

Effect of seed treatments with plant growth regulators on reducing damping-off and root rot diseases in common bean under greenhouse conditions

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Abstract

This study was carried out to evaluate the efficiency of some plant growth regulators (PGRs) to protect common bean plants against damping-off and root rot which caused by Rhizoctonia solani (Kuhn). Fourteen isolates of R. solani were isolated from common bean plants. Pathogenicity tests of these isolates were carried out on common bean (Giza 6 cv.) in greenhouse experiments during growing season in 2014. Results showed that all the tested isolates of R. solani were pathogenic to common bean plants with different degrees of infection. PGRs (indole 3-butric acid, gibberellic acid and N_6 -benzyladenine) have been evaluated as soaking seeds treatments before sowing at concentrations 50 and 100 ppm under greenhouse conditions. Treatments with the tested PGRs were effective in reducing disease severity (DS) percent at the tested parameters, i.e. damping-off and root rot. Also, data indicate that the highest significant reduction of diseases severity was observed in the case of N₆-benzyladenine treatment (100 ppm), followed by indole 3-butyric acid (100 ppm). The concentration 50 ppm of PGRs was the most effective in reducing both diseases (damping-off and root rot). Application of the tested PGRs has significantly enhanced the height and the fresh weight of common bean plants compared to the control and fungicides (Moncut 25%). Gibberellic acid (100 ppm) was the most effective in increasing the height and the fresh weight of plants than the other treatments at seasons 2015 and 2016. Laboratory estimates, showed that treated plants with the tested PGRs content more phenolic compounds compared to untreated plants. The treatment with N₆benzyladenine was more content of phenolic compounds than the other treatments. There is an inverse relationship between the level of phenolic compounds in treated plants and the disease severity.

Keywords: common bean, phenolic compounds, plant growth regulators, Rhizoctonia solani.



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Introduction

Common bean (Phaseolus vulgaris L.) is one of the most widely cultivated food legume species in the world (Baudoin et al., 2001). It is a major source of low cost calories, protein, dietary fiber, minerals poor populations vitamins for and (Hillocks et al., 2006). Rhizoctonia solani [Teleomorph: **Thanatephorus** Kuhn cucumeris (Frank) Donk] is a soilborne pathogen causing severe damage to a wide range of crops. In tropical regions, this pathogen cause different types of symptoms on bean, including pre- and post-emergence damping-off, root and hypocotyl rot and foliar blight (Fenille et al., 2002). Damping-off and root rot are destructive diseases of green, snap, lima and dry beans worldwide (Miller et al., 1995). Root rot is a major constraint leading to severe crop losses of common bean production in many parts of the world, especially under tropical and subtropical climate conditions (Matloob & Juber, 2013; D'aes et al., 2011). Regulation of plant growth by the use of plant growth regulators can modify crops, making them more productive and more efficient, and can change the timing of normal events such as flowering or germinating (Parker, 2010). Recently, attempts have been made to use plant growth regulators not only to increase yield but also to control or minimizing the damage and loss caused by plant pathogens. There are many researches explained the role of PGRs in plant diseases control, such as brown patch caused by R. solani (Burpee, 1998), gray leaf spot caused by Pyricularia grisea (McDonald et al., 2006; Uddin & Soika, 2000). anthracnose caused bv Colletotrichum cereale (Inguagiato et al., 2008) and Alternaria leaf spot disease on faba bean (Abd El-Hai. 2015). Gibberellins (GAs) play an essential role

in many aspects of plant growth and development, such as seed germination (Maske et al., 1997), stem elongation and flower development (Yamaguchi & Kamiya, 2000). Application of GA3 (100 ppm) led to increase in plant height, average number of leaves, leaf area per plant and dry weight of shoot in Vicia faba (Ibrahim et al., 2007). Treatment of wounds with IAA at 20 µg/ml 24 h before pathogen inoculation resulted in significant inhibition of Penicillium expansum spore germination and host infection (Yu et al., 2009). The objectives of this study were to: (i) evaluate the efficacy of some plant growth regulators on reducing damping-off and root rot of common bean, and; (ii) study their effect on plant growth and the level of phenolic compounds in treated plants.

