Egypt. J. Phytopathol., Vol. **44**, No. 2, pp. 85-102 (2016)

Impact of certain Biological Treatments in Controlling Damping-off Disease of Soybean

N.M. Abou-Zeid, Noher A. Mahmoud and Rania A. Saleh
Plant Pathol. Res. Inst., Agric. Res. Center, Giza, Egypt.

Soybean damping-off caused by *Rhizoctonia solani* is considered one of the most important diseases in Egypt. Seven isolates were collected from different locations and tested for their virulence on Crawford cv. *R. solani* (R5) was the highest virulent isolate, which significantly increased pre-and post-emergence damping-off and reduced the survived plants. The ten *Trichoderma* spp. significantly increased the growth reduction of *R. solani in vitro*. *T. hamatum* and *T. harzianum* were the most effective bioagents which gave the highest growth reduction. All treatments of the biocides Bio-Arc, Bio-Zeid, Bio-4 and Bio-Nagi, *T. hamatum* and *T. harzianum*, the green alga (*Ulva* sp.) and the fungicide Vitavax-200 decreased the incidence damping-off of the four tested cultivars (Crawford, Giza-35, Giza-111 and Giza-83) under greenhouse conditions. Bio-Nagi was the best treatment, which gave the highest survived plants followed by *T. hamatum* and Bio-Zeid when compared to the untreated control. Under field condition, all treatments decreased damping-off incidence of the four tested cultivars and increased the survived plants during two consecutive seasons (2014 and 2015). Seed yield was positively correlated with plant height, number of pods/plant, 100-seed weight. The fungicide Vitavax-200 was the most effective treatment for controlling damping- off of the four tested cultivars during the growing seasons of 2014 and15, followed by Bio-Nagi then *T. hamatum*.

Keyword: Bio-4, Bio-Arc, Bio-Nagi, Bio-Zeid, damping-off, soybean, *T. hamatum*, *T. harzianum*, *Ulva* sp and Vitavax-200.

Soybean seeds are used to make high protein meal El-Abady *et al.* (2008), which is used largely as supplement to cereal seeds in feed for poultry dairy and beef animals (Iqbal *et al*., 2003). Damping-off of soybean caused by *R. solani* is one of the most destructive diseases (Mahmoud and Abo-Elyousr, 2014). *R. solani* is a widely-distributed soil-borne plant parasitic-saprophytic fungus (Sneh *et al*., 1991). Therefore, the pathogen is difficult to control because of its persistence in the soil and wide host range. Some chemicals are effective in controlling the disease but those chemicals are expensive and not environmental friendly (Abou-Zeid *et al*., 1987).

Also, the incessant and indiscriminate application of fungicides has caused health hazards in animals and human due to residual toxicity. Therefore, alternative control methods are needed for managing this pathogen, including induced resistance by using biotic treatments (Abd El-Monaim *et al*., 2011). *Trichoderma* spp. well documented as effective biological control agents of plant diseases caused by soil-borne fungi including *R. solani* and *Pythium* spp. (Coley-Smith *et al.*, 1991; Harman *et al*., 2004 and Howell, 2006).

This work aimed to evaluate the efficacy of some different biocides and bioagents on controlling damping-off of different soybean cultivars.

Materials and Methods

1. Isolation and identification of the causal pathogens:

Soybean seedlings showing damping-off and root-rot symptoms, were collected from five soybean growing Governorates (Kafr El-Sheikh, Fayoum, Dakahlia, Ismailia and Gharbia) in Egypt. The isolated fungi were purified and identified as *R. solani* Kühn according to their morphological features (Gilman 1957 and Singh 1982), at the Unit of Identification of Microorganisms, Plant Pathol. Res. Inst., Agric. Res. Cen. (A.R.C).

1.1. Pathogenicity test:

Inocula of the seven isolates of *Rhizoctonia solani* were prepared by growing in sterilized glass bottles (500-ml) containing sorghum grain medium (150 g sorghum seeds, 50 g sand and 200 ml water) and autoclaved at 1.5 Ib for 30 min. then, incubated at 25^{o}C for 15 days. Sterilized pots (20 cm in diameter) were filled with sterilized clay soil and infested with any of the tested isolates at the rate of 3%. Each pot was planted with ten soybean seeds of the Crawford cv. (Amer, 2005 and Abd El-Monem, 2007). Three pots were used for each isolate as replicates. Non-infested pots were used as control. Percentages of pre-and post-emergence damping-off were calculated 15 and 30 days after sowing, respectively, while survived plants were recorded at the end of the experiment (60 days after sowing). Disease estimation in each stage was calculated based on the number of seeds that were sown per each pot and survived plants were assessed by subtraction number of pre-and post-emergence seedlings from the number of sown seeds.

2. Isolation of the antagonistic micro-organisms:

Different ten of *Trichoderma* isolates were isolated from the rhizosphere of healthy soybean plants collected from naturally heavily infested field, purified and identified according to their morphological features (Ellis, 1971 and Bissett, 1991) at the Identification of Micro-organisms Unit, Plant Path. Res. Inst., (A.R.C).

2.1. Antagonistic effect of Trichoderma spp., on mycelial growth of R. solani in vitro:

The antagonistic effect of ten *Trichoderma* spp. was evaluated against *R. solani* in vitro. One disc (5 mm) of 4-days old culture of any of the antagonistic isolate of *Trichoderma* spp. and an isolate of *R. solani* were inoculated at the opposite 1 cm from the plate edge in each plate (9 cm) contained 10 ml PDA medium. Three plates were used as replicates for each treatment. Plates inoculated only at one side of Petri plate with *R. solani* as a control treatment. The inoculated plates were incubated at 28$\pm$1^{o}C until the mycelial growth cover the medium surface in control treatment. The plates were then examined and radial growth of each treatment was measured to determine the reduction in radial growth according to Yeh and Sinclair (1980), by using the following formula: G= C-T/C X100

Where:
G= Percentage of fungal growth reduction.
C= Fungal growth of control (Pathogen alone).
T= Fungal growth of treatment (Pathogen against the antagonist).

2.2. Identification of the antagonistic micro-organisms:

The antagonistic isolates were identified using the Biolog-FF Micro Plats™ (Anon., 2010), at the Identification of Micro-organisms Unit, Plant Path. Res. Inst., (A.R.C).

2.3. Parasitism of T. hamatum and T. harzianum on R. solani:

A laboratory technique was developed to determine the mycoparasitic activity of the most effective *T. hamatum* and *T. harzianum* against the most virulent isolate of *R. solani* (R5), using the method described by Mohamed (2015).

In this technique a sterilized microscope glass slide was covered by a thin film sterilized diluted water agar medium. Disc from the antagonist was inoculated at one side, whereas a disc from the pathogen was inoculated at the other side of the glass slide. Inoculated slides were placed in sterilized Petri-dishes containing two filter papers (Whitman No.1) saturated with 10 ml of sterilized distilled water just to maintain humidity around the inoculated slides. Plates with slides were incubated at 18ºC. Slides were stained with lactophenol cotton-blue before examination periodically using light microscope with fixed camera to observe interaction between the pathogen and the antagonist.

