</sup>	1.03 <sup>d</sup>	10.51 <sup>c</sup>	9.38 <sup>b</sup>

Abbreviations as those stated for Table (4)

Inoculation onion with *A. chroococcum*, *B. subtilis* or *B. megaterium* scored higher values of nitrogen uptake of onion compared to other tested PGPR strains. Additionally, the treatments inoculated with *B. megaterium*, *B. circulans*, or *B. subtilis* scored higher values of phosphorus uptake compared to other tested PGPR strains. Regarding potassium uptake, data revealed that the inoculation onion with *B. circulans* or *B. megaterium* gave higher values than other tested PGPR strains in case of infestation with *F. solani*. While the inoculation with *B. circulans* or *B. subtilis* gave higher values of potassium uptake in case of infestation with *R. solani*. The highest value of N uptake of onion was recorded with plants inoculated with *A. chroococcum*, while the highest value of P uptake was recorded with plants inoculated with *B. megaterium*.

According to its role as silicate solubilizer, the inoculation with *B. circulans* gave the highest value of K uptake of onion. The obtained results are in agreement with Li *et al.*, (2020) who reported that the inoculation with N<sub>2</sub>-fixing bacteria, phosphate dissolving bacteria and potassium releasing bacteria scored significantly increases in N, P and K plant uptake as compared to untreated treatments. The increment of NPK uptake in inoculated onion may be due to the enhancement of various PGPR treatments in increase activity of some soil enzymes (as shown in Table 6) as well as improve soil nutrition. Similar results were obtained by Nosheen and Bano (2014) and Järvan *et al.*, (2014).

**Effect of PGPRs and compost on onion production under field condition**

**Effects on microbial enzymes activity**

Dehydrogenase activity was determined in the soil rhizosphere as an indicator for respiration rate and total microbial activity. However alkaline phosphatase activity was determined for its importance in the mineralization process of organic phosphorus compounds into available phosphorus. While nitrogenase activity was determined as an indication of symbiotic N<sub>2</sub>-fixers activity. Data presented in Table (9a, b, and c) showed the effect of investigated treatments on microbial enzymes activity and indicated that there are significant increases in DH, N-ase,

and AP activity of green onion rhizosphere in some inoculated treatments compared to uninoculated one. Generally, high values of DH and N-ase activity were recorded ascendingly with treatments that treated with T9 (compost + biofertilizing-PGPR + *B. subtilis* + *P. fluorescens* + *T. viride*), and T7 (compost + biofertilizing-PGPR + *B. subtilis* + *T. viride*). Respecting AP activity the high values were recorded with T7 (compost + biofertilizing-PGPR+ *B. subtilis* + *T. viride*) and T8 (compost + biofertilizing-PGPR + *P. fluorescens* + *T. viride*).

The highest value of DH and AP activity was recorded with (T7) soil treated with compost + biofertilizing-PGPR + *B. subtilis* + *T. viride* at both seasons. While the highest values of N-ase activity were recorded with (T9). Moreover, the activity of all investigated microbial enzymes was increased gradually until 60 DAT and decrease again and the highest records of their activity were recorded at 60 DAT.

The increment of microbial enzymes values under application of PGPR strains are in agreement with those obtained by (Salazar *et al.*, 2011 and Kumar *et al.*, 2013) who reported that dehydrogenases are used as an indicator of overall soil microbial activity. Inoculation with PGPR increasing soil microbial activity. Moreover, higher records of nitrogenase activity were observed in manured soil that inoculated with biofertilizer than that treated with compost only. (Bellenger *et al.*, 2020 and Abdel-Rahman & Darwesh, 2020), and phosphatase activities were increased in the rhizosphere after inoculation with PGPR. (Bechtaoui *et al.*, 2020 and Gupta *et al.*, 2015).

It is important to mention that the three enzymes activity values were higher at the flowering stage (60 DAT) rather than in another growth stage, it could be attributed to the beneficial effect of root exudates which increase during this stage of cultivated plants, on microbial activities. This result is in harmony with those obtained by Shams *et al.*, (2013) and Abdel-Rahman *et al.*, (2017) who found that the densities of microbial enzymes in the rhizosphere were higher at the flowering stage of lettuce and tomato plants than other plant growth stages.

