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Efficacy of some *Trichoderma* Species on Management of Sunflower Head-Rot

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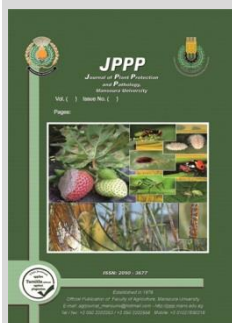


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ABSTRACT

The efficacy of different *Trichoderma* species were evaluated *in vitro* and under field conditions against five pathogenic of sunflower head-rot fungi (HRF). Bioagents were applied by three application methods during 2015 and 2016 growing seasons. *In vitro*, six species of *Trichoderma harzianum*, *T. viride*, *T. hamatum*, *T. koningii*, *T. reesei* and *T. polysporum* were selected for field studies. Both of *T. harzianum* and *T. viride* recorded the highest antagonistic potential against almost of the pathogenic fungi followed by *T. reesei*. In field experiments, the tested bioagents were studied in series of experiments using different three application methods, i.e. foliar spray [F.S.], soil drench [S.D.] and foliar spray with soil drench [F.S.+ S.D.]. These bioagents were applied at three stages i.e. 30, 45 and 60 days after sowing. Head diameter, stem diameter, leaf area, seed number per head, 1000 seed weight, seed yield per plant, yield per plot and yield per feddan were determined as crop parameters. Results indicated that the three application methods gave the lowest values of disease incidence and increased crop parameters compared with untreated plants. The highest antagonistic activity was achieved with *T. harzianum* when applied as foliar spray which gave the highest percentage of disease reduction and increased crop parameters during both seasons, followed by strain of *T. viride* when applied as soil drench and *T. reesei* when applied as (soil drench and foliar spray), respectively.

Keywords: Head-rot sunflower- *Trichoderma* spp. - Biological control- Disease management- Crop parameters - Induce systemic resistance.



INTRODUCTION

Sunflower (*Helianthus annuus L.*) is the third most important oil seed crop belonging to Asteraceae and is a rich source of edible oil (40-52%) having anticholesterol properties due to the presence of polyunsaturated fatty acids (55-65% linoleic acid and 20-30% oleic acid) (Joksimovic *et al.*, 2006). Area under sunflower cultivation in the world accounts for (20) million hectares. Presently in Egypt, sunflower is cultivated over an area of 38550 feddan with a productivity of 970kg - 1200kg / feddan (Mini. of Agric. and Land Recl., 2016). More than 30 diseases have been identified on sunflower (Gulya *et al.*, 1994), head rot disease as effective by destructive pathogenic fungi which are often called the head rot fungi [HRF] on sunflower and among them i.e. *Fusarium Head-rot* [FHR], *Botrytis Head rot* [BHR] and *Rhizopus Head rot* [RHR] (Rodriguez *et al.*, 2004), were a worldwide distributed necrotrophic pathogen, attacking more than 400 plant species including sunflower (Boland and Hall 1994). The pathogenic of head rot fungus can attack sunflower plants causing diverse symptoms in leaves, stalks and flowers and causing significant losses in seed yield, damage to oil and has a wide host range causing average yield reductions of 10 to 20%. On the other hand, head rot are considered a major disease in Europe, Argentina, USA and Egypt causing average yield reduction reached to 20% (Al-Taweil *et al.*, 2009), also when it occurs it can cause significant damage to oilseed crops, where can be cultivated for many years, caused an increase in the incidence and severity in production areas (Steadman, 1983).

Disease control of head rot infection is difficult, since the pathogens persists in the soil for long periods and at high inoculum levels (Rodriguez *et al.*, 2004). The diseases can be controlled by appropriate application of fungicides, while the most economic method can be controlled these diseases by use biological control, where increased the effectiveness of the disease management.