3. In vivo experiment:

3.1. Effect of biological treatments on controlling damping-off of four soybean cultivars under greenhouse conditions:

Active ingredients and rate of applications are shown (Table, 1). Eight treatments were used to study their effect on controlling damping-off on four soybean cvs. (Crawford, Giza-35, Giza-111 and Giza-83) under greenhouse condition. The experiment was designed as a complete randomized block design with three replicates. Pots (20 cm in diameter) were dipped in 5% formalin solution for 10 min and left to dry in open air. The inoculum of *R. solani* was prepared as mentioned before in pathogenicity test and mixed with the soil at the rate of 3%. Inoculated potted soil was mixed thoroughly to ensure the distribution of the tested pathogen, watered daily for one week. Each of the antagonistic fungus (*T. hamatum* and *T. harzianum*) was grown on PDA plates for 10 days at 28 ± 1°C then its growth was flooded with sterile-distilled water, scraped with a slide then filtered through two layers of cheesecloth. The resulted spore suspensions were counted using haemocytometer and adjusted to approximately 10^7 spore/ml (Abou-Zeid *et al*., 2003). Bio Arc® and Bio Zeid® are commercial biocides labelled on different crops in Egypt and used at the recommended dose (Table, 1). However, Bio Nagi (*Tricoderma asperellum*) was recorded as a first record in Egypt by Abou-Zeid and Mahmoud (2012). Both Bio-Four and Bio-Nagi are still under registration and used at rate 2.5 g/L (Mahmoud *et al*., 2013). Green alga (*Ulva* sp.) used at rate 2.5 g/L (Mahmoud *et al*., 2012). All biocides and green alga (*Ulva* sp.) were obtained from Unit of Identification of Micro-organisms, Plant Path. Res. Inst. (A.R.C). *Ulva* sp. isolated from the coastal zone of Baltim city on the Mediterranean Sea, Kafr El-Sheikh Governorate then left for dry at room temperature and grounded to fine powder. The collected macroalgal samples were identified according to Dawson (1955) and Aleem (1993). Three pots were used for each treatment as replicates; each pot was planted with 10 treated soybean seeds. Disease incidence was determined as mentioned before in the pathogenicity test.

Table 1. Active ingredients of the fungicide Vitavax-200, biocides, *Ulva* sp. and the most active Trichoderma isolates

Tested products	Active ingredients	Types	Rate
Bio-ARC	*Bacillus megaterium* 6% (w/w)	Biocide	2.5 g/L
Bio-Zeid	*Trichoderma album* 2.5 % (w/w)	Biocide	2.5 g/L
Bio-Nagi	*T. asperellum*	Biocide	2.5 g/L
Bio-4	*B. subtilis + B. megaterium* *B. licheniformish + B. pumilus*	*Bioagent*	2.5 g/L
Ulva sp.	Green alga	-	2.5 g/L
Vitavax-200	Carboxin + Thiram WP 75%	Fungicide	1.5 g/kg
T. hamatum	*T. hamatum*	Fungus	$1x10^7$/ml
T. harzianum	*T. harzianum*	Fungus	$1x10^7$/ml

3.2. Effect of biological control treatments on soybean damping-off disease under field conditions:

Field experiment was carried out at Giza Res. Stat. in a field known to have the causal pathogen of damping-off *R. solani* during 2014 and 2015 growing seasons to study the efficiency of some biological treatments and the fungicide Vitavax-200 on controlling soybean damping-off, as well as their effects on some soybean crop parameters. The experiment was designed as a complete randomized block design with three replicates for each particular treatment. The field plot was 3.0 x 4.0 m^2 in four rows with about 10 -15 cm distances between sowing holes (about 2 seed/hell). All agricultural practices were done as normal for all treatments. The biological treatments and the fungicide, which were used in the greenhouse experiment, were evaluated in the current experiment. Disease assessment was measured as percentages of pre-and post-emergence damping-off and survived plants and crop parameters were also estimated. At harvest, ten plants were randomly taken from each plot to determine the following parameters: plant height, number of pods/plant, number of seeds/pod and 100-seed weight (g). Meanwhile, at full maturity, plants in each replicate were harvested and left to dry for ten days, then seed yield/plot and seed yield ton/ feddan were recorded for each treatment.

Statistical analysis:

Statistical analysis was done using complete randomize block and split design for analyzing the obtained data according to Snedecor and Cochran (1980).

Results

1. Isolation, purification and identification of the causal pathogens:

The isolated fungal isolates were purified and identified as *Rhizoctonia solani* Kühn according to their morphological features.

1.1 Pathogenicity test of seven R. solani isolates on soybean plants (Crawford cv.) under greenhouse conditions:

Data in Table (2) proved that the seven tested *R. solani* isolates were significantly varied in their virulence on inducing pre-and post-emergence damping-off disease on Crawford cv. R5 was the most aggressive one which gave the highest percentages of pre- and post-emergence damping-off. On the other hand, R7 caused the lowest percentages of damping-off. According these results R5 was chosen to carry out the greenhouse experiment.

Table 2. Pathogenicity test of seven *R. solani* isolates on soybean (Crawford cv.) under greenhouse conditions

Isolate No.	Governorate	Location	Damping-off (%)		Survived Plants (%)
			*Pre	**Post	
R1	Kafr El- Sheikh	Sakha	13.33	13.34	73.33
R2	Fayoum	Manshiet El-Fayoum	13.33	20.00	66.67
R3	Fayoum	Senoras	3.33	36.67	60.00
R4	Dakahlia	Mansoura	10.00	26.67	63.30
R5	Ismailia	Qasasen	60.00	33.30	6.67
R6	Ismailia	Abou-Sweer	46.67	33.33	20.00
R7	Gharbia	Zefta	3.33	6.67	90.00
L.S. D at 5%	-	-	8.82	10.35	8.55

*Pre: pre-emergence, **Post: post-emergence.

2. Antagonistic effect of ten Trichoderma isolates on the mycelial growth of R. solani in vitro:

Data presented in Table (3) show that all ten *Trichoderma* spp., which identified according to their morphological features, significantly reduced the radial growth of R5 *in vitro*. T9 and T3 were the most effective bio-agent, which gave the highest growth reduction of *R. solani.* These highly antagonistic isolates were evaluated for their effectiveness in controlling soybean damping-off in greenhouse and field experiments.

