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Biocontrol of Tomato Root-Rot Caused by *Rhizoctonia solani*

Naglaa A.S. Muhanna; Seham, S.M. Ragab and Abeer A. El Ghunm

Plant Pathol. Res. Inst., Agric .Res. Centre, Giza, Egypt.

Four experiments were designed to study the effect of fertilization (NPK), *Trichoderma harzianum*, mycorrhiza and an algae extract, on tomato stem canker disease severity. Tomato plants (*Lycopersicon esculentum* Mill.) were grown in *Rhizoctonia solani* infested soil under protected greenhouse conditions, to study the effect of some biocontrol agents on the disease severity and to assay the total phenols in tomato tissues. The results showed that inoculation with *T. harzianum* alone as a bioagent resulted in a distinct decrease in disease severity and to less extent after inoculation with AM fungi, algae extract, fertilizer (NPK), either single or in combination. The mycorrhiza treatments, however, significantly increased the total phenols in the infected tomato plants compared to the infected check plants. The most promising elevation in total phenol was recognized in infected plants treated with a mixture of (AM) mycorrhiza, *Chlorella vulgaris* (A1) and *Spirulina platensis* (A2). *Trichoderma harzianum* plus the usual NPK dose fertilization with, produced similar effect compared to check plants infected with *R. solani* alone, similar results were recognized in case of mixing Trichoderma and mycorrhiza treatment.

Keywords: Algae, *Lycopersicon esculentum* Mill., mycorrhizae, NPK, phenolic compounds, *Rhizoctonia solani*, tomato and *Trichoderma harzianum.*

Rhizoctonia solani Kühn, the anamorph of *Thanatephorus cucumeris* (Frank) Donk, is a soilborne pathogen that causes diseases on many plant species world-wide including agricultural and vegetable crops (Ogoshi, 1987), within different anastomosis groups (Bolkan and Ribeiro, 1985). Tomato (*Lycopersicon esculentum* L.) is a popular vegetable widely grown in the temperate and in the tropics which is an excellent source of vitamin A and vitamin C as well as antioxidants (Renata *et al.*, 2013), along with iron and phosphorus. Tomato production under greenhouse conditions is subject to several pathogens, including *R. solani*, *Fusarium oxysporum* f.sp. *lycopersici* and *Pyrenochaeta lycopersici* which affect the plant stand, the yield and its quality (Campbell and Shishkoff, 1990). Disease control in the greenhouse is largely dependent on fungicides, thus biological control has been utilized to avoid pesticides hazards against a wide number of agricultural pests including fungi, insects and weeds (Stiling and Cornelissen, 2005). A voluminous literature has been accumulated concerning the biological control of plant diseases on different crops, and still attracting interest of many pathologists (Cook and Baker, 1983; Bargabus *et al.*, 2004 and Zahoor *et al*., 2012).

Biological control by *Trichoderma* species has been studied by many pathologists for different reasons. Firstly to overcome the passive effects of the used chemical control procedures resulting in the pollution to the environment, and producing of resistant strains (Naseby *et al.*, 2004). Biological control activity by different *Trichoderma* species requires a number of indirect and direct interactions with the pathogen. The combination of these mechanisms resulting in death, suppression or reduction in the pathogen biomass with consequent decrease of plant infection and disease incidence (Vinale *et al*., 2008). Moreover, the biological control by algae became an interesting approach in antibiosis studies. Algal extracts containing plant hormones, amino acids, fatty acids and trace elements that assumed responsible for the improvement of plant growth and for improving the resistance to pathogens, algae extracts stimulated seed germination, growth and yield of different crops (Khan *et al*., 2009). The algal active ingredients may stimulate nitrate reductase and other plant enzymes responsible for absorbing minerals and their transformation in the plant, and thus they act as physioactivators (Joubert and Lefranc, 2008). Although the arbuscular mycorrhiza may be described, as a symbiotic form of diseases, like nodulating nitrogen fixing bacteria. Their use for improvement of plant vigour, thus increasing plant tolerance to diseases has been suggested. Vesicular arbuscular mycorrhizal obligate fungi (VAM) are widespread group of soil inhabitant that can enhance yield of several agricultural crops (Thanuja, 2002). VAM has distinguishing importance due to improving soil structure (Miller and Jastrow, 2000) and great capability to increase plant growth and yield of pepper under different conditions (Durgapal *et al*., 2002). This increment was reported due to various mechanisms such as increased nutrients uptake especially phosphorous content, which has special effect on physiological parameters in plants (Soleimanzadeh, 2010).