**Table 9a. Dehydrogenase activity in the rhizosphere of onion affected by application of PGPRs and organic fertilizer under field conditions.**

Treatments	Dehydrogenase activity (µg TPF.g <sup>-1</sup> dry soil. 24 <sup>-1</sup> )					
	S1			S2		
	30	60	90	30	60	90
Compost	4.34 <sup>g</sup>	8.27 <sup>c</sup>	7.12 <sup>e</sup>	2.23 <sup>g</sup>	12.23 <sup>d</sup>	8.71 <sup>f</sup>
Comp+Biofertilizing-PGPR	9.16 <sup>c</sup>	32.42 <sup>b</sup>	13.77 <sup>c</sup>	7.34 <sup>c</sup>	35.68 <sup>bc</sup>	18.60 <sup>de</sup>
Comp+Biofertilizing-PGPR+Bs	9.31 <sup>c</sup>	31.00 <sup>b</sup>	10.20 <sup>d</sup>	4.01 <sup>e</sup>	33.81 <sup>c</sup>	17.35 <sup>e</sup>
Comp+Biofertilizing-PGPR+Pf	8.31 <sup>d</sup>	30.97 <sup>b</sup>	10.87 <sup>c</sup>	7.75 <sup>c</sup>	36.57 <sup>abc</sup>	19.33 <sup>d</sup>
Comp+Biofertilizing-PGPR+Tv	6.01 <sup>f</sup>	30.16 <sup>b</sup>	20.37 <sup>b</sup>	1.60 <sup>h</sup>	37.31 <sup>ab</sup>	21.91 <sup>bc</sup>
Comp+Biofertilizing-PGPR+Bs+Pf	9.38 <sup>c</sup>	37.16 <sup>a</sup>	21.27 <sup>b</sup>	7.49 <sup>c</sup>	36.79 <sup>ab</sup>	23.48 <sup>b</sup>
Comp+Biofertilizing-PGPR+Bs+Tv	12.01 <sup>a</sup>	39.01 <sup>a</sup>	27.25 <sup>a</sup>	11.8 <sup>a</sup>	39.49 <sup>a</sup>	26.46 <sup>a</sup>
Comp+Biofertilizing-PGPR+Pf+Tv	7.23 <sup>e</sup>	31.53 <sup>b</sup>	10.58 <sup>d</sup>	3.20 <sup>f</sup>	35.67 <sup>bc</sup>	21.35 <sup>c</sup>
Comp+Biofertilizing-PGPR+Bs+Pf+Tv	10.60 <sup>b</sup>	38.20 <sup>a</sup>	26.66 <sup>a</sup>	9.64 <sup>b</sup>	39.38 <sup>a</sup>	25.85 <sup>a</sup>

Comp: compost recommended rate; Biofertilizing-PGPR: *Azotobacter chroococcum*, *B. megaterium*+*B. circulans*; Bs: *B. subtilis*; Pf: *P. fluorescens*; Tv: *T. viride*; S1: season one; S2: season two. A column followed by the same letter are not significantly different at P= 0.05 when compared by Duncan test

**Table 9b. Alkaline phosphatase activity in the rhizosphere of onion affected by application of PGPRs and organic fertilizer under field conditions.**

Treatments	Alkaline phosphatase activity ( $\mu\text{g pNP g}^{-1} \text{h}^{-1}$ )					
	S1			S1		
	30	60	90 DAP	30	60	90 DAP
Compost	12.27 <sup>d</sup>	25.83 <sup>d</sup>	20.31 <sup>ef</sup>	11.34 <sup>cd</sup>	12.53 <sup>e</sup>	10.94 <sup>e</sup>
Comp+Biofertilizing-PGPR	13.01 <sup>d</sup>	32.14 <sup>c</sup>	25.40 <sup>ab</sup>	11.44 <sup>cd</sup>	33.00 <sup>d</sup>	16.63 <sup>c</sup>
Comp+Biofertilizing-PGPR+Bs	12.69 <sup>d</sup>	36.73 <sup>b</sup>	22.17 <sup>de</sup>	12.24 <sup>abc</sup>	35.98 <sup>abc</sup>	13.17 <sup>d</sup>
Comp+Biofertilizing-PGPR+Pf	12.63 <sup>d</sup>	28.12 <sup>d</sup>	22.92 <sup>cd</sup>	10.86 <sup>de</sup>	33.76 <sup>cd</sup>	13.60 <sup>d</sup>
Comp+Biofertilizing-PGPR+Tv	14.50 <sup>c</sup>	37.57 <sup>ab</sup>	24.67 <sup>bc</sup>	12.04 <sup>bc</sup>	33.72 <sup>cd</sup>	16.17 <sup>c</sup>
Comp+Biofertilizing-PGPR+Bs+Pf	12.57 <sup>d</sup>	39.97 <sup>b</sup>	24.99 <sup>ab</sup>	10.25 <sup>e</sup>	32.23 <sup>d</sup>	16.74 <sup>bc</sup>
Comp+Biofertilizing-PGPR+Bs+Tv	19.11 <sup>a</sup>	38.81 <sup>a</sup>	26.98 <sup>a</sup>	13.21 <sup>a</sup>	37.15 <sup>a</sup>	18.27 <sup>a</sup>
Comp+Biofertilizing-PGPR+Pf+Tv	16.28 <sup>b</sup>	37.86 <sup>ab</sup>	24.36 <sup>bc</sup>	12.60 <sup>ab</sup>	36.64 <sup>ab</sup>	17.95 <sup>ab</sup>
Comp+Biofertilizing-PGPR+Bs+Pf+Tv	11.98 <sup>d</sup>	32.45 <sup>c</sup>	22.74 <sup>cd</sup>	10.49 <sup>de</sup>	34.31 <sup>bcd</sup>	16.13 <sup>c</sup>