Table 3. Antagonistic effect of ten Trichoderma isolates on the mycelial growth of *R. solani* (R5) on PDA medium at 28±1°C for 5 days *in vitro*

Trichoderma isolates	Governorate	Location	Radial growth (mm)	Growth reduction (%)
T 1	Kafr El-Sheikh	Sakha	36	60.00
T 2	Kafr El-Sheikh	Dosok	35	61.11
T 3	Fayoum	Senoras	15	88.30
T 4	Dakahlia	Mansoura	31	65.50
T 5	Dakahlia	Aga	33	63.33
T 6	Ismailia	Abou-Sweer	38	57.70
T 7	Ismailia	Qasasen	37	58.50
T 8	Fayoum	Manshiet El-Fayoum	26	71.11
T 9	Gharbia	Zefta	5	94.44
T10	Gharbia	Tanta	22	75.50
Control	-	-	90	-
L.S.D at 5%	-	-	5	5.36

2.1. Identification of the antagonistic micro-organisms:

To confirm our morphological identification of the tested bioagents, the most active antagonistic fungal isolates were purified and identified as *Trichoderma hamatum* and *T. harzianum* using Biolog-Micro-plates.TM

2.2. Parasitism of T. hamatum and T. harzianum on R. solani:

The interaction of *T. hamatum* and *T. harzianum* against the virulent isolate of *R. solani* (R5) was examined under light compound microscope. The morphological changes in the hyphal growth of the pathogenic fungus were illustrated in Fig. (1). *Trichoderma* spp. hyphae coiled around the host hyphae of *R. solani* consisting of different types of parasitism in forms of coiling, pressing and adhering on hyphae. In addition, aggregation of the mycelium and lysis of pathogen hyphae were noticed. Control treatment showed intact and normal hyphal growth of the tested pathogen.

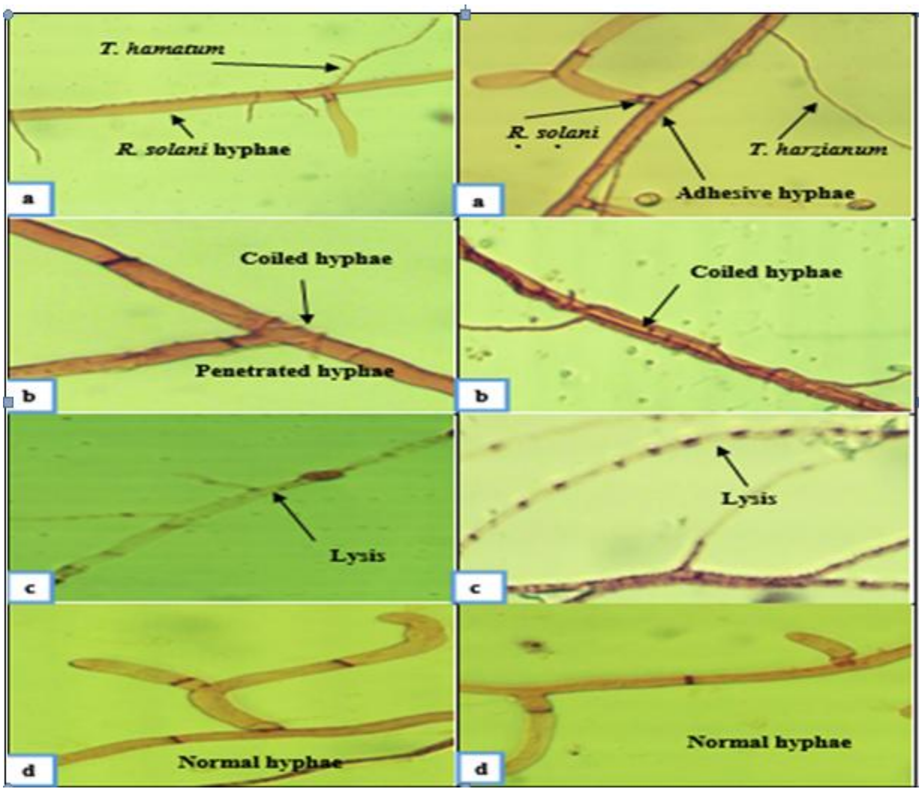

Fig.1. Mycoparasitism of *R. solani* by *T. hamatum* and *T. harzianum*

Light compound microscope (X100) showing different types of *Trichoderma* parasitism on *R. solani* hyphae: (a) the hyphae of *Trichoderma* pressing and adhering on the hyphae of *R. solani*. (b) *Trichoderma* hyphae coiled around the host hyphae of *R. solani*. (c) *Trichoderma* malformed and lysis hyphae of *R. solani.* (d) Normal hyphae of *R. solani.*

3. In vivo experiment:

3.1. Effect of biological treatments on controlling damping-off of four soybean cultivars under greenhouse conditions.

Data in Table (4) indicate that the interaction between the four tested cvs. and all tested treatments decreased soybean damping-off in the greenhouse. Giza-35 followed by Giza-83 were the lowest susceptible cvs. against *R. solani*. However, Crawford and G-111 were the most susceptible ones. Vitavax-200 significantly was superior to the other treatments in controlling damping-off on the tested cultivars. Bio-Nagi and *T. hamatum* were the best bio-treatments for controlling damping-off followed by Bio-Zeid and *T. harzianum* when compared to Vitavax-200. In addition, Bio-Nagi and *T. hamatum* were the best treatments for controlling damping-off of the four tested cvs. followed by Bio-Zeid and *T. harzianum* in controlling damping-off on G-35 and G-83 cvs. On the other hand, *Ulva* sp. was the lowest treatment in this respect.

Table 4. The effect of bioagents and biofungicide on controlling *R. solani* damping-off of different cultivars of soybean compared to fungicide in the greenhouse

Treatment	% of pre & post emergence damping off and survival plants of Soybean cultivars											
	Crawford			Giza-35			Giza-111			Giza-83		
	Pre	Post	*Sur.	Pre	Post	Sur.	Pre	Post	Sur.	Pre	Post	Sur.
Bio-Arc	40.0	13.3	46.7	20.0	13.3	66.7	40.0	6.7	53.3	33.3	6.7	60.0
Bio-Zeid	46.7	0.0	53.3	20.0	6.7	73.3	20.0	20.0	60.0	26.7	6.7	66.7
Bio-Nagi	40.0	0.0	60.0	13.3	6.7	80.0	20.0	6.7	73.3	13.3	6.7	80.0
Bio-4	46.9	6.7	46.4	46.7	0.0	53.3	40.0	13.3	46.7	20.0	13.3	66.7
Ulva sp.	60.0	6.7	33.3	33.4	13.3	53.3	46.7	13.3	40.0	46.6	6.7	46.7
T. hamatum	26.7	20.0	53.3	13.3	6.7	80.0	26.7	6.7	66.7	20.0	6.7	73.3
T. harzianum	40.0	13.3	46.7	26.7	6.7	66.7	40.0	0.0	60.0	40.0	0.0	60.0
Vitavax-	26.7	0.0	73.3	13.3	0.0	86.7	20.0	0.0	80.0	13.3	0.0	86.7
Control	73.3	20.0	6.7	73.3	0.0	26.7	93.3	0.0	6.7	66.7	20.0	13.3
Mean	44.5	8. 9	46.7	28.9	5.9	65.2	38.5	7.4	54.1	31.1	7.4	61.5

L.S.D at 5% for	Pre- emergence	Post- emergence	Survival plants
Treatments (T)	5.95	3.56	6.26
Cultivars (C)	3.27	1.96	3.44
T x C	9.80	5.87	10.31

*Sur.: Survival plants

3.2. Effect of the bioagents and biocides compared to fungicide on controlling damping-off of four soybean cultivars in the field experiment during growing season's 2014 and 2015.