Vigour improvement of plant growth either by chemical or biological means was also tried. The objectives of nutrient applications to crops for protection from pathogens are being practiced to avoid plant stress, which may allow crops to better withstanding of the pathogen attack, and to manipulate nutrients to the advantage of the plant and disadvantage of the pathogen (Palti, 1981). Not only is the supply of an individual nutrient important, but also balanced, crop specific nutrient ratios are crucial for improving plant health by adequately supplying the plant during its development under varying environmental conditions. Through an understanding of disease interactions with each specific nutrient, the effects on the plant, pathogen and environment can be effectively modified to improve disease control, enhance production efficiency and increase crop quality (Walters and Bingham, 2007). Mineral nutrients however may reduce the incidence of diseases in certain cases or increase them in others, depending on the particular mineral nutrient, the host plant, the pathogen and other factors.

This investigation was planned to determine the effect of the usual NPK fertilization dose, along with the algae extract, *Trichoderma harzianum* and mycorrhizae (VAM) applications on tomato root-rot incidence.

Materials and Methods

Isolation and identification:

Naturally infected tomato plants showing root-rot symptoms were collected from Kaha Research Station, Qalubiya Governorate, for isolation of the causal organisms. Roots were thoroughly washed with tap water, surface sterilized with sodium hypochlorite 0.5 % for one minute, washed several times with sterilized water and then dried. Infected pieces were then placed on PDA medium and incubated at 28°C for 5 days. The developed fungal colonies were purified using the hyphal tip technique (Brown, 1924).

Purified fungi were placed on PDA slants and kept in a refrigerator for further studies. Identification of the isolated fungi was carried out according to their cultural properties and morphological characteristics as described by Barnett (1965); Rifai (1969) and Baruch *et al.* (1991).

Inoculation procedures:

Rhizoctonia solani propagation was made by inoculating autoclaved sand corn medium (25g washed sand, 75g corn and enough tap water, to cover the prepared mixture in 500ml bottles). The inoculation was made by using agar discs obtained from the periphery of 7 days old colony of the desired fungus and incubated at 28 ± 2^{O}C for two weeks. Each pot (25-cm-diam.) was filled with disinfested soil, and then infested with the desired inoculum at the rate of 3% of the filing soil weight. Inocula were thoroughly mixed with the soil and watered regularly for one week before planting to ensure the uniform distribution and even growth of the tested fungi. Pots used for check were filled with the same soil untreated and distributed under the greenhouse conditions in complete randomized design with 3 replicates for each treatment. *Trichoderma harzianum* was cultured on PDA plates and incubated at 28^{O}C for 4 days, (washed sand 25g, corn 75g and enough tap water to cover the prepared mixture in 500ml bottles) the sterilized bottles were inoculated with agar discs obtained from the periphery of 7 days old colony of *T. harzianum* and incubated at 28 ± 2^{O}C for two weeks.

Host plant and soil:

Tomato seeds (cv. Castle Rock) were used throughout this study. Sandy clay loam soil was used in the greenhouse experiment.

Greenhouse experiment:

Pots (25-cm-diam.) containing sterile soil (3kg/pot) were transplanted with tomato seedlings (5 seedlings/pot). The biocontrol agent inocula were mycorrhiza (AMF) 30gm/kg soil, and Trichoderma at 30gm, fertilizer NPK was applied twice at rate of 6g/pot, and extracted algae preparation was applied twice at the rate of 5 gm/l water.

Microorganisms:

The effective fungal antagonists, mycorrhiza and algae extracts are shown in Table (1). Their high antagonistic effect against wide spectrum of plant pathogens was proved at their respective departments (El-Sayed, 2004; El-Sayed *et al*., 2011; Abouziena *et al*., 2013 and Essa *et al*., 2014).