Trichoderma spp. are considered as most efficient biocontrol agents (Perchepeid *et al.*, 2010). Major mechanisms involved in the biocontrol activity of *Trichoderma* spp. are competition for space and nutrients, production of diffusible and/or volatile antibiotics, hydrolytic enzymes like chitinase and B-1,3-glucanase. These hydrolytic enzymes partially degrade the pathogen cell wall and lead to its parasitization (Lorito *et al.*, 2010). Also, many recent findings suggest that plant development and biochemistry were strongly affected by *Trichoderma* strains. *Trichoderma* colonize were gave penetrate plant head tissues and initiate a series of morphological and biochemical changes in the plant, considered to be part of the plant defense response, which in the end leads to induced systemic resistance in the entire plant. *Trichoderma* species have the ability to interact with head of diverse plant species leading to induce systemic resistance responses to a wide spectrum of pathogens and adverse environmental conditions (Shoresh, *et al.*, 2010). Other ecological success of these strains are their capability to synthesize antagonistic compounds [proteins, enzymes and antibiotics] and growth promoting substances [vitamins, hormones and minerals] enhance their biocontrol activity (Al-Taweil *et al.*, 2009).

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There are several studies dealing with the use of biological control agents, mainly *Trichoderma* spp. to control head-rot diseases, caused by the genera *Botrytis*, *Rhizopus*, *Rhizoctonia*, *Aspergillus* and *Fusarium* (Elad and Stewart 2007). There are some studies conducted with *Trichoderma* bioagents for the control of head-rot fungi [HRF] of sunflower and promising results have been obtained by many researchers. (Raouf *et al.*, 2003, Chagas, 2009, and Demant *et al.*, 2010). In addition, (Raj *et al.*, 2004) reported that *Trichoderma* species, i.e. *T. harzianum*, *T. viride*, *T. hamatum* and *T. koningii* used as spray and soil drench, induced head rot diseases resistance and enhanced promotes plant growth and fruit yield (Sudisha *et al.*, 2006).

The objective of this study was to evaluate the effectiveness of *Trichoderma* spp. isolates with different three application methods to reduce sunflower head-rot, as well as the economic importance of *Trichoderma* spp. in increased yield quantity under field conditions at two seasons (2015, 2016)

MATERIALS AND METHODS

1-Antagonistic effect of *Trichoderma* isolates on linear growth of sunflower head-rot fungi:

All *Trichoderma* spp. were kindly obtained from the Dept. of Mycol. Res., Pl. Pathol. Res. Inst., ARC, Giza, Egypt., were subjected under lab. conditions to evaluate their antagonistic effect on the majority of phytopathogenic fungi causing head rot of sunflower i.e. *B. cinerea*, *R. solani*, *R. oryzae*, *A. flavus* and *F. moniliforme* were collected from (onion and garlic) and oil crops diseases Res. Dept. Institute (ARC), Giza, Egypt. In such experiment, plates were inoculated with 7 days old pure culture discs (5 mm in diameter) of the pathogenic fungal isolates at the preferal of the plate surface. The antagonistic organism (5 mm diam. disc) was inoculated on the opposite side of the plate, then incubated at 27°C and periodically examined every 24 hrs. Four replicates were used for each particular treatment. After the complete growth of the control treatment, reduction in mycelial growth was estimated according to the method adopted by Ferreira *et al.*, (1991) in which:

$$R = \frac{A-B}{A} \times 100.$$

Where: R= Percentage of growth reduction.

A= Average growth diameter of control.

B= Average growth diameter of pathogen.

Degree of antagonistic effect was determined according to the scale adopted by Bell *et al.*, (1982).

2-Field Experiments:-

-Preparation of bioagents inoculum:-

Trichoderma isolates were grown on broth medium (Allens *et al.*, 1983) by inoculation of 5 ml of conidial suspension from sporulating cultures on PDA slants to 250ml of medium in 500 ml Erlenmeyer flasks, the potato dextrose broth was employed for multiplication of *Trichoderma* isolates. The *Trichoderma* suspension was adjusted to be contain 5×10^{10} cfu/ml by using a mechanical shaker, at 180 rpm at 28°C and centrifuged at 18,000 rpm for 15 min in sharples on line centrifuge. These conidial suspensions were used for further experiments.