Materials and methods

Isolation and identification of R. solani isolated from roots of common bean plants: Common bean plants in various stages with symptoms of Rhizoctonia root rot disease were collected from different localities in Egypt (El- Behera and Assiut governorates). Diseased plant roots were washed carefully under running tap water to remove the adjacent soil particles followed by sterile water, then dried between two filter papers to remove the excess of water. Pieces of tissues from the margins of lesion on roots were cut into small pieces (1cm). Pieces surface sterilized using 1% sodium hypochlorite solution for 3 min, then washed three times thoroughly in sterilized distilled water. Sterilized pieces were dried above sterilized filter papers and placed on Potato Dextrose Agar (PDA) medium amended with streptomycin sulphate (5 mg/L)to

suppress bacterial growth. Petri dishes were incubated at 28°C and examined every 24 h to observe the hyphal growth in the plates. Purification of the isolates found was done using the hyphal tip technique. Pure cultures were identified according to the cultural properties, the morphological and the microscopical characteristics as described by Sneh et al. (1991). Hyphal tipped isolates tentatively identified as *R. solani* were transferred to PDA medium. Pure culture of isolates were kept in refrigerator at 4°C on PDA slants and stored for further studies.

Pathogenicity test: The pathogenicity of the obtained R. solani isolates were tested on common bean plants (Giza 6 cv.) during growing season in 2014 under greenhouse conditions. Inoculum of each solani isolate was prepared by *R*. autoclaved barley medium in 500 ml glass bottles (150g barley + 50 g clean sand + 4 g glucose + 0.2 g yeast extract + 200 ml water). The bottles were inoculated with the isolates and incubated at 25°C in the dark for two weeks. The test was carried out using sterilized plastic bags (30 cm in diam.) containing sterilized clay-sand soil. The sterilization of plastic bags and soil was carried out by using 5% of formaldehyde solution. The content of bottles was thoroughly mixed in plastic container and used as a source of inoculum. Inoculum of each isolate of the pathogen was added at a rate of 1 % to the plastic bags soil (w/w), one week before sowing, mixed well and then thoroughly irrigated. Sterilized six seeds of common bean were sown in each plastic bag. Three replicates were used for each treatment. Plastic bags containing non-infested soil were used as control. Then, the plastic bags were kept under greenhouse conditions in natural light (Agricultural Botany Department, Faculty of Agriculture, Al-Azhar University, Assiut Branch. Assiut. Egypt) and irrigated when required. The disease severity was determined by recording the percentage of preemergence damping-off (% of unemerged seeds) 15 days after sowing as well as post-emergence damping-off (% of dead plants) 45 days after sowing (Carling et al., 1999). After 60 days from sowing, disease severity of root rot was determined in survival plants using the scales as described by Nelson et al. (1996) where: 1 = no symptoms, 2 =lesion(s) < 3mm and/or < 25% girdling, 3 = lesion (s) 3 to 6 mm and/or > 25 to 50% girdling, 4 = lesions > 6 mm and/or> 50% girdling, and 5= 75% of leaves wilted or plant dead. Disease severity index (DSI) was calculated using the following formula (Liu et al. 1995):

Disease Severity Index % =
$$\frac{\sum d}{d_{max} \times n} \times 100$$

Where, d= the disease rating possible, $d_{max}=$ the maximum disease rating and n= the total number of plants examined in each replicate.

Effect of plant growth regulators on reducing damping-off and root rot diseases severity: Plant growth regulators (indole 3-butric acid. gibberellic acid and N₆-benzyladenine) have been evaluated as seed treatments against damping-off and root rot of common bean under greenhouse conditions. Highly pathogenic isolates (4 and 7) were selected for this study. Inocula of tested pathogenic isolates and soil infestation were done as mentioned before. Surface sterilized common bean

seeds were soaking in the tested plant growth regulators and the fungicide (Moncut 25%) at concentrations 50 and 100 ppm for 30 min before sowing (Tzortzakis, 2009). Seeds were left to dry then sown in infested soil. Six seeds were sown in each plastic bag and three replicates for each treatment have been used. Soaked seeds in distilled water were sown in infested soil as control. Percent of pre and post-emergence damping-off were recorded as mentioned before and three plastic bags were used as replicates for each treatment. After 60 from sowing, survival plants days carefully removed for determination of root rot disease severity, average plant height (cm) and fresh weight (gm/plant).