Table 1. List of treatments used to control tomato root-rot

Treatment	Name	Rate of application	Source
1- Mycorrhiza (AM)	Mycorrhiza	3%/kg soil	Plant Pathol. Dept., National Res. Centre, El-Behouth St., Dokki, 12662 Giza, Egypt
2- Algae extract (A1)	*Chlorella vulgaris*	5gm/l water	
3- Algae extract (A2)	*Spirulina platensis*	5gm/l water	
4- Fertilization	(NPK) 1:1:1	2gm/ kg soil	Veg. Res. Dept., Hort. Res. Inst., Agric. Res. Centre, Giza, Egypt
5- Trichoderma (T)	*T. harzianum*	3% /kg soil	Plant Pathol. Res. Inst., Agric. Res. Centre, Giza, Egypt
6- Pathogen	*R. solani*	3%/kg soil	

Tested treatments:

1- *R. solani* + Mycorrhiza (AM)
2- *R. solani* + *Chlorella vulgaris* (A1)
3- *R. solani* + *Spirulina platensis* (A2)
4- *R. solani* + Fertilization (NPK)
5- *R. solani* + Trichoderma (T)
6- *R. solani* + (AM) + (A1)
7- *R. solani* + (AM) + (A2)
8- *R. solani* + (AM) + (NPK)
9- *R. solani* + (AM) + (T)
10- *R. solani* + (A1) + (NPK)
11- *R. solani* + (A1) + (T)
12- *R. solani* + (A2) + (NPK)
13- *R. solani* + (A2) + (T)
14- *R. solani* + (AM) + (A1) + (A2) + (NPK) + (T)
15- *R. solani* only
16- Check.

Determination of growth parameters:

Growth parameters of tomato plants, *i.e*. shoot height (cm), root length (cm), plant fresh and dry weights (g/plant) and number of leaves/plant were measured 40 days after transplanting.

Disease assessment:

The plants were carefully removed two months after planting and maintained for root-rot severity determination according to the following scale. 0 = healthy roots; 1 = secondary roots are rotten; 2 = secondary root and part of taproot are rotten; 3 = taproot is rotten, 4 = taproot and crown are rotten and 5=death of the plant. Disease severity was calculated according to (Carling and Summer, 1992).

Assay of total phenolic content:

Total phenolic compounds were calorimetrically determined as described by Snell and Snell (1953) using the phosphotungestic-phosphomolybdic acid reagent (Fonlin-Ciocalteu).

Statistical analysis:

Obtained data were analysed by the analysis of variance according to the procedures of Snedecor and Cochran (1980). Means of all treatments were compared by the least significant difference LSD at 5% level.

Results

Isolation and identification:

Isolated fungi from rotted roots of tomato plants were identified and their frequency percentages were calculated. Data presented in Table (2) show that *Rhizoctonia solani* recorded the highest percentage followed by *Alternaria alternata* and *Fusarium solani*, being 37.3; 18.2 and 17.3%, respectively.

Table 2. Frequency (%) of fungi isolated from the rotted root of tomato plants grown in Kaha Research Station

Isolated fungus	Frequency (%)
Rhizoctonia solani Kuhn.	37.3
Alternaria alternata (Fr.) Keissl.	18.2
Aspergillus sp.	9.6
Fusarium solani (Mart.) Sacc.	17.3
Trichoderma harzianum Rafai.	10.7
Other fungi	6.7

Results in Table (3) show that disease severity was significantly increased in tomato plants infected by *R. solani* compared to healthy check. On the other hand, results showed that inoculation with *T. harzianum* along with the pathogen resulted in a lower level of disease severity followed by inoculation with AM fungi, algae extract of *C. vulgaris*, fertilizer (NPK), being 26.6, 28.8, 40.0 and 44.4%, respectively.