In this experiment, efficacy of different tested treatments *i.e.* biocides, the fungicide Vitavax- 200 and effective bioagents were evaluated against pre-and post-emergence damping-off on four cvs. in the field. Data in Tables (5-a, b, c and d) showed that the fungicide Vitavax-200 was the most effective treatment for controlling damping- off on the four tested cvs. in both growing seasons, followed by Bio-Nagi, *T. hamatum*. However, *Ulva* sp. was the lowest effective treatment in this respect.

Table 5a. Effect of bioagents, biocides and the fungicide Vitavax-200 on controlling damping-off of Giza-35 soybean cultivar under field conditions during 2014 and 2015 growing seasons

Treatment	2014 growing season					2015 growing season				
	Damping-off %			*Effic. %	Survived plants%	Damping-off %			Effic. %	Survived plants %
	Pre	Post	Total			Pre	Post	Total		
Bio-Arc	11.0	6.0	17.0	49.40	83.0	9.7	5.3	15.0	49.66	85.0
Bio-Zeid	8.4	4.1	12.5	62.80	87.5	6.0	3.3	9.3	68.79	90.7
Bio -Nagi	4.6	2.9	7.5	77.68	92.5	3.0	1.7	4.7	84.23	95.3
Bio-4	11.5	5.7	17.2	48.81	82.8	9.8	5.4	15.2	48.99	84.8
Ulva sp.	15.2	8.5	23.7	29.46	76.3	14.6	7.1	21.7	27.18	78.3
T. hamatum	7.6	3.3	10.9	67.56	89.1	5.7	3.1	8.8	70.47	91.2
T.harzianium	7.8	3.6	11.4	66.07	88.6	6.5	3.5	10.0	66.44	90.0
Vitavax-200	3.7	2.0	5.7	83.04	94.3	2.4	1.3	3.7	87.58	96.3
Control	21.7	11.9	33.6	0.00	66.4	19.2	10.6	29.8	0.00	70.2
L.S.D at 5%	0.95	1.19	-	1.43	0.58	1.14	0.70	-	1.77	0.72

*Effic. % (Efficiency %) = Total damping off in treatment – Total damping off in control / Total damping off in control x 100

Table 5b. Effect of bioagents, biocides and the fungicide Vitavax-200 on controlling damping-off of Crawford soybean cultivar under field conditions during 2014 and 2015 growing seasons

Treatments	2014 growing season					2015 growing season				
	Damping-off %			Effic. %	Survived plants%	Damping-off %			Effic. %	Survived plants %
	Pre	Post	Total			Pre	Post	Total		
Bio-Arc	17.2	9.9	27.1	34.06	72.9	15.9	9.5	25.4	32.63	74.6
Bio-Zeid	14.9	8.1	23.0	44.04	77.0	12.5	8.5	21.0	44.03	79.0
Bio -Nagi	10.0	6.7	16.7	59.37	83.3	7.9	4.8	12.7	66.31	87.3
Bio-4	17.5	10.8	28.3	31.14	71.7	16.3	9.8	26.1	30.76	73.9
Ulva sp.	18.3	10.9	29.2	28.95	70.8	17.5	10.3	27.8	26.26	72.2
T. hamatum	13.0	7.2	20.2	50.85	79.8	10.6	6.5	17.1	54.64	82.9
T.harzianium	15.3	10.0	25.3	38.44	74.7	14.1	9.4	23.5	37.67	76.5
Vitavax-200	6.7	4.1	10.8	73.72	89.2	4.3	4.0	8.3	77.98	91.7
Control	26.6	14.5	41.1	0.00	58.9	24.6	13.1	37.7	0.00	62.3
L.S.D at 5%	2.76	1.47	-	4.34	4.92	1.96	0.99	-	2.42	5.79

Table 5c. Effect of bioagents, biocides and the fungicide Vitavax-200 on controlling damping-off of Giza-111soybean cultivar under field conditions during 2014 and 2015 growing seasons

Treatments	2014 growing season					2015 growing season				
	Damping-off %			Effic. %	Survived plants%	Damping-off %			Effic. %	Survived plants %
	Pre	Post	Total			Pre	Post	Total		
Bio-Arc	15.8	7.6	23.4	33.52	76.6	12.7	7.0	19.7	40.12	80.3
Bio-Zeid	11.0	7.1	18.1	50.55	81.9	9.7	5.1	14.8	55.02	85.2
Bio -Nagi	7.50	4.6	12.1	66.94	87.9	5.9	2.5	8.4	74.47	91.6
Bio-4	16.4	9.5	25.9	29.23	74.1	14.8	8.2	23.0	30.09	77.0
Ulva sp.	17.1	9.9	27.0	26.23	73.0	15.4	9.0	24.4	25.84	75.6
T. hamatum	10.5	6.9	17.4	52.45	82.6	9.6	5.1	14.7	55.32	85.3
T.harzianium	12.7	7.5	20.2	44.81	79.8	11.5	5.5	17.0	48.33	83.0
Vitavax-200	5.6	3.6	9.2	74.86	90.8	4.1	2.4	6.5	80.24	93.5
Control	23.7	12.9	36.6	0.00	63.4	21.7	11.2	32.9	0.00	67.1
L.S.D at 5%	3.03	2.16	-	2.98	1.13	2.55	1.37	-	3.29	2.61

Table 5d. Effect of bioagents, biocides and the fungicide Vitavax-200 on controlling damping-off of Giza-83 soybean cultivar under field conditions during 2014 and 2015 growing seasons

Treatments	2014 growing season					2015 growing season				
	Damping-off %			Effic. %	Survived plants%	Damping-off %			Effic. %	Survived plants %
	Pre	Post	Total			Pre	Post	Total		
Bio-Arc	13.6	8.0	21.6	38.64	78.4	12.1	6.9	19.0	39.49	81.0
Bio-Zeid	10.5	6.1	16.6	52.84	83.4	8.8	4.2	13.0	58.60	87.0
Bio -Nagi	4.9	3.7	8.6	75.57	91.4	4.7	2.0	6.7	78.66	93.3
Bio-4	15.8	8.6	24.4	30.68	75.6	13.5	7.8	21.3	32.17	78.7
Ulva sp.	16.7	10.3	27.0	23.30	73.0	15.1	8.7	23.8	24.20	76.2
T. hamatum	9.5	5.7	15.2	56.82	84.8	7.6	4.1	11.7	62.74	88.3
T.harzianium	12.2	5.9	18.1	48.58	81.9	9.3	5.4	14.7	53.18	85.3
Vitavax-200	3.3	1.1	4.4	87.50	95.6	2.6	1.4	4.0	87.26	96.0
Control	22.1	13.1	35.2	0.00	64.8	20.5	10.9	31.4	0.00	68.6
L.S.D at 5%	1.28	0.93	-	2.24	1.61	1.15	1.26	-	0.96	1.22

3.3. Effect of the tested treatments on plant growth parameter and yield components.

Data in Tables (6-a, b, c and d) and (7-a, b, c and d) illustrated the effect of the tested biological treatments and the check fungicide on yield and yield components (plant height, No. of pods/plant and 100 seed weight) of four soybean cultivars under field conditions during growing seasons 2014 and 15. Obtained data indicate that all the tested bioagents, biocides and the fungicide Vitavax-200 significantly increased yield and the yield components of four soybean cultivars during the two growing seasons (2014 and 2015).