Abbreviations as those stated for Table (9a)

**Table 9c. Nitrogenase activity in the rhizosphere of onion affected by application of PGPRs and organic fertilizer under field conditions.**

Treatments	Nitrogenase activity ( $\mu\text{L C}_2\text{H}_4\text{g}^{-1} \text{dry soil.h}^{-1}$ )					
	S1			S1		
	30	60	90 DAP	30	60	90 DAP
Compost	5.2 <sup>f</sup>	11.8 <sup>f</sup>	6.3 <sup>f</sup>	9.3 <sup>g</sup>	8.3 <sup>f</sup>	11.7 <sup>f</sup>
Comp+Biofertilizing-PGPR	8.6 <sup>e</sup>	31.9 <sup>e</sup>	23.8 <sup>e</sup>	11.8 <sup>f</sup>	53.7 <sup>e</sup>	23.6 <sup>e</sup>
Comp+Biofertilizing-PGPR+Bs	10.3 <sup>d</sup>	44.7 <sup>d</sup>	38.1 <sup>b</sup>	13.5 <sup>f</sup>	48.9 <sup>e</sup>	31.7 <sup>d</sup>
Comp+Biofertilizing-PGPR+Pf	12.5 <sup>c</sup>	55.2 <sup>b</sup>	35.1 <sup>c</sup>	25.7 <sup>e</sup>	63.8 <sup>d</sup>	37.9 <sup>b</sup>
Comp+Biofertilizing-PGPR+Tv	11.7 <sup>c</sup>	49.3 <sup>c</sup>	27.2 <sup>d</sup>	27.5 <sup>e</sup>	59.2 <sup>d</sup>	34.8 <sup>c</sup>
Comp+Biofertilizing-PGPR+Bs+Pf	14.7 <sup>b</sup>	58.4 <sup>ab</sup>	39.9 <sup>b</sup>	32.8 <sup>c</sup>	77.4 <sup>bc</sup>	38.7 <sup>b</sup>
Comp+Biofertilizing-PGPR+Bs+Tv	18.8 <sup>ab</sup>	62.4 <sup>a</sup>	39.3 <sup>b</sup>	36.2 <sup>b</sup>	78.6 <sup>b</sup>	37.5 <sup>b</sup>
Comp+Biofertilizing-PGPR+Pf+Tv	13.9 <sup>b</sup>	55.7 <sup>b</sup>	38.8 <sup>b</sup>	29.7 <sup>d</sup>	72.9 <sup>c</sup>	31.8 <sup>d</sup>
Comp+Biofertilizing-PGPR+Bs+Pf+Tv	19.1 <sup>a</sup>	61.7 <sup>a</sup>	44.5 <sup>a</sup>	41.7 <sup>a</sup>	89.7 <sup>a</sup>	43.8 <sup>a</sup>

Abbreviations as those stated for Table (9a)

**Effect on total NPK and plant uptake**

Data in Table (10) indicated that significant increases were found in nitrogen, phosphorus and potassium percentage and uptake of onion that inoculated with investigated PGPR strains compared with uninoculated one. Additionally, high values of NPK percentage and uptake were shown in onion that inoculated with *T. viride*, *B. subtilis* + *T. viride* or *B. subtilis* + *P. fluorescens*+Tv (T5, T7and T9). The highest value of NPK percentage and uptake of onion were recorded with plants inoculated with compost+biofertilizing-PGPR+ *B. subtilis* + *T. viride* (T7). While the lowest values for NPK

percentage and uptake of onion plant were recorded with plants that were treated with compost solely (T1).