Table3. Effect of different treatments on tomato root-rot severity under greenhouse conditions

Treatment *	Diseases severity (%)	Reduction (%) to infested check
R. solani + (AM)	28.8	52.0
R. solani + (A1)	40.0	33.3
R. solani + (A2)	51.1	14.8
R. solani + (NPK)	44.4	26.0
*R. solan*i + (T)	26.6	55.6
*R. solan*i + (AM) + (A1)	37.7	37.1
*R. solan*i + (AM) + (A2)	35.5	40.8
*R. solan*i + (AM) + (NPK)	40.0	33.3
R. solani + (AM) + (T)	24.4	59.3
R. solani + (A1) + (NPK)	44.4	26.0
R. solani + (A1) + (T)	35.5	40.8
R. solani + (A2) + (NPK)	44.4	26.0
R. solani + (A2) + (T)	35.5	40.8
R. solani + (AM) + (A1) + (A2) + (NPK) + T	22.2	63.0
Check (*R. solani* infested soil)	60.0	0.0
Check (uninfested soil)	46.6	22.3
LSD at 5%	1.21	----

* Mycorrhiza = (AM); Algae = *Chlorella vulgaris* (A1) ; Algae = *Spirulina platensis* (A2); Fertilization = (NPK); Trichoderma= (T); Check= uninfested soil; Check= infested soil with *R. solani*.

Data presented in Table (4) show the effect of various treatments on certain growth parameters, *i.e.* stem height, root length, dry and fresh weights of the plants compared to the check treatments. The observed significant growth due to *T. harzianum*, mycorrhiza, algae extract, and fertilizer (NPK), are quite clear especially for the combined treatment with [mycorrhiza (AM), *C. vulgaris* (A1), *S. platensis* (A2), NPK and *T. harzianum* (T)].

Table 4. Effect of the tested treatments on the parameters of tomato plants (cv. Castle Rock) grown under greenhouse conditions

Treatment *	Plant fresh weight (g)	Plant dry weight (g)	Shoot height (cm)	Root length (cm)	No. of leaves
R. solani + (AM)	16.8	2.7	27.5	4.8	4.6
R. solani + (A1)	13.5	1.8	22.8	3.4	5.9
R. solani + (A2)	14.5	1.6	21.5	3.3	4.3
R. solani + (NPK)	15.0	1.5	24.0	3.6	3.8
*R. solan*i + (T)	17.8	2.5	33.0	4.2	5.7
*R. solan*i + (AM) + (A1)	18.7	2.2	28.0	5.2	5.4
*R. solan*i + (AM) + (A2)	19.6	2.2	30.6	5.1	5.3
*R. solan*i + (AM) + (NPK)	21.5	2.0	34.5	5.6	5.0
R. solani + (AM) + (T)	23.5	3.2	36.5	5.5	5.8
R. solani + (A1) + (NPK)	20.3	1.7	26.0	4.5	4.5
R. solani + (A1) + (T)	22.5	2.2	29.5	5.2	5.2
R. solani + (A2) + (NPK)	20.0	1.8	25.0	5.0	4.0
R. solani + (A2) + (T)	22.0	2.8	28.5	5.3	4.2
R. solani + (AM)+(A1)+(A2)+(NPK)+T	26.5	3.5	38.0	8.7	6.7
Check (*R. solani* infested soil)	8.5	1.0	13.8	2.8	2.1
Check (uninfested soil)	11.5	1.5	20.5	3.0	3.1
LSD at 5%	1.25	0.5	2.8	0.4	0.4

* As described in footnote of Table (3).

Results presented in Table (5) indicate that the formation of soluble phenols was significantly increased in the infected plants subject to various treatments, compared to healthy checks. The effect of mixed treatments with fungi antagonistic to *R. solani* on phenol contents of the infected tomato plants was considered in the present study. Mycorrhiza treatments significantly increased the total phenol content in the infected tomato plants (0.83) mg/g fresh weight compared to the infected plants without mycorrhiza treatment (0.34) mg/g fresh weight, and the healthy check plants (0.46) mg/g fresh weight. The most promising treatments for total phenol increase may be recognized for infected plants treated by a mixture of mycorrhiza, *Chlorella vulgaris* (A1) and *Spirulina platensis* (A2), Trichoderma plus fertilization with the usual NPK, being (1.59) mg/g fresh weight compared to (0.34) mg/g fresh weight for plants infected with Rhizoctonia alone. Similar results are recognized in case of infected plants treated with mixed *Trichoderma* and mycorrhiza treatments, and treatments with either Trichoderma or mycorrhiza alone.