Table 6a. Effect of bioagents, biocides and the fungicide Vitavax-200 on some crop components of Crawford soybean cultivar under field conditions during 2014 growing season

Treatments	Plant height (cm)	No. of pods /plant	100 seed weight (g)	Seed yield	
				Kg/plot (12 m^2)	Ton /fed
Bio-Arc	83.80	44.55	19.00	3.87	1.355
Bio-Zeid	95.80	48.48	20.80	4.15	1.453
Bio -Nagi	104.75	51.25	22.60	4.62	1.617
Bio-4	74.00	44.32	17.90	3.71	1.299
Ulva sp.	85.80	45.15	16.40	3.18	1.113
T. hamatum	101.25	50.69	22.80	4.42	1.547
T. harzianum	83.00	43.21	19.60	3.65	1.278
Vitavax-200	98.25	49.31	21.50	4.35	1.523
Control	62.75	33.59	13.90	2.83	0.991
L.S.D at 5%	4.15	0.58	0.45	0.71	0.250

Table 6b. Effect of bioagents, biocides and the fungicide Vitavax-200 on some crop components of Giza-111 soybean cultivar under field conditions during 2014 growing season

Treatments	Plant height (cm)	No. of pods /plant	100 seed weight (g)	Seed yield	
				Kg/plot (12 m^2)	Ton /fed
Bio-Arc	91.00	50.14	21.91	3.62	1.267
Bio-Zeid	95.80	56.82	22.53	4.34	1.519
Bio -Nagi	113.20	66.94	25.83	4.85	1.698
Bio-4	90.20	50.39	21.08	3.53	1.236
Ulva sp.	86.25	44.46	18.09	3.30	1.155
T. hamatum	108.25	65.20	23.90	4.62	1.677
T. harzianum	93.80	55.96	21.29	3.53	1.236
Vitavax-200	106.80	60.40	25.00	4.79	1.666
Control	75.00	37.40	14.80	3.00	1.050
L.S.D at 5%	3.60	0.44	1.07	0.61	0.212

Table 6c. Effect of bioagents, biocides and the fungicide Vitavax-200 on some crop components of Giza-35 soybean cultivar under field conditions during 2014 growing season

Treatments	Plant height (cm)	No. of pods /plant	100 seed weight (g)	Seed yield	
				Kg/plot (12 m^2)	Ton /fed
Bio-Arc	94.75	55.40	22.69	4.43	1.551
Bio-Zeid	96.00	64.10	24.12	4.62	1.617
Bio -Nagi	120.25	71.36	27.18	5.19	1.817
Bio-4	95.80	67.03	23.28	4.59	1.607
Ulva sp.	77.40	62.33	22.85	4.22	1.477
T. hamatum	111.00	69.8	25.83	5.17	1.811
T. harzianum	100.00	54.02	21.37	4.67	1.635
Vitavax-200	107.80	70.91	26.40	5.13	1.796
Control	78.75	51.25	17.81	3.40	1.190
L.S.D at 5%	1.87	2.81	0.45	0.69	0.240

Table 6d. Effect of bioagents, biocides and the fungicide Vitavax-200 on some crop components of Giza-83 soybean cultivar under field conditions during 2014 growing season

Treatments	Plant height (cm)	No. of pods /plant	100 seed weight (g)	Seed yield	
				Kg/plot (12 m^2)	Ton /fed
Bio-Arc	84.40	53.10	22.97	4.17	1.460
Bio-Zeid	100.00	62.99	23.36	4.31	1.509
Bio -Nagi	111.00	69.77	25.07	5.23	1.831
Bio-4	82.30	55.94	21.64	4.11	1.439
Ulva sp.	81.75	43.10	19.28	3.72	1.302
T. hamatum	107.50	64.10	24.76	4.41	1.544
T. harzianum	82.50	56.90	24.35	4.13	1.446
Vitavax-200	98.50	67.67	23.20	4.95	1.733
Control	66.75	36.43	16.67	2.90	1.015
L.S.D at 5%	8.95	1.54	1.13	0.86	0.302

Table 7a. Effect of bioagents, biocides and the fungicide Vitavax-200 on some crop components of Crawford soybean cultivar under field conditions during 2015 growing season

Treatments	Plant height (cm)	No. of pods /plant	100 seed weight (g)	Seed yield	
				Kg/plot (12 m^2)	Ton /fed
Bio-Arc	89.98	46.12	19.33	4.12	1.442
Bio-Zeid	106.61	52.79	20.06	4.53	1.586
Bio -Nagi	115.27	55.60	23.72	4.95	1.733
Bio-4	87.74	47.70	19.18	4.01	1.402
Ulva sp.	87.15	46.87	16.85	3.43	1.201
T. hamatum	107.68	54.78	22.96	4.60	1.610
T. harzianum	87.95	45.37	19.96	3.77	1.319
Vitavax-200	105.01	53.76	22.27	4.14	1.449
Control	60.50	34.84	13.97	3.06	1.071
L.S.D at 5%	0.91	1.06	0.97	0.83	0.291

Table 7b. Effect of bioagents, biocides and the fungicide Vitavax-200 on some crop components of Giza-111soybean cultivar under field conditions during 2015 growing season

Treatments	Plant height (cm)	No. of pods /plant	100 seed weight (g)	Seed yield	
				Kg/plot (12 m^2)	Ton /fed
Bio-Arc	100.67	52.26	17.40	3.94	1.379
Bio-Zeid	99.39	63.12	22.90	4.72	1.652
Bio -Nagi	118.24	68.42	23.20	5.27	1.845
Bio-4	96.60	53.04	20.30	3.84	1.344
Ulva sp.	92.37	48.16	20.80	3.58	1.253
T. hamatum	109.37	66.76	25.00	5.21	1.823
T. harzianum	100.46	56.30	19.00	3.84	1.344
Vitavax-200	114.38	65.34	21.90	5.02	1.757
Control	87.82	39.26	15.30	3.26	1.141
L.S.D at 5%	10.00	1.18	0.54	0.79	0.276