Determination of the total phenolic compounds: Two grams of treated common bean plants were cut into small portions, immediately plunged into 95% ethanol alcohol for 10 min, in order to kill the tissues then extracted for 8-10 h in soxhlet units using 70% ethanol till the percolate was colorless. The combined ethanol extracts were filtered and evaporated on water path at 70°C to near dryness. The dried residues were redissolved in 50% isopropanol. Then, isopropanol extracts were used for determining free, total and conjugated phenols using Folin and Ciocatalteus reagent as described by Snell and Snell (1953). Total phenols were determined as follows: ten drops of conc. HCl (70%) were added to the samples, heated rapidly to boiling point and placed in a boiling water bath for 10 min. After cooling 1 ml of the reagent and 5 ml of 20 % Na2CO3 were added. The mixture was diluted to 10 ml and determination was carried out by spectrophotometer at 520 after 30 min. nm Phenolic compounds were calculated as milligrams equivalent of catechol/gm fresh weight of shoot plant using the following formula:

Sample conc. (mg) = $\frac{(\text{Reading of sample } \times \text{ total volume of sample (cm) } / \text{ sample volume (ml))}}{\text{Standard curve of catechol } \times \text{Fresh weight (gm)}} \times 100$

Statistical analysis: The obtained data were subjected to statistical analysis using the MSTAT-C program version 2.10 (1991). The least significant difference (L.S.D., p = 0.05) has been used for the comparison between means of treatments as stated by Gomez and Gomez (1984).

Results

Pathogenicity tests of *R. solani* **isolates on common bean plants:** Results presented in Table (1) indicate that all the tested isolates of *R. solani* were pathogenic to common bean plants (Giza 6 cv.) with different degrees of infection and disease severity. Data also indicate that isolates No. 4 followed by No. 6 then No. 13 were the highest pathogenic and caused the highest percent of preemergence damping-off, while the isolates No. 5 followed by No. 7 then No. 2 exhibited the lowest disease severity of pre-emergence damping-off. The isolates No. 3 followed by No. 2 the No. 4 caused the highest percent of postdamping-off; while. emergence the isolates No. 13 followed by No. 6 then No. 11 gave the lowest percent. The

isolate No. 7 gave the highest percent of root rot followed by isolates No. 13 then No. 14. While, the other tested isolates gave the lowest percentage of diseases severity.

| Table 1: Pathogenicity of R. solani isolates on Giza 6 common | bean cultivar under greenhouse conditions. |
|---|--|
|---|--|

| No. of | | Dise Damping-o | | |
|--------------------|-----------|--|--------------------------------------|---------------------|
| tested isolates | Location | Pre-emergence (% of un-emerged seeds) | Post-emergence (% of dead plants) | Root rot (%) |
| 1 | Assiut | 44.45 ^{efg} | 24.44 ^{abcd} | 18.51 ^{gh} |
| 2 | El-Behera | 27.78 ^{hij} | 36.11 ^{ab} | 46.20^{d} |
| 3 | El-Behera | 38.89 ^{fgh} | 41.11 ^a | 36.25 ^e |
| 4 | El-Behera | 83.33 ^a | 33.33 ^{abc} | 11.40^{i} |
| 5 | Assiut | 16.67 ^j | 28.89 ^{abcd} | 13.86 ^{hi} |
| 6 | El-Behera | 77.78 ^{ab} | 16.67 ^{cde} | 27.03^{f} |
| 7 | El-Behera | 22.22 ^{ij} | 26.67 ^{abcd} | 89.10^{a} |
| 8 | Assiut | 61.11 ^{cd} | 27.78 ^{abcd} | 16.74^{ghi} |
| 9 | El-Behera | 55.56 ^{cde} | 22.22 ^{bcd} | 20.13 ^g |
| 10 | Assiut | 33.33 ^{ghi} | 30.00 ^{abc} | 18.14 ^{gh} |
| 11 | Assiut | 50.00^{def} | 19.44 ^{bcd} | 50.87^{d} |
| 12 | El-Behera | 50.00^{def} | 27.78 ^{abcd} | 26.52^{f} |
| 13 | El-Behera | 66.67 ^{bc} | 11.11 ^{de} | 73.49 ^b |
| 14 | Assiut | 44.44 ^{efg} | 30.55 ^{abc} | 63.18 ^c |

Mean followed by the same letter in a column are not significantly different according to Duncan's multiple range test (P < 0.05).