Table 5. Effect of treatments on phenol contents of tomato plants cv. Castle Rock grown in soil artificially infested tested with *R. solani*, 40 days after transplanting, under greenhouse conditions

Treatment *	Phenol components as mg/g fresh weight in plant		
	Free	Conjugated	Total
R. solani + (AM)	0.230	0.600	0.830
R. solani + (A1)	0.128	0.390	0.518
R. solani + (A2)	0.210	0.350	0.560
R. solani + (NPK)	0.176	0.370	0.546
*R. solan*i + (T)	0.134	0.790	0.924
*R. solan*i + (AM) + (A1)	0.177	0.430	0.607
*R. solan*i + (AM) + (A2)	0.220	0.400	0.620
*R. solan*i + (AM) + (NPK)	0.103	1.180	1.283
R. solani + (AM) + (T)	0.330	1.050	1.380
R. solani + (A1) + (NPK)	0.260	0.590	0.850
R. solani + (A1) + (T)	0.280	0.660	0.940
R. solani + (A2) + (NPK)	0.233	0.330	0.563
R. solani + (A2) + (T)	0.027	0.860	0.887
R. solani + (AM) + (A1) + (A2) + (NPK) + T	0.110	1.480	1.590
Check (*R. solani* infested soil)	0.090	0.250	0.340
Check (uninfested soil)	0.155	0.310	0.465

* As described in footnote of Table (3).

Discussion

The present investigation highlights the possibility of controlling root-rot of tomato by certain antagonistic microorganisms such as *Trichoderma harzianum* and mycorrhizal fungi alone with algae extract [*Chlorella vulgaris* (A1), *Spirulina platensis* (A2)] and fertilizer (NPK). The recognized increased growth of the crops due to mycorrhiza infection may be attributed to improve phosphorous and micronutrient uptake by the host plant. These treatments also caused an increase in dry weight of plants raised under controlled conditions in the greenhouse as a consequence to the greater uptake of macronutrients and possibly micronutrients compared to the check treatments. The effect of micronutrients in increasing dry weight is of course of paramount importance and requires deep investigation (Jeffries *et al*., 2003). Moreover, the combined treatment with both *T. harzianum* and AMF also significantly promoted growth as expressed by an increase in stem length, shoot and root dry weights. The multiple effect of dual inoculation with *T. harzianum* and AMF has previously been reported to enhance significantly the growth more than the single fungus application (Ozbay and Newman, 2004). The treatment with a combination of *T. harzianum*, AMF, algae *C. vulgaris* (A1), *S. platensis* (A2) and NPK also significantly decreased the disease severity compared to the individual treatments. There are many available literatures on the antibiosis of the of *T. harzianum*, and the check plant pathogens concentrating on

several mechanisms including mycoparasitism and induction of systematic defence mechanism (Yedida *et al*., 1999 and Harman, 2000) that being resulted in a range of biopesticides is now available commercially, most of which are based on the fungal genus *Trichoderma* (Woo *et al*., 2006). Some strains of Trichoderma have been reported to elicit induced systemic resistance and, moreover, colonized roots appear to be primed for an intense defence response to subsequent pathogen attack (Reglinski *et al*., 2012). On the other hand AMF has been known to increase plant resistance to infection through improved plant nutrition (Declerck *et al*., 2002). Classically, four major groups of mycorrhizal mode of action mechanisms that mediated bio-protection have been considered: (1) direct competition, (2) mechanism mediated by alteration in plant growth, nutrition and morphology, (3) biochemical and molecular changes in mycorrhizal plants that induce pathogen resistance and (4) alterations in the soil microbiota and development of pathogen antagonism (Vierheilig *et al*., 2008).

In the present study, however, there is an evidence on the increased disease control of *R. solani* with mixed treatment using algal extract (*Chlorella vulgaris* (A1) and *Spirulina platensis* (A2) as shown by the decreased disease severity, the combination of algal extracts with either *T. harzianum* or mycorrhiza had the best effect on the disease severity compared to check treatments. Mixed *T. harzianum* with *C. vulgaris* (A1), *T. harzianum* with *S. platensis* (A2) and mycorrhiza with *C. vulgaris* (A1), mycorrhiza, *S. platensis* (A2) but no effect was found when combination was made with NPK treatment.