Table 7c. Effect of bioagents, biocides and the fungicide Vitavax-200 on some crop components of Giza-35 soybean cultivar under field conditions during 2015 growing season

Treatments	Plant height (cm)	No. of pods /plant	100 seed weight (g)	Seed yield	
				Kg/plot (12 m^2)	Ton /fed
Bio-Arc	102.42	63.58	22.40	4.68	1.638
Bio-Zeid	103.78	67.63	24.70	4.92	1.722
Bio -Nagi	129.99	74.64	28.70	5.58	1.953
Bio-4	103.56	65.50	23.90	4.87	1.705
Ulva sp.	83.67	63.70	23.50	4.50	1.575
T. hamatum	119.99	73.21	27.80	5.51	1.929
T. harzianum	108.10	66.39	22.10	4.97	1.740
Vitavax-200	111.13	71.95	25.20	5.49	1.922
Control	78.64	52.54	18.30	3.62	1.267
L.S.D at 5%	1.040	0.19	1.93	0.89	0.313

Table 7d. Effect of bioagents, biocides and the fungicide Vitavax-200 on some crop components of Giza-83 soybean cultivar under field conditions during 2015 growing season

Treatments	Plant height (cm)	No. of pods /plant	100 seed weight (g)	Seed yield	
				Kg/plot ($12\ m^2$)	Ton /fed
Bio-Arc	90.92	58.02	23.80	4.65	1.628
Bio-Zeid	103.94	65.32	24.30	4.79	1.677
Bio -Nagi	112.99	73.40	26.60	5.56	1.946
Bio-4	80.29	63.89	18.10	4.61	1.614
Ulva sp.	93.09	59.59	19.90	4.03	1.411
T. hamatum	110.56	67.94	25.00	4.91	1.719
T. harzianum	90.06	57.80	22.30	4.66	1.631
Vitavax-200	106.60	72.55	26.40	5.39	1.887
Control	68.08	40.50	17.90	3.27	1.145
L.S.D at 5%	2.11	0.72	0.43	0.63	0.221

Discussion

Soybean is one of the world's most important sources of oil and protein. It has the highest protein content among leguminous crops (El-Abady *et al.*, 2008). Damping-off of soybean caused by *R. solani* is one of the most destructive common soil borne fungus (Mahmoud and Abo-Elyousr, 2014). The disease is commonly observed early in the growing season when the soil moisture content is often high. Characteristic symptoms include root rotting, often originating at the distal tip of the young root and gradual yellowing and wilting of foliage (Dubey and Dwivedi, 2000). This pathogen is difficult to control because of their persistence in the soil and wide host range (Nelson *et al.,* 1996).

The results of the current study indicated that *R. solani* isolates were significantly deferred in their virulence on Crawford cv. *R. solani* isolate R5, was the highest virulent one which significantly increased pre- and post-emergence damping-off and reduced the survived plants. (Mahmoud and Abo-Elyousr, 2014 and Yang 1999).

Light compound microscope showed the effect of most effective *T. harzianum* and *T. hamatum* on *R. solani* hyphae, which caused hyper-parasitism, coiling and adhesive hyphae, lysis and changing the colour in the growing hyphae of the pathogen. These findings are in agreement with those obtained by Ahmed (2005); Harman (2006) and Mohamed (2015) who showed that *Trichoderma* spp. have been widely used as antagonistic fungal agents against several pathogenic fungi. *Trichoderma* spp. coiled around the pathogen hyphae, adhered to parallel hyphae looking for penetration within it, causing malformation and complete destruction for the parasitized mycelium.

Some chemicals are effective in controlling this disease but those chemicals are expensive and not environmental friendly. Therefore, alternative control methods are needed for managing this pathogen.

Also, the incessant and indiscriminate application of fungicides has caused health hazards to animals and human due to residual toxicity (Agrios, 2005). All different treatments of biocides, effective bioagents, *Ulva* sp. and the fungicide (Vitavax 200) decreased the incidence of damping-off on the four tested cvs. in both greenhouse and field experiments. Bio-Nagi and *T. hamatum* were the best treatments for controlling damping-off followed by Bio-Zeid and on the four tested soybean cultivars.

Biological control of *Trichoderma* species against plant pathogens has been studied extensively (Hjelijord *et al.*, 2001 and Krause *et al.*, 2001). A number of commercial formulations, based on *T. harzianum* and *T. virens* are available for the control of soil borne and foliar diseases in a range of horticultural crops (Samuels *et al.*, 1996). The best treatment reduced the incidence of bean damping off *R. solani* was obtained by *T. hamatum*, followed by *T. viride*, *T. album* and *T. harzianum* respectively (Abd-El-Khair *et al*., 2011). All ten *Trichoderma* spp. significantly decreased the radial growth of *R. solani in vitro*. Both *T. hamatum* and *T. harzianum* were the most effective bio agents, which gave the highest growth reduction of *R. solani*. These in results agreement with these obtained by Harmann *et al*. (2004); Howell (2006) and Siame to *et al*. (2011) who reported that the efficiency of *T. harzianum* in inhibiting fungal growth could be through competition for space and nutrients, mycoparasitism and production of antibiotic compounds. In addition, they found that the hyphae of *T. harzianium* coil around the hyphae of the pathogen and penetrate the host mycelium through degrading cell wall by secretion hydrolytic enzymes followed by assimilation of cell contents. Barari and Foroutan (2016) reported that the inhibition zone could be due to the effect of diffusible inhibitory substances produced by the *Trichoderma* strains, which suppressed the growth of *M. phaseolina*. *Trichoderma* spp. are well documented as effective biological control agents of plant diseases caused by soil borne fungi including *R. solani* and *Pythium* spp. (Coley-Smith *et al.*, 1991; Harman *et al*., 2004 and Howell, 2006).

Greenhouse and field trials showed that treatments with *Trichoderma* isolates could decrease damping-off incidence, increased the 100-seed weight and total yield of the four-tested soybean cvs. In the current study, Vitavax-200 followed by Bio-Nagi, *T. hamatum* and Bio-Zeid were effective for controlling damping-off compared with control. These are results in agreement with the reported by Hudge (2015) which observed that all the bio agents significantly reduced seed rot over inoculated control. Also, Barari and Foroutan (2016) found that Trichoderma isolates tested could be used for controlling soybean charcoal rot. Likewise, similar to the results revealed by Yedidia *et al*. (1999) who showed that treatment of cucumber plants in soil with *T. harzianum* (T-203) resulted in increasing of root area and cumulative root lengths. They also mentioned that there is significant increase in dry weight, shoot length and leaf area over that of the untreated control, which *T. harzianum* modifies and strengthens plant cell walls in cucumber. Ilyas *et al*. (1985) reported that *T. harzianum* isolates significantly increased plant height and dry stem weight percentages compared to non-treated healthy sunflower plants in a pot experiment. Yedidia *et al*. (2001) suggested a direct role for *T. harzianum* in mineral uptake by the plant at a very early stage of the fungal-plant association. Maisuria and

Patel (2009) found that *T. viride* promoted an increased root length, shoot length and seed germination. Hussein *et al*. (2006) and Kandil *et al*. (2012) found that soybean cvs. showed high difference in seed yield and its components (number of pods/ plant, 100 seed weight and seed yield). Giza -35 was the best resistant cvs. in this regard followed by Giza-83 and Giza-111 among the four cv. These results were in agreement with in these reported by Abd El-Mohsen *et al*. (2013).