The increment of NPK percentage and uptake in inoculated plants may be due to the positive role of inoculation with PGPR strains in improving the availability and absorption of these nutrients. These results are in agreement with those obtained by Reimer *et al.*, (2020) who reported that the use of PGPR strains as bio-inoculants would improve the supply of nutrients in the soil, decrease the use of artificial fertilizers, reduce emissions, and encourage sustainable agriculture.

**Table 10. Total NPK and plant uptake of onion affected by application of PGPRs and organic fertilizer under field conditions.**

Treatments	Nitrogen				Phosphorus				Potassium			
	Total (%)		Uptake (mg/plant)		Total (%)		Uptake (mg/plant)		Total (%)		Uptake (mg/plant)	
	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2
Compost	3.2 <sup>bc</sup>	3.1 <sup>b</sup>	176.0 <sup>f</sup>	170.5 <sup>g</sup>	0.3 <sup>c</sup>	0.4 <sup>c</sup>	1.65 <sup>g</sup>	2.20 <sup>e</sup>	2.2 <sup>de</sup>	2.3 <sup>c</sup>	12.10 <sup>d</sup>	12.65 <sup>c</sup>
Comp+Biofertilizing-PGPR	3.5 <sup>ab</sup>	3.2 <sup>b</sup>	325.5 <sup>ab</sup>	313.6 <sup>b</sup>	0.4 <sup>b</sup>	0.5 <sup>b</sup>	3.72 <sup>d</sup>	4.90 <sup>a</sup>	2.9 <sup>a</sup>	2.8 <sup>a</sup>	26.97 <sup>b</sup>	27.44 <sup>a</sup>
Comp+Biofertilizing-PGPR+Bs	3.5 <sup>ab</sup>	3.0 <sup>b</sup>	269.5 <sup>e</sup>	243.0 <sup>f</sup>	0.3 <sup>c</sup>	0.5 <sup>b</sup>	2.31 <sup>f</sup>	4.05 <sup>b</sup>	2.7 <sup>ab</sup>	2.7 <sup>a</sup>	20.79 <sup>c</sup>	21.87 <sup>b</sup>
Comp+Biofertilizing-PGPR+Pf	3.5 <sup>ab</sup>	3.1 <sup>b</sup>	294.0 <sup>cd</sup>	266.6 <sup>abc</sup>	0.4 <sup>b</sup>	0.4 <sup>c</sup>	3.36 <sup>e</sup>	3.44 <sup>d</sup>	2.7 <sup>ab</sup>	2.6 <sup>ab</sup>	22.68 <sup>c</sup>	22.36 <sup>b</sup>
Comp+Biofertilizing-PGPR+Tv	3.2 <sup>bc</sup>	3.1 <sup>b</sup>	304.0 <sup>bc</sup>	300.7 <sup>bc</sup>	0.5 <sup>a</sup>	0.4 <sup>c</sup>	4.75 <sup>b</sup>	3.88 <sup>bc</sup>	2.7 <sup>ab</sup>	2.4 <sup>bc</sup>	25.65 <sup>b</sup>	23.28 <sup>b</sup>
Comp+Biofertilizing-PGPR+Bs+Pf	3.1 <sup>c</sup>	3.1 <sup>b</sup>	279.0 <sup>de</sup>	282.1 <sup>cd</sup>	0.5 <sup>a</sup>	0.4 <sup>c</sup>	4.50 <sup>b</sup>	3.64 <sup>cd</sup>	2.4 <sup>cd</sup>	2.4 <sup>bc</sup>	21.60 <sup>c</sup>	21.84 <sup>b</sup>
Comp+Biofertilizing-PGPR+Bs+Tv	3.6 <sup>a</sup>	3.5 <sup>a</sup>	345.6 <sup>a</sup>	360.5 <sup>a</sup>	0.5 <sup>a</sup>	0.6 <sup>a</sup>	5.05 <sup>a</sup>	5.15 <sup>a</sup>	2.9 <sup>a</sup>	2.6 <sup>ab</sup>	29.29 <sup>a</sup>	26.78 <sup>a</sup>
Comp+Biofertilizing-PGPR+Pf+Tv	3.1 <sup>c</sup>	3.1 <sup>b</sup>	295.2 <sup>cd</sup>	257.3 <sup>ef</sup>	0.5 <sup>a</sup>	0.5 <sup>b</sup>	4.10 <sup>c</sup>	4.98 <sup>a</sup>	2.6 <sup>bc</sup>	2.6 <sup>ab</sup>	21.32 <sup>c</sup>	21.58 <sup>b</sup>
Comp+Biofertilizing-PGPR+Bs+Pf+Tv	3.6 <sup>a</sup>	3.2 <sup>b</sup>	313.1 <sup>bc</sup>	304.0 <sup>bc</sup>	0.4 <sup>b</sup>	0.4 <sup>c</sup>	3.84 <sup>cd</sup>	3.80 <sup>bc</sup>	2.8 <sup>ab</sup>	2.8 <sup>a</sup>	26.88 <sup>b</sup>	26.60 <sup>a</sup>