In this regard macro- and micro- elements have long been recognized as being associated with size, quality, and yield of crops, and also with changes in levels of the incidence of the disease (Rush *et al*., 1997). Microbial activity in the rhizosphere is a major factor that determines the availability of nutrients to plants and has a significant influence on plant health and productivity (Jeffries *et al*., 2003).

The effect of mixed treatments with fungi antagonistic to *R. solani* on phenol contents of affected tomato plants was considered in the present study. The mycorrhiza treatments significantly increased the total phenol content in the infected tomato plants (0.83) mg/g compared to the infected plants without mycorrhiza treatment (0.34) mg/g, and the healthy check plants (0.46) mg/g. The most promising treatments for total phenol increase may be recognized for infected plants treated by a mixture of mycorrhiza, *C. vulgaris* (A1) and *S. platensis* (A2), Trichoderma plus fertilization with the usual NPK, being (1.59) mg/g compared to (0.34) mg/g for plants infected with *R. solani* alone. Similar results may be recognized in the case of infected plants treated with mixed Trichoderma and mycorrhiza treatments, and treatments with either Trichoderma or mycorrhiza each alone.

Generally, the role of phenolic compounds in defence is related to their antimicrobial and nutritional or unpalatable properties**.** Plant phenolics may be divided in two classes: (i) preformed phenolics that are synthesized during the normal development of plant tissues and (ii) induced phenolics that are synthesized by plants in response to physical injury, infection or when stressed by suitable elicitors such as heavy metal-salts, UV-irradiation, temperature, *etc.* (phytoalexins).

The mechanism is due to the direct effect of the antagonist in addition to stimulation of plant resistance by producing active phenols (Cherif *et al*., 2007).

References

Abouziena, H.F.; Zaghloul, A.; El-Ashry, S.; Hobballa, E.M. and Saber, M. 2013. Some chemical and biological additives with hyper accumulator plants for amendment the sandy soil contaminated for long term by sewage water. *Internat. J. Water Res. Arid Environ*., **3**(1): 15-25.

Bargabus, R.L.; Zidack, N.K.; Sherwood, J.E. and Jacobsen, B.J. 2004. Screening for the identification of potential biological control agents that induce systemic acquired resistance in sugar beet. *Biological Control*, **30**: 342-350.

Barnett, H.L. 1965. *Illustrated Genera of Imperfect Fungi*. Burgess Pub. Co. 218 pp.

Baruch, S.; Burpee, l. and Ogoshi, A. 1991. *Identification of Rhizoctonia Species*. APS. Press. 133pp.

Bolkan, H.A. and Ribeiro, W.R.C. 1985. Anastomosis groups and pathogenicity of *Rhizoctonia solani* isolates from Brazil. *Plant Dis*., **69**: 599-601.

Brown, N. 1924. Two mycological methods. II. A method of isolating single strains of fungi by cutting out a hyphal tip. *Ann. Bot*., **38**: 402-406.

Campbell, R. and Shishkoff, N. 1990. Survival of *Pyrenochaeta lycopersici* and the influence of temperature in cultivar resistance on the development of corky root of tomato. *Plant Dis*., **74**(11): 889-894.

Carling D.E. and Summer, D.R. 1992. Rhizoctonia. Pages: 157-165. In: *Methods for Research on Silborne Phytopathogenic Fungi.* L.L. Singleton, J.D. Mihail and C.M. Rush (eds.). The Amer. Phytopathol. Soc. Press, St Paul, Minnesota, USA.

Cherif, M.; Arafaoui, A. and Rhaiem, A. 2007. Phenolic compounds and their role in bio-control and resistance of chickpea to fungal pathogenic attacks. *Tunisian J. Plant Prot*., **2**: 7-21.

Cook, R. and Baker, K.F. 1983. *The Nature and Practice of Biological Control of Plant Pathogens*, Amer. Phytopathol. Soc., St Paul, Minnesota, USA. 539pp.

Declerck, S.; Risede, J.M.; Rufyikiri, G. and Delvaux, B. 2002. Effect of arbuscular mycorrhizal fungi on severity of root rot of bananas caused by *Cylindrocladium spathiphylli*. *Plant Pathol*., **51**: 109-115.