- Disease assessment:

Suspension concentrates of bioagents 5×10^{10} conidia/ml was used for different application methods as i.e. foliar spray (F.S.), soil drench (S.D.) and both application methods (F.S.+ S.D.) to determine the most effective

application method against head Rot fungi (HRF) of sunflower. On the other hand, the broth liquid of *Trichoderma* isolates namely, *T. harzianum*, *T. viride*, *T. Hamatum*, *T. Koningii*, *T. Reesei* and *T. Polysporum* were used in doses at (5ml/L) at three stages, the first dose was given 30 days after sowing followed by second dose after 45 days followed by the third dose after 60 days after sowing. Disease assessment was measured as percentages of standing plant with head rot symptoms 90 days after planting, and disease incidence was determined according to

$$\text{Percentage of infection} = \frac{\text{Number of infected plants}}{\text{Total number of plants}} \times 100$$

The experiment was repeated through two seasons to determine the accurate application on head rot sunflower.

- yield components and growth characters:

A field experiments were conducted during seasons 2015 and 2016 at Etai EL-Baroud Experi Resr stat, the location chosen was fields naturally infested with head rot diseases that had been planted of sunflower. Each experiment was carried out in split plot design and the sunflower (c.v.sakha 53) were sown in the experiments under field condition, resulted and area 574 m² were divided to four plots, Each plot consisted of 7 ridges, 50 within each ridge included treatments i.e. *T. harzianum*, *T. virid.*, *T. hamatum*, *T. koningii*, *T. reesei*, *T. polysporum* and the untreated control plants, each ridges of treatments divided to three rows, the row was 3m in length and 50cm width, seeds were sown on the third week of May at 20cm spacing within each row and, three replications were maintained for each treatment. *Trichoderma* bioagents were applied at 30,45,60 days after sowing. At the end of experiment, the following data were recorded during two seasons 2015/2016 i.e. head diameter [cm], stem diameter [cm], leaf area [cm²], seed number per head, 1000 seed weight [g], seed yield per plant [g], yield per plot [kg], total yield per feddan [ton]. Leaf area per plant in square decimeters of three samples were measured according to the method described by (Shaneiter, 1978) using the formula

$$(La = (l \times w \times 0.6683) - 2.45)$$

La: leaf area L : length W: wide

-Statistical analysis:

Data obtained from all experiments were subjected to analysis of variance (ANOVA), the studied treatments means according to Gomez and Gomez (1984). The treatment mean was compared using the least significant differences (L.S.D.) test at 5% level.

RESULTS AND DISCUSSION

This investigation aimed to study the effect of six bioagents with three application methods on the incidence of sunflower head rot disease. The results were evaluated *in vitro* and determined during 2015, 2016 growing seasons under field conditions.

1-Effect of different isolates of *Trichoderma* spp. against the growth of pathogenic fungi:

Data presented in Table (1 and 2) revealed the effect of six isolates of *Trichoderma* spp. on sunflower head rot pathogens in terms of growth diam., for the pathogen and reduction in colony diameter (%) due to the application. These data indicate that a significant difference was observed between each of the tested isolates and the control treatment. This might be due to the effective agents against wide range of sunflower pathogenic fungi (Hegde 2002). The highest reduction (%) was recorded with *T. harzianum* followed by

T. Viride and *T.reesei*. On the other hand, the least reduction in radial growth of test sunflower head rot fungi was recorded with *T.hamatum* followed by *T. polysporum*. This effect might be due to *Trichoderma* spp. isolates proved to exhibit high growth level of indurance to the causal agents of head rot fungi of sunflower (Chandra *et al.* , 2009). Although, *T. harzianum* and *T. Viride* given a large number of antagonism and the isolates are capable of producing antibiotics in laboratory cultures. If these, or similar compounds are involved at least in part or in the biocontrol activity of the organism. (Adams *et al.*, 2007).

Table 1. Effect of *Trichoderma* spp. on linear growth of *B.cinearea* and *R.solani* causal organisms of sunflower head rot disease.

Bioagent	<i>B. cinearea</i>		<i>R. solani</i>	
	G.D (mm)	R%	G.D (mm)	R%
<i>T. harzianum</i>	40.7	54.7	30.9	65.6
<i>T.viride</i>	40.1	55.3	40.3	55.2
<i>T. hamatum</i>	50.8	43.5	60.0	33.3
<i>T. koningii</i>	50.3	44.0	50.7	43.6
<i>T. reesei</i>	40.9	54.4	40.1	55.4
<i>T. polysporum</i>	50.7	43.6	50.7	43.6
Control	90.0	00.0	90.0	00.0
Mean	51.9	42.2	51.8	42.38
L.S.D at 5%	0.83		0.50	

[G.D]: Growth Diameter [R]: % Reduction

Table 2. Effect of *Trichoderma* spp on linear growth of *R.oryzae*, *A. flavus* and *F.moniliformum* causal organisms of sunflower head rot disease.