Abbreviations as those stated for Table (9a)

**Effects on growth characteristics**

Data in Table (11) showed significant increases in plant height, leaves number and dry weight of onion plant that inoculated with various PGPR strains compared with uninoculated one. It is worth mentioning that there are low significant differences among all inoculate treatments with PGPR strains. The highest value of all estimated vegetative

growth characters was recorded with plants treated with compost + biofertilizing-PGPR + *B. subtilis* + *T. viride* (T7). While, the lowest values for plant height, leave numbers, dry weight, and vegetable yield of onion plants were found at the treated plants with compost only(T1).

Regarding vegetative yield of green onion data in Table (11) indicated that there was a significant increase in

the plant that inoculated with various PGPR strains in comparison with those treated with compost only. The high values vegetative yield of green onion plant recorded with (T7, T6 and T5) compared to other tested PGPR strains on both seasons. The highest value of vegetative yield of green onion plant was recorded with plants inoculated with compost + biofertilizing-PGPR + *B. subtilis* + *T. viride* (T7). While the lowest values for the vegetative yield of green onion plant were found at the (T1) treated with

compost only. The obtained results are in agreement with Abou Zaid *et al.*, (2011) and Abdel-Rahman *et al.*, (2017) who reported that PGPRs as biofertilizers produce plant growth promoting substances led to enhancement plant vegetative growth parameters and support yield production. Moreover Samayoa *et al.*, (2020) reported that PGPRs are beneficial microbes that increase growth and yield of onion (*Allium cepa* Linn.).

**Table 11. Plant height, Leaf number, Fresh weight and vegetative yield of onion affected by application of PGPRs and organic fertilizer under field conditions.**