Effect of PGRs as soaking common bean seeds treatments on reducing damping-off and root rot diseases under greenhouse conditions: Plant growth regulators, i.e. indole-3-butric acid, gibberellic acid and N_{6} benzyladenine had a beneficial effect when treated to common bean seeds as soaking treatments at the concentrations 50 and 100 ppm for 30 min before sowing. Data presented in Tables (2 and 3) show that treatment seeds with PGRs significantly reduced disease severity percentages of damping-off and root rot diseases compared to the control. In general, all the **PGRs** tested at concentration 50 ppm were the most effective in reducing the disease severity in both diseases more than concentration

100 There were significant ppm. differences among the PGRs in reducing the both diseases (damping-off and root also showed that N₆rot). Data benzyladenine gave the best reduction of both diseases compared to other tested PGRs. As, pre-emergence damping-off was recorded (13.89 and 16.67%), postemergence damping-off (12.22 and 12.28%) and root rot percent (18.38 and 13.33%) at seasons 2015 and 2016, respectively. Treatment with the fungicide (Moncut 25%) at concentration 100 ppm showed the lowest percent of pre-emergence damping-off (4.8 and 11.11%), post-emergence damping-off (7.50 and 8.33%) and disease severity of root rot (16.74 and 13.72%) at seasons 2015 and 2016 respectively, compared to PGRs.

Effect of seed treatments with some plant growth regulators on plant height and fresh weight of common plants under bean greenhouse conditions: Data presented in Table (4) indicated that the soaking of common bean seeds in plant growth regulators solutions before sowing at the concentrations 50 and 100 ppm improved the plant growth characters as the height and the fresh weight of common bean plants (Giza 6 cv.) in both seasons (2015 and 2016) compared to untreated plants. The highest plant height was observed in the case of the gibberellic acid treatment at the concentration of 100 ppm with isolate No. 7 in both seasons 2015 and 2016 (33.77 and 43.17 cm), respectively. However, the highest fresh weight was found when common bean seeds treated with isolate No. 7 at the concentration

100 ppm of GA3 (16.17 and 19.55 g/plant) in both seasons 2015 and 2016 respectively. While, the lowest height (18.67 cm) and fresh weight (6.63 g/plant) of plants has been recorded with N₆-benzyladenine at concentration 50 ppm. The gibberellic acid treatment caused the best increase of length and fresh weight of plants. Moreover, the best concentration of PGRs gave highest height and fresh weight of plants was obtained at 100 ppm at both seasons 2015 and 2016. Treated seeds with the fungicide (Moncut 25%) showed the lowest plant height (16 cm) and fresh weight (6.84 g/plant) of plants compared to PGRs. Finally, all tested PGRs were effective in increasing the percent of germination as well as in the of the plant improvement growth characters compared to the untreated plants.

| Table 2: Efficacy of some plant growth regu | ators on reducing Rhizoctonia | diseases severity of common bean un | nder |
|---|-------------------------------|-------------------------------------|------|
| greenhouse conditions at growing sea | son 2015. | | |