In conclusion, the fungicide Vitavax-200 and the tested biological treatments such as *Trichoderma harzianum* and *T. hamatum* as well as the two biocides, *i.e* Bio-Nagi and Bio-Zeid were the most effective treatments in reducing soybean damping-off, as well as improving soybean crop parameters. This might lead to suggest using of biological control as an alternative to synthetic fungicides.

References

Abd-El-Khair, H.; Khalifa, R.K.M. and Haggag, Karima H. 2011. Effect of *Trichoderma* species on damping off diseases incidence, some plant enzymes activity and nutritional status of bean plants. *J. American Sci.*, **7**(1):156-167.

Abd El-Mohsen, A.A., Mahmoud, G.O. and Safina, S.A. 2013. Agronomical evaluation of six soybean cultivars using correlation and regression analysis under different irrigation regime conditions. *J. Plant Breed. Crop Sci.*, **5**: 91-102.

Abd El-Monaim, M.F.; Ismail, M.E. and Morsy, K.M. 2011. Induction of systematic resistance in soybean plants against Fusarium wilt disease by seed treatment with benzothiadiazole and humic acid. *Afri. J. Agric. Res.*, **6**: 5747-5756.

Abd El-Monem H.M. 2007. Biochemical and Pathological Studies on Important Crops in New Reclaimed Lands. Ph.D. Thesis, Agric. Sci. (Biochemistry), Fac. Fac. Agric., Cairo Univ., 216 p.

Abou-Zeid, N.M.; Abada, K.A. and Ragab, Mona, M. 1987. Effect of different fungicides *in vitro* and *in vivo* on the control of soybean damping-off disease. *Agric. Res. Rev.*, **65**: 263 - 269.

Abou-Zeid, N.M.; Arafa, M.K. and Attia, S. 2003. Biological control of post emergence diseases of faba bean, lentil and chickpea in Egypt. *Egypt. J. Agric. Res.*, **81**(4): 1491-1503.

Abou-Zeid, N.M. and Mahmoud, Noher, A. 2012. First record of *Tricoderma asperellum* in Egypt. *Egypt. J. Phytopathol.*, **40**(1), (Abstract).

Agrios, G.N. 2005. Plant Pathology. 5th Ed. Academic Press, San Diego, USA.

Ahmed, M.F. 2005. Effect of adding some biocontrol agents on non-target microorganisms in root diseases infecting soybean and broad bean plants. M.Sc. Thesis, Fac. Agric. Moshtohor, Benha Univ., 142 p.

Aleem, A.A. 1993. The Suez Canal as habitat and pathway for marine algae and seagrasses. Marine Science of the North-West Indian Ocean and adjacent water. Proceeding of the Mabalith-John Morray. Int. Symp. Egypt, 3-6.

Amer, M.A. 2005. Reaction of selected soybean cultivars to Rhizoctonia root rot and other damping-off disease agents. *Commun. Agric. Appl. Biol. Sci.*, **70**(3): 381-390.

Anonymous 2010. Micro station™ system release 3.50 Biolog, Inc. 21124 Cabot Blvd Hayward, CA 94545 U.S.A.

Barari, H. and Foroutan, A. 2016. Biocontrol of soybean charcoal root rot disease by using *Trichoderma* spp. Cercetări Agronomic Moldova, **166**:41-51.

Bissett, J. 1991. A revision of the genus *Trichoderma* II: Infragenic classification. *Can. J. Bot.*, **69**: 2357-2317.

Coley-Smith, J.R.; Ridout, C.J.; Mitchell, C.M. and Lynch, J.M. 1991. Control of buttom rot disease of lettuce (*Rhizoctonia solani*) using preparations of *Trichoderma viride*, *T. harzianum* or tolclofos-methyl. *Plant Pathol.*, **40**: 359-366.

Dawson, E.Y. 1955. Pictured-Key to the Common Genera of Macroscopic Marine Algae of the United States. In: How to know the seaweeds. WM. C. Brown Company Publishers, Dubuque, Iowa, pp. 28-179.

Dubey, S.C. and Dwivedi, R.P. 2000. Diseases of leguminous crops caused by *Rhizoctonia solani* and their management. Advances in plant disease management. Advance Publishing Concept, New Delhi, India.

El-Abady, M.I.; Seadh, S.E.; Attia, A.N. and El-Saidy, Aml, E.A. 2008. Impact of foliar fertilization and its time of application on yield and seed quality of soybean. The 2*nd* Field Crops Conference, FCRI, AV, Giza, Egypt.

Ellis, M.B. 1971. Dematiaceous Hyphmycetes. Commonwealth Mycological Institute, Kew, Surrey, England, 608 p.

Gilman, J.C. 1957. A Manual of Soil Fungi. 2nd Ed., The Iowa State College Press, Ames, Iowa, USA, 450 p.

Harman, G.E. 2006. Overview of mechanisms and uses of *Trichoderma* spp. *Phytopathology*, **96**:190-194.

Harman, G.E.; Howell, C.R.; Viterbo, A.; Chet, I. and Lorito, M. 2004. *Trichoderma* species-opportunistic, avirulent plant symbionts. Nat. *Rev. Microbiol.*, **2**: 43-56.

Hjelijord, L.G.; Stensvand, A. and Transmo, A. 2001. Antagonism of nutrient activated conidia of *Trichoderma harzianum* (atroviridae) P1 against *Botrytis cinerea. Phytopathology*, **91**:1172-1180.

Howell, C.R. 2006. Understanding the mechanisms employed by *Trichoderma virens* to effect biological control of cotton diseases. *Phytopathology*, **96**: 178-180.

Hudge, B.V. 2015. Management of damping-off disease of soybean caused by *Pythium ultimum* Trow. Int. *J. Curr. Microbiol. App. Sci.*, **4**: 799-808.

Hussein, T.F.; Darweish, G.A. and Rattiba, M.M. 2006. Effect of planting dates on growth yield and quality of some soybean cultivars. *J. Agric. Sci., Mansoura Univ*., **31**: 587-594.

Ilyas, M.B.; Randhawa, M.A. and Ayub, M. 1985. Biological control of charcoal rot of sunflower. *Pakistan J. Agri. Sci.*, **23**: 67-73.