Treatments	Plant height (cm/plant)		Leaves number plant <sup>-1</sup>		Dry weight (g/plant)		Vegetative yield (Ton/fed. <sup>-1</sup> )	
	S 1	S 2	S 1	S 2	S 1	S 2	S 1	S 2
Compost	41.3 <sup>de</sup>	41.6 <sup>de</sup>	5.7 <sup>de</sup>	6.0 <sup>c</sup>	5.5 <sup>e</sup>	5.5 <sup>e</sup>	4.20 <sup>f</sup>	4.26 <sup>e</sup>
Comp+Biofertilizing-PGPR	46.3 <sup>ab</sup>	46.7 <sup>ab</sup>	6.4 <sup>bc</sup>	6.8 <sup>b</sup>	9.3 <sup>b</sup>	9.8 <sup>ab</sup>	6.81 <sup>cde</sup>	6.88 <sup>cd</sup>
Comp+Biofertilizing-PGPR+Bs	42.2 <sup>cde</sup>	42.7 <sup>bcde</sup>	5.9 <sup>cde</sup>	6.2 <sup>c</sup>	7.7 <sup>d</sup>	8.1 <sup>d</sup>	6.32 <sup>e</sup>	6.42 <sup>d</sup>
Comp+Biofertilizing-PGPR+Pf	45.0 <sup>abcd</sup>	46.2 <sup>abc</sup>	5.9 <sup>cde</sup>	6.1 <sup>c</sup>	8.4 <sup>cd</sup>	8.6 <sup>cd</sup>	6.91 <sup>bcd</sup>	6.97 <sup>bcd</sup>
Comp+Biofertilizing-PGPR+Tv	43.9 <sup>abcde</sup>	44.6 <sup>abcd</sup>	6.1 <sup>cd</sup>	6.4 <sup>bc</sup>	9.5 <sup>ab</sup>	9.7 <sup>ab</sup>	7.23 <sup>abc</sup>	7.33 <sup>abc</sup>
Comp+Biofertilizing-PGPR+Bs+Pf	45.8 <sup>abc</sup>	46.4 <sup>abc</sup>	6.3 <sup>bc</sup>	6.8 <sup>b</sup>	9.0 <sup>bc</sup>	9.1 <sup>bc</sup>	7.45 <sup>ab</sup>	7.52 <sup>ab</sup>
Comp+Biofertilizing-PGPR+Bs+Tv	46.9 <sup>a</sup>	47.2 <sup>a</sup>	7.4 <sup>a</sup>	7.7 <sup>a</sup>	10.1 <sup>a</sup>	10.3 <sup>a</sup>	7.62 <sup>a</sup>	7.70 <sup>a</sup>
Comp+Biofertilizing-PGPR+Pf+Tv	42.4 <sup>bcde</sup>	42.6 <sup>cde</sup>	6.7 <sup>b</sup>	6.0 <sup>c</sup>	8.2 <sup>d</sup>	8.3 <sup>d</sup>	6.61 <sup>de</sup>	6.63 <sup>d</sup>
Comp+Biofertilizing-PGPR+Bs+Pf+Tv	44.8 <sup>abcd</sup>	45.1 <sup>abcd</sup>	6.2 <sup>bcd</sup>	6.4 <sup>bc</sup>	9.6 <sup>ab</sup>	9.5 <sup>b</sup>	6.93 <sup>bcd</sup>	6.94 <sup>cd</sup>

Abbreviations as those stated for Table (9a)

**Effects on defense enzymes activity in onion.**

Data in Table (12) showed significant differences in peroxidase and polyphenoloxidase activity between a plant that inoculated with various PGPR strains and uninoculated plants. Moreover, the values of peroxidase and polyphenoloxidase activity of the green onion plants were higher in treatments (T9, T8 and T4) than other inoculated ones. The highest value of peroxidase and polyphenoloxidase activity of green onion plant were recorded with plants inoculated with compost+biofertilizing-PGPR+ *B. subtilis* + *P. fluorescens* + *T. viride* (T9). While the lowest values for peroxidase and polyphenoloxidase activity of green onion plant were found at plants treated with compost only (T1).

Similar results of the positive effect of inoculation with PGPR strains on peroxidase and polyphenoloxidase production were found by Gailite *et al.*, (2005) who reported that the content of both peroxidase and polyphenoloxidase increased after the treatment with plant growth-promoting bacteria or fungi.

**Table 12. Peroxidase and polyphenoloxidase activity of onion affected by application of PGPRs and organic fertilizer under field conditions.**

Treatments	Peroxidase		Polyphenoloxidase	
	S 1	S 2	S 1	S 2
Compost	0.62 <sup>d</sup>	0.64 <sup>ef</sup>	0.11 <sup>e</sup>	0.11 <sup>d</sup>
Comp+Biofertilizing-PGPR	0.72 <sup>b</sup>	0.70 <sup>cd</sup>	0.12 <sup>de</sup>	0.13 <sup>c</sup>
Comp+Biofertilizing-PGPR+Bs	0.73 <sup>b</sup>	0.74 <sup>bcd</sup>	0.14 <sup>bc</sup>	0.14 <sup>bc</sup>
Comp+Biofertilizing-PGPR+Pf	0.83 <sup>a</sup>	0.80 <sup>ab</sup>	0.15 <sup>b</sup>	0.19 <sup>a</sup>
Comp+Biofertilizing-PGPR+Tv	0.64 <sup>cd</sup>	0.69 <sup>cd</sup>	0.06 <sup>f</sup>	0.15 <sup>b</sup>
Comp+Biofertilizing-PGPR+Bs+Pf	0.70 <sup>b</sup>	0.69 <sup>cd</sup>	0.13 <sup>cd</sup>	0.14 <sup>bc</sup>
Comp+Biofertilizing-PGPR+Bs+Tv	0.69 <sup>bd</sup>	0.68 <sup>de</sup>	0.12 <sup>de</sup>	0.14 <sup>bc</sup>
Comp+Biofertilizing-PGPR+Pf+Tv	0.73 <sup>b</sup>	0.75 <sup>bc</sup>	0.14 <sup>bc</sup>	0.15 <sup>b</sup>
Comp+Biofertilizing-PGPR+Bs+Pf+Tv	0.86 <sup>a</sup>	0.84 <sup>a</sup>	0.18 <sup>a</sup>	0.19 <sup>a</sup>