| | ч | | | | Dis | ease Sev | erity % | | | | | |
|-------------------------------|-------|---------|-------------------------|----------------|--------|--------------------|--------------|--------|--------|-------|--|--|
| | uıdd | |] | | | | | | | | | |
| Treatments | C.] | Pre | e-emergen | Post-emergence | | | Root rot (%) | | | | | |
| | Conc. | (% of u | (% of un-emerged seeds) | | | (% of dead plants) | | | | | | |
| | 0 | Iso. 4 | Iso. 7 | Mean | Iso. 4 | Iso. 7 | Mean | Iso. 4 | Iso. 7 | Mean | | |
| Indole 3-butric acid | 50 | 16.67 | 11.11 | 13.89 | 21.67 | 22.22 | 21.95 | 27.72 | 25.00 | 26.36 | | |
| | 100 | 33.33 | 27.78 | 30.56 | 27.78 | 25.00 | 26.39 | 33.33 | 27.35 | 30.34 | | |
| Gibberellic acid | 50 | 33.33 | 22.22 | 27.78 | 27.78 | 25.00 | 26.39 | 35.14 | 30.41 | 32.78 | | |
| | 100 | 38.89 | 33.33 | 36.11 | 36.11 | 25.56 | 30.84 | 41.67 | 33.33 | 37.50 | | |
| N ₆ -benzyladenine | 50 | 16.67 | 11.11 | 13.89 | 13.33 | 11.11 | 12.22 | 20.08 | 16.67 | 18.38 | | |
| | 100 | 27.78 | 16.67 | 22.23 | 18.89 | 13.40 | 13.65 | 24.50 | 20.91 | 22.71 | | |
| Moncut 25% | 50 | 16.67 | 11.11 | 13.89 | 13.33 | 12.22 | 12.78 | 21.44 | 16.67 | 19.06 | | |
| | 100 | 11.11 | 5.56 | 8.34 | 8.33 | 6.67 | 7.50 | 18.83 | 14.64 | 16.74 | | |
| Control (untreated seeds) | - | 44.44 | 50.00 | 47.22 | 55.56 | 44.44 | 50.00 | 50.00 | 46.67 | 48.34 | | |
| Mean | - | 26.54 | 20.99 | - | 24.75 | 20.62 | - | 30.30 | 25.74 | - | | |
| L.S.D. at 5% for | | | | | | | | | | | | |
| Treatments (A) | | | 7.82 | | | 11.37 | | | 2.99 | | | |
| Concentration (B) | | | 4.95 | | | 7.19 | | | 1.89 | | | |
| Isolates (C) | | | 6.76 | | | 8.20 | | | 1.48 | | | |
| Interaction (A×B×C) | | | 21.37 | | | 25.94 | | | 4.67 | | | |

| | ч | | | | Dis | ease Sev | erity % | | | | | |
|-------------------------------|-------|---------|---------------|-----------------|--------|--------------------|--------------|--------|--------|-------|--|--|
| | uıdd | | Damping-off % | | | | | | | | | |
| Treatments | IC.] | Pre | e-emergend | Post-emergence | | | Root rot (%) | | | | | |
| | Conc. | (% of u | n-emerged | -emerged seeds) | | (% of dead plants) | | | | | | |
| | 0 | Iso. 4 | Iso. 7 | Mean | Iso. 4 | Iso. 7 | Mean | Iso. 4 | Iso. 7 | Mean | | |
| Indole 3-butric acid | 50 | 22.22 | 16.67 | 19.45 | 22.22 | 20.00 | 21.11 | 26.67 | 25.00 | 25.84 | | |
| | 100 | 27.78 | 27.78 | 27.78 | 33.33 | 25.00 | 29.17 | 33.33 | 20.00 | 26.67 | | |
| Gibberellic acid | 50 | 33.34 | 33.33 | 33.33 | 26.11 | 26.67 | 26.39 | 40.13 | 26.67 | 33.40 | | |
| | 100 | 38.89 | 27.78 | 33.34 | 36.11 | 21.67 | 28.89 | 46.67 | 33.33 | 40.00 | | |
| N ₆ -benzyladenine | 50 | 16.67 | 16.67 | 16.67 | 13.33 | 12.22 | 12.78 | 13.33 | 13.33 | 13.33 | | |
| | 100 | 22.22 | 16.67 | 19.45 | 18.89 | 15.00 | 16.95 | 20.00 | 13.33 | 16.67 | | |
| Moncut 25% | 50 | 16.67 | 11.11 | 13.89 | 13.33 | 11.11 | 12.22 | 20.00 | 17.78 | 18.89 | | |
| | 100 | 11.11 | 11.11 | 11.11 | 8.33 | 8.33 | 8.33 | 13.33 | 14.10 | 13.72 | | |
| Control (untreated seeds) | - | 44.45 | 55.56 | 50.01 | 50.00 | 44.44 | 47.22 | 50.00 | 46.67 | 48.34 | | |
| Mean | - | 25.93 | 24.08 | - | 24.63 | 20.49 | - | 29.27 | 23.36 | - | | |
| L.S.D. at 5% for | | | | | | | | | | | | |
| Treatments (A) | | | 10.27 | | | 8.63 | | | 1.67 | | | |
| Concentration (B) | | | 6.50 | | | 5.46 | | | 1.05 | | | |
| Isolates (C) | | | 8.59 | | | 8.23 | | | 1.42 | | | |
| Interaction (A×B×C) | | | 27.17 | | | 26.02 | | | 4.49 | | | |

Table 3: Efficacy of some plant growth regulators on reducing Rhizoctonia diseases severity of common bean under greenhouse conditions at growing season 2016.