Durgapal, A.; Pandey, A. and Palni L.S. 2002. The use of rhizosphere soil for improved establishment of conifers at nursery stage for application in plantation programmers. *J. Sustain. Forest*, **15**(3): 57-73.

El-Sayed, A.B. 2004. Screening and growth characterization of the green life stock of drill water from Jeddah, Saudi Arabia. I- Isolation and growth characterization of *Scenedesmus* sp. *Egypt. J. Microbiol*., **8**: 376-385.

El-Sayed, A.B.; Abdel-Maguid, A.A. and Hoballah, E.M. 2011. Growth response of *Chlorella vulgaris* to acetate carbon and nitrogen forms. *Nature Sci.,* **9**(9): 53-58.

Essa, T.A.A.; Muhanna, Naglaa A.S. and Mansour, A.S. 2014. Longevity of *Trichoderma harzianum* and *T. viride* in alginate preparations in relation to the stock propagation media. Egypt. *J. Biol. Pest Control*, **24**(2): 471-476.

Harman, G.E. 2000. Myths and dogmas of biocontrol. Changes in perceptions derived from research on *Trichoderma harzianum* T-22. *Plant Dis*., **84**: 377-393.

Jeffries, P.; Gianinazzi, S.; Perotto, S.; Turnau, K. and Barea, J.M. 2003. The contribution of arbuscular mycorrhizal fungi in sustainable maintenance of plant health and soil fertility. *Biol. Fert. Soils,* **37**: 1-16.

Joubert, J.M. and Lefranc, G. 2008. Seaweed phytostimulants in agriculture: recent studies on mode of action two types of products from algae: growth and nutrition stimulants and stimulants of plant defence reactions. Book of abstracts: Biostimulators in modern agriculture. Warsaw, 7-8 February, 2008.

Khan, W.; Rayirath, U.P.; Subramanian, S.; Jithesh, M.N.; Rayorath, P.; Hodges, D.M.; Critchley, A.T.; Craigie, J.S.; Norrie, J. and Prithiviraj, B. 2009. Seaweed extracts as biostimulants of plant growth and development. *J. Plant Growth Reg*., **28**: 386-399.

Miller, R.M. and Jastrow, J.D. 2000. Mycorrhizal fungi influence soil structure. Pages: 3-18. In: *Arbuscular Mycorrhizas: Physiology and Function.* Y. Kapulnik and D.D. Douds (eds.). Kluwer Academic, Dordrecht.

Naseby, D.C.; Pascual, J.A. and Lynch, J.M. 2004. Effect of biocontrol strains of Trichoderma on plant growth, Pythium ultimum populations, soil microbial communities and soil enzyme activities. *J. Appl. Microbiol*., **88**: 161-169.

Ogoshi, A. 1987. Ecology and pathogenicity of anastomosis and intraspecific groups of *Rhizoctonia solani* Kühn. *Ann. Rev. Phytopathol*., **25**: 125-143.

Ozbay, N. and Newman, E.S. 2004. Effect of *T. harzianum* strains to colonize tomato roots and improve transplant growth. *Pak. J. Biol. Sci*., **7**: 253-257.

Palti, J. 1981. *Cultural Practices and Plant Diseases*. Springer-Verlag, Berlin.

Reglinski, T.; Rodenburg, N.; Taylor, J.T.; Northcott, G.L.; Chee, A.A.; Spiers, T.M. and Hill, R.A. 2012. *Trichoderma atroviride* promotes growth and enhances systemic resistance to *Diplodia pinea* in radiata pine (*Pinus radiata*) seedlings. *Forest Pathol*., **42**: 75-78.

Renata, G.B.; Deborah, H.M.B.; José, M.M.; Fernanda, S.C.; Da SilvaTorres, Elizabeth, A.F. 2013. Antioxidant potential of tomatoes cultivated in organic and conventional systems. *Braz. Arch. Biol. Technol*., **56**(4): 521-529.

Rifai, M.A.A. 1969. Revision of the genus *Trichoderma*. *Mycol. Papers*. **116**: 1–56.