Iqbal, S.; Mahmood T.; Tahira, A.M.; Anwar, M. and Sarwar, S. 2003. Path-coefficient analysis in different genotypes of soybean (*Glycine max* (L) Merrill). *Pakistan J. Biol. Sci.*, **6**: 1085-1087.

Kandil, A.A.; Sharief A.E.; Morsy A.R and El-Sayed, M.A.I. 2012. Performance of some promising genotypes of soybean under different planting dates using biplots analysis. *J. Basic Appl. Sci.*, **8**:379-385.

Krause, M.S.; Madden, L.V. and Hoitink, H.A.J. 2001. Effect of potting mix microbial carrying capacity on biological control of Rhizoctonia damping-off of radish and Rhizoctonia crown and root rot of poinsettia. *Phytopathology*, **91**: 1116-1123.

Mahmoud, A.F.A. and Abo-Elyousr, K.A.M. 2014. Genetic diversity and biological control of *Rhizoctonia solani* associated with root rot of soybean in Assiut Governorate, Egypt. *J. Plant Physiol., Pathol.*, **2**:4.

Mahmoud, Noher, A.; Ashrei, A.A.M. and Mekhemar, G.A.A. 2012. Evaluation of some products in controlling chocolate spot of faba bean and their effect on the growth and yield parameters. *Egypt. J. Appl. Sci*., **90**:991-1000.

Mahmoud, Noher, A.; Khalifa, M.M.A. and Abou-Zeid, N.M. 2013. Performance of some biofungicides on the most onion economic disease compared to recommended fungicide in Egypt. *Egypt. J. Appl. Sci*., **28:**66-92.

Maisuria, K.M., and Patel, S.T. 2009. Seed germ inability, root and shoot length and vigour index of soybean as influenced by rhizosphere fungi. *Karnataka J. Agri. Sci.*, **22**: 1120-1122.

Mohamed, A.M. 2015. Efficiency of some bioagents and nemastop compound in controlling damping-off and root-rot diseases on peanut plants. *Int. J. Adv. Res. Biol. Sci.*, **2**: 77-86.

Nelson, B.; Helms, T.; Christianson, T. and Kural, I. 1996. Characterization and pathogenicity of Rhizoctonia from soybean. *Plant Dis.*, **80**: 74-80.

Samuels, G.J. 1996. *Trichoderma*: a review of biology and systematics of the genus. *Mycol Res.*, **100**: 923-935.

Siameto, E.N.; Okoth, S.; Amugune, N.O. and Chege, N.C. 2011. Molecular characterization and identification of biocontrol isolates of *Trichoderma harzianum* from Embu District, Kenya. *Tropical Subtropical Agroecosystems*, **13**: 81 – 90.

Singh, R.S. 1982. Plant pathogens "the fungi". Oxford and IBH Publishing Co. New Delhi, Bombay, Calcuta, 443 p.

Snedecor, G.W. and Cochran W.G. 1980. Statistical Methods. 7*th* ed. Ames: Iowa State University Press.

Sneh, B.; Burpee, L. and Ogashi, A. 1991. Identification of *Rhizoctonia* species. *Amer. Phytopathol. Soc.* Press. St. Paul, Minnesota, 133p.

Yang, X.B. 1999. Rhizoctonia dampig-off and root-rot. Pages: 45-46. In: Compendium of Soybean Diseases. Hartmann, G. L.; Sinclair, J. B. and Rupe, J.C. (eds.). APS, St Paul, MN, USA.

Yedidia, I.I.; Benhamou, N. and Chet, I. 1999. Induction of defense responses in cucumber plants (*Cucumis sativus* L.) by the biocontrol agent *Trichoderma harzianum. Appl. Environ. Microbiol.*, **65**: 1061-1070.

Yedidia, I.; Srivastva, A.; Kapulnik, Y. and Chet, I. 2001. Effective of *Trichoderma harzianum* microelement concentration and increased growth of cucumber plants. *Plant Soil*, **235**: 235-242.

Yeh, C.C. and Sinclair, J.B. 1980. Effect of *Chaetomium cupreum* on germination and antagonism to other seed borne fungi of soybean. *Plant Dis.,* **64**: 468-470.

(Received 05/09/2016;
in revised form 10/10/2016

تأثير كفاءة بعض المعاملات الحيوية في مقاومة مرض موت بادرات فول الصويا

ناجى محمد أبو زيد، نهير عبد النظير محمود ، رانيا عباس صالح

معهد بحوث امراض النباتات - مركز البحوث الزراعية- الجيزة.

يعتبر مرض موت البادرات المتسبب عن فطر *Rhizoctonia solani* من أهم الأمراض التى تصيب فول الصويا في مصر. تم تجميع سبع عزلات من مناطق مختلفة من الفطر ريزوكتونيا سولاني وتم اختبارها لدراسة قدرتها المرضية على الصنف الحساس (كروفورد). تميزت عزلة ريزوكتونيا سولاني R5 بضرواتها حيث سببت زيادة معنوية فى نسبة موت البادرات قبل وبعد إنبثاقها فوق سطح التربة كما قللت من نسب النباتات النامية. سببت عزلات فطر *Trichoderma* spp. العشرة انخفاض معنوى في النمو الميسليومى للفطر *R. solani* تحت ظروف المعمل. كانا الفطران *T. hamatum* و *T. harzianum* هما أكثر المعاملات التي تم استخدامها فى خفض النمو الميسليومى للفطر الريزوكتونيا سولانى. قللت كل المعاملات الحيوية المختبرة سواء من المبيدات الحيوية الأربعة المختبرة (بيوأرك، بيوزيد، بيو فور، بيوناجي)، وفطري *T. hamatum* و *T. harzianum*، وطحلب أولفا والمبيد الكيماوى (فيتافاكس 200) من الإصابة بمرض موت البادرات للأصناف الأربعة المختبرة (كروفورد، جيزة- 111، جيزة- 35 وجيزة- 83) تحت ظروف العدوى الصناعية بالصوبة. كان المبيد الحيوى بيوناجى هو أفضل المعاملات فى مكافحة مرض موت البادرات والذى أعطى أعلى نسبة نباتات نامية متبوعا بالمعاملة بفطر *T. hamatum* والمبيد الحيوي بيوزيد مقارنة بالكنترول. تحت ظروف العدوى الطبيعية بالحقل خلال موسمى 2014، 2015 أدى استخدام كل المعاملات الحيوية المختبرة إلى خفض الإصابة بمرض سقوط البادرات للأربعة أصناف المختبرة مع زيادة نسبة النباتات النامية ومحصول البذور والذى يرتبط ارتباطا إيجابيا وثيقا بالصفات المحصولية من ارتفاع النبات، عدد القرون/نبات، ووزن 100 بذرة. كان المبيد الحيوى بيوناجى هو أكثر المعاملات فعالية فى خفض نسبة سقوط البادرات في موسمى الزراعة 2014 و2015 تلاه في ذلك المعاملة بـ *T. hamatum* ثم المبيد الحيوي بيوزيد مقارنة بالكنترول.