Table 4: Effect of seed treatments with certain plant growth regulators on the height and fresh weight of common bean plants under greenhouse conditions at growing seasons 2015 and 2016.

| | n | Season 2015 | | | | | | Season 2016 | | | | | |
|---|-----------|-------------|------------------------------|--------------|--------|------------------------------|---------|-------------|------------------------------|-------|--------|------------------------------|---------|
| Treatments | Conc. ppm | | | height m) | | Fre weigh (gi | t/plant | 1 | Plant heigh (cm) | t | Fı | esh weigh (gm) | t/plant |
| | Ŭ | Iso. 4 | Iso. 7 | Mean | Iso. 4 | Iso. 7 | Mean | Iso. 4 | Iso. 7 | Mean | Iso. 4 | Iso. 7 | Mean |
| Indole 3-butric acid | 50 | 26.13 | 28.67 | 27.40 | 8.32 | 13.12 | 10.72 | 26.67 | 34.47 | 30.57 | 6.53 | 10.02 | 8.28 |
| muole 5-buttic acid | 100 | 24.00 | 29.27 | 26.64 | 12.51 | 13.19 | 12.85 | 28.37 | 39.10 | 33.74 | 8.99 | 10.95 | 9.97 |
| Gibberellic acid | 50 | 27.50 | 31.87 | 29.69 | 11.15 | 12.93 | 12.04 | 31.07 | 34.90 | 32.99 | 9.86 | 13.73 | 11.80 |
| Gibberenic acid | 100 | 28.50 | 33.77 | 31.14 | 13.64 | 16.17 | 14.91 | 39.30 | 43.17 | 41.24 | 10.92 | 19.55 | 15.24 |
| N. housededening | 50 | 18.67 | 27.67 | 23.17 | 6.63 | 10.62 | 8.63 | 30.67 | 31.33 | 31.00 | 8.70 | 13.34 | 11.02 |
| N ₆ -benzyladenine | 100 | 28.33 | 28.73 | 28.53 | 8.74 | 13.32 | 11.03 | 25.67 | 31.97 | 28.82 | 7.98 | 10.01 | 8.00 |
| Manuart 250/ | 50 | 16.00 | 19.33 | 17.67 | 7.51 | 6.84 | 7.18 | 25.13 | 28.97 | 27.05 | 7.77 | 8.93 | 8.35 |
| Moncut 25% | 100 | 18.53 | 21.00 | 19.77 | 8.29 | 7.54 | 7.92 | 31.03 | 34.13 | 32.58 | 8.30 | 9.13 | 8.72 |
| Control | 0 | 15.83 | 19.00 | 17.42 | 5.20 | 7.00 | 6.10 | 26.60 | 26.27 | 26.44 | 4.63 | 6.57 | 5.60 |
| Mean | - | 22.61 | 26.59 | - | 9.11 | 11.19 | - | 29.39 | 33.81 | - | 8.19 | 11.36 | - |
| L.S.D. at 5% for Treatments (A) Concentration (B) Isolates (C) Interaction (A×B×C) | | | 3.22 2.04 2.23 7.05 | | | 3.60 2.28 2.16 6.85 | | | 3.43 2.17 2.13 6.74 | | | 2.99 1.89 1.81 5.73 | |

Effect of treatments with PGRs on content of the total phenolic compounds in treated common bean plants: Data presented in Figure (1) indicate that all common bean plants treated with PGRs (50 and 100 ppm) as soaking seeds treatments showed the highest contents of total phenolic compounds compared to the untreated plants (healthy and diseased). The highest amounts of total phenolic compounds were found in the treated by N_6 -benzyladenine at the plants concentration 100 ppm, which were infected by the isolates No. 4 (8.64 mg/g

fresh weight) and No. 7 (8.89 mg/g fresh weight). In general, increasing the concentration of the plant growth regulators in different treatments (50 and 100 ppm) increased the levels of total phenolic compounds in the treated plants.