Rush, C.M.; Piccinni, G. and Harveson, R.M. 1997. Agronomic measures. In: *Environmentally Safe Approaches to Crop Disease Control*. N.A. Rechcigel and J.E. Rechcigel (eds.). CRC Pub., Boca Raton, FL, USA.

Snedecor, G.W. and Cochran, W.G. 1980. *Statistical Methods*. 7th Ed. Oxford and J.P.J. Pub. Comp.

Snell, F.D. and Snell, C.T. 1953. Colorimetric methods analysis including some turbidimetric and nephelometric methods. D. Van., Nostrand Company IVC., Toronto, New York, London, **111**: 606-612.

Soleimanzadeh, H. 2010. Effect of VA-mycorrhiza on growth and yield of sunflower (*Helianthus annuus* L.) at different phosphorus levels. *World Acad. Sci. Eng. Technol*., **71**: 414-417.

Stiling, P. and Cornelissen, T. 2005. What makes a successful biocontrol agent? A meta-analysis of biological control agent performance. *Biol. Control*, **34**: 236-246.

Thanuja, T.V. 2002. Induction of rooting and root growth in black pepper cuttings (*Piper nigrum* L.) with the inoculation of arbuscular mycorrhizae. *J. Sci. Hort*., **92**(3-4): 339-346.

Vierheilig, H.; Steinkellner, S.; Khaosaad T. and Garcia-Garrido, J.M. 2008. The biocontrol effect of mycorrhization on soilborne fungal pathogens and the autoregulation of the am symbiosis: one mechanism, two effects?. Pages: 307-320. In: Mycorrhiza: Genetics and Molecular Biology, Eco-Function, Biotechnology, Eco-Physiology, Structure and Systematics. Varma, A. (ed.). Springer-Verlag, Heidelberg, Germany.

Vinale, F.; Sivasithamparam, K.; Ghisalberti, E.L.; Marra, R.; Woo, S.L. and Lorito, M. 2008. *Trichoderma*-plant-pathogen interactions. *Soil Biol. Biochem*., **40**: 1-10.

Walters, D.R. and Bingham, I.J. 2007. Influence of nutrition on disease development caused by fungal pathogens: implications for plant disease control. *Ann. Appl. Biol*., **151**: 307-324.

Woo, S.L.; Scala, F.; Ruocco, M. and Loritto, M. 2006. The molecular biology of the interactions between *Trichoderma* spp., phytopathogenic fungi, and plants. *Phytopathology*, **96**: 1061-1070.

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

Zahoor, A.; Saifulla, F.R.; Hakim, K. and Muhammad, I. 2012. Chemical and biological control of root rot of okra. *Pak. J. Bot*., **44**: 453-457.

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المقاومه البيولوجية لمرض جذور الطماطم المتسبب عن *Rhizoctonia solani*

نجلاء عبد الباقى سلام مهنا ، سهام سمير محمد رجب ، عبير عبد الرحمن الغانم
معهد بحوث أمراض النباتات - مركز البحوث الزراعية - الجيزة.

إستهدفت الدراسة إستخدام ٤ مواد لمقاومة مرض جذور الطماطم المتسبب عن الفطر *Rhizoctonia solani* ، وذلك بدراسه تأثير هذه المواد كل منهم على حده وتأثير خلطهم معاً على شده الإصابة ومستويات الفينول فى الأنسجة النباتية. وكانت نتيجه إستخدام التريكودرما بمفردها ادى الى إنخفاض شدة الإصابة ، وعند الخلط بالميكروهيزا والطحالب والسماد المعدنى أعطت نتيجة أقل وذلك مقارنة بالكنترول. كما أدت المعامله بالميكروهيزا إلى زياده معنوية فى مستويات الفينول الكلية فى النباتات المصابة بالفطر *Rhizoctonia solani* مقارنه بالنباتات السليمة. كما أظهرت معامله النباتات بنوعى الطحالب تحت الدراسة و الميكروهيزا والسماد المعدنى والتريكودرما أعلى نسبه لمستويات للفينول الكليه مقارنه بالنبات المصاب بالفطر *Rhizoctonia solani* فقط. كما أدت معاملة التريكودرما والميكروهيزا كذلك إلى زيادة فى نسبه الفينولات الكلية.