Abbreviations as those stated for Table (9a)

**CONCLUSION**

From data of current study, it could be concluded that using all PGPR strains suppressed *F. solani* and *R. solani*. The inhibition zone increased by increasing incubation time. Under greenhouse condition, onion inoculation with various PGPR strains reduced disease severity and disease incidence of *F. solani* and *R. solani*. Also, the application with PGPR had a positive effect on plant defense enzymes, nutrients uptake and growth parameters. The application of tested PGPR strains with compost were used for green onion production in the open field under the organic agriculture system. During the experiment the microbial enzymes activity, total NPK and plant uptake, defense enzymes activity, growth characteristics and vegetative yield were estimated. Obtained data showed that onion treated with compost combined with biofertilizing-PGPR, *B. subtilis* and *T. viride* (T7) gave the highest values of all estimated parameters. So, it can be recommended as integrated fertilizing program to promote plant growth, increase crop production, decrease production costs.

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### تطبيق بعض أساليب الزراعة العضوية لتعزيز نمو وإنتاج البصل الأخضر

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تحت ظروف المعمل تم دراسة التأثير المضاد للعديد من سلالات البكتيريا المشجعة لنمو النبات وهي *A. chroococcum* و *B. megaterium* و *B. subtilis* و *Ps. fluorescens* و فطر *T. viride* ضد الفطريات المسببة لأعغان الجذور *F. solani* و *R. solani*. أظهرت النتائج أن استخدام جميع سلالات البكتيريا المشجعة لنمو النبات قد ثبتت من نمو كلا من *F. solani* و *R. solani* وقد زادت منطقة التثبيط بزيادة زمن التحضين. وكذلك تحت ظروف الصوبة، قد أظهرت النتائج أن تلقيح البصل الأخضر بسلالات البكتيريا المشجعة لنمو النبات المختلفة إلى خفض شدة المرض الناتج عن الإصابة بـ *F. solani* بنسبة تتراوح بين (79.4-93.5%). في حين أنها خفضت من شدة المرض الناتج عن الإصابة بـ *R. solani* بنسبة تتراوح بين (84.5-93.6%). وعليه فقد انخفض معدل الإصابة بالمرض بنسبة تراوحت بين (68.4-81.8%) و (65.5-81.8%) في حالة الإصابة بالفطر *F. solani* و *R. solani* على التوالي. علاوة على ذلك، كان للتلقيح بسلالات البكتيريا المشجعة لنمو النبات تأثير إيجابي على إنزيمات الدفاع في النبات وكذلك على امتصاص العناصر الغذائية وصفات النمو. وعند زراعة البصل في الحقل المفتوح في ارض تابعة للمعمل المركزي للزراعة العضوية لدراسة التأثير المتكامل لاستخدام سلالات البكتيريا المشجعة لنمو النبات تحت الدراسة وعوامل المكافحة الحيوية والسماد العضوي لتحسين خصائص نمو ومحصول البصل الأخضر في ظل نظام الزراعة العضوية. خلال التجربة تم تقدير نشاط الإنزيمات الميكروبية في التربة، وكذلك النيتروجين والفوسفور والبوتاسيوم الكلي والملتص في النبات، ونشاط الإنزيمات الدفاعية، وأخيرا خصائص النمو والمحصول الأخضر. أظهرت النتائج المتحصل عليها أن البصل المعامل بالكومبوست مع التسميد الحيوي بسلالات البكتيريا المشجعة لنمو النبات وعوامل المقاومة الحيوية *B. subtilis* و *T. viride* (المعاملة 7) قد أعطت أعلى القيم في جميع القياسات المقدره باستثناء نشاط الإنزيمات الدفاعية. لذلك، يمكن التوصية بالمعاملة السابقة كبرنامج تسميد متكامل لتعزيز نمو وزيادة إنتاج محصول البصل الأخضر وتقليل تكاليف الإنتاج.