Also, results indicate that the different treatments with PGRs enhanced the accumulation of total phenols compounds in the treated common bean plants compared to the healthy and diseased plants.

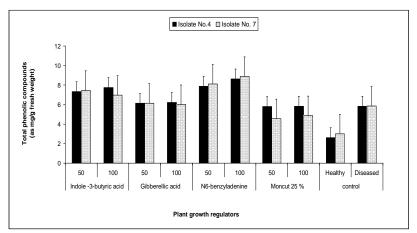


Figure 1: Accumulation of total phenolic compounds in common bean plants treated with plants growth regulators and control (healthy and diseased plants).

Discussion

Fourteen isolates of R. solani were isolated from different localities in Egypt (El-Behera and Assiut governorates). Pathogenic potentialities of the tested isolates on Giza 6 common bean cultivar were different and ranged from moderate to severe. Such results are in agreement with those reported by D'aes et al. (2011) and Rashad et al. (2012). Results found indicated that the plant growth regulators, *i.e.* indole 3-butric acid, gibberellic acid and N6-benzyladenine had a beneficial effect when it has been treated to common bean seeds as soaking treatments before sowing at different concentrations (50 and 100 ppm). The results of this work indicated that all treatments with PGRs have significantly

reduced the percent of damping-off and root rot diseases compared to the control. The obtained results are in agreement with those obtained by many other researchers (Abd El-Hai, 2015; Abd El-Hai et al., 2010; Metwally et al., 2006). N₆-benzyladenine gave the highest reduction of diseases severity (dampingoff and root rot). PGRs, i.e. indole butyric acid, gibberellic acid, ethrel and paclobutrazol at any concentration reduced significantly the severity and the incidence of Alternaria leaf spot disease (Abd El-Hai, 2015). Inhibitory effects of PGRs on fungal diseases may be due to its effects on the growth, the sporulation and the sclerotial formation of fungi (Khalifa, 2003). Application of indole acetic acid (IAA) at the concentration 20 µg/ml reduced Penecillium also

expansion infection when it was applied 48 h before pathogen inoculation in the intact fruit (Yu et al., 2009). Recent evidence, indicates IAA displays in-vitro antifungal activity against Ustilago maydis and Saccharomyces cerevisiae (Prusty et al., 2004), and in-vivo activity against Gibberella pulicaris and *Phytophthora* infestans in potato (Slininger et al., 2004; Noel et al., 2001). Similar results were obtained with P. infestans on the detached potato leaves in which a reduction in the late blight infection was observed when the leaves were incubated in a solution of IAA for 24 h and then inoculated with P. infestans (Noel et al., 2001). Data also showed that all tested PGRs were effective in increasing the percent of germination and improved plant growth characters, *i. e.* plant height and fresh weight compared to untreated plants. These results are in agreement with those obtained by Dhoran and Gudadhe (2012), they found that GA3 had a significant effect on germination rate as compared to control of IAA, IBA and NAA during light and dark periods. The stimulatory effect of GA3 on plant height may be due to increase in cell division, number of internodes and/or elongation of each internode (Deotale et al., 1998; Bruce, 1990). The highest amounts of total phenolic compounds were found in treated plants with N₆-benzyladenine at the concentration 100 ppm. Increase levels of total phenolic compounds following treatments of common bean seeds with PGRs reported herein was previously suggested to be involved in resistant mechanisms of plants to other plants pathogens (Abd El-Hai et al., 2010; Gogoi et al., 2001). The obtained results are in agreement with those

obtained by Chowdhury (2003) who found that treated seeds with plant growth regulators led to increase in total phenol, which protect plants against pathogen stress. Gogoi et al. (2001) reported that, the first step of plant defence mechanism involves a rapid accumulation of phenols at the infection site, which restricts or slows the growth of the pathogen. Abd El-Hai et al. (2010) added that ethrel and indole butyric acid increased the total phenol. In conclusion, the results of this study clearly showed that treatments with all the tested PGRs were effective in reducing the diseases severity and improved the agronomic characters of plants.

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