Bioagent	<i>R.oryzae</i>		<i>A.flavus</i>		<i>F.moniliformum</i>	
	G.D(mm)	R%	G.D(mm)	R%	G.D(mm)	R%
<i>T. harzianum</i>	30.6	65.9	20.3	77.3	20.4	77.2
<i>T.viride</i>	40.2	55.3	30.4	66.2	20.4	77.2
<i>T. koningii</i>	40.5	54.9	30.3	66.2	30.1	66.4
<i>T.reesei</i>	40.2	55.3	20.1	77.5	20.7	76.9
<i>T. polysporum</i>	40.9	54.4	30.1	66.5	30.2	66.4
Control	90.0	0.00	90.0	0.00	90.0	00.0
Mean	46.1	48.6	35.9	59.9	34.6	61.4
L.S.D at 5%	0.33		0.50		0.44	

[G.D]: Growth Diameter [R]: % Reduction

2. Field experiments:

Disease incidence:

Three application methods. i.e. foliar spray [F.S.], foliar spray with soil drench[F.S.+S.D.] and soil drench [S.D.] of sunflower plants were applied in dose at three stages at 30,45,60 days after sowing were carried under field conditions during the two seasons 2015 and 2016. Data in Table (3) show that sunflower head rot incidence in treatments with *Trichoderma* isolates led to significant increase in disease reduction of head rot when used as foliar spray of sunflower plants in comparison with the other two application methods. This data might be due to application with foliar spray which induces systemic resistance as the main mechanism of the activity to the plant (Azarmi *et al.*, 2011 and Al- Ani *et al.*, 2011). On the other hand, among all the different application methods as foliar spray, foliar with soil drench and soil drench by *Trichoderma* isolates as solution spraying were found to be the most effective against sunflower head rot fungi which recorded the lowest disease incidence compared with untreated control. This effect might be due to *Trichoderma* bioagents which produce different types of antibiotics (Adames *et al.*, 2007).

Table 3. Effect of three application methods, with different bioagents on disease incidence (%) of head rot sunflower disease, during growing seasons 2015 and 2016.

Bioagent	2015 season			2016 season		
	Foliar spray	Foliar spray+ Soil drench	Soil drench	Foliar spray	Foliar spray+ Soil drench	Soil drench
<i>T.harzianum</i>	1.6a	6.6a	6.6b	0.0a	5.0a	3.3b
<i>T.viride</i>	5.0b	8.3b	3.3a	3.3a	6.6b	1.6a
<i>T.hamatum</i>	13.3e	15.0e	13.3f	10.0a	11.6d	10.0e
<i>T.koningii</i>	10.0d	11.6d	11.6e	8.3a	13.3e	11.6f
<i>T.reesei</i>	8.3c	6.6a	10.0d	6.6a	6.6b	8.3d
<i>T.polysporum</i>	10.0d	10.0c	8.3c	8.3a	10.0c	6.6c
Control	45.0f	45.0f	45.0g	43.3b	43.3f	43.3g
Mean	13.3	14.7	14.0	11.3	13.7	12.1
L.S.D. at 5%	0.56	0.55	0.54	0.35	0.18	0.14

The efficacy of *Trichoderma* bioagents against sunflower head rot fungi was tested during two growing seasons field trials, *T. harzianum* and *T. viride* were as equal effect against head rot with maximum disease reduction over other *Trichoderma* species isolates. The tested *Trichoderma* bioagents showed different little disease incidence ranged from 1.67 – 15.0% comparing with the untreated plants 43.3-45%, during 2015 /2016 growing season. The lowest values of disease incidence was recorded with *T. harezianum* when applied as foliar spray. Results in the same Table indicate that, all the individual bioagent treatment were effective with high disease reduction compared with the untreated control. This might be due to *Trichoderma* bioagents which have several methods to control the disease such as production of antifungal compounds and induce of systemic resistance in the intire plant (Chandra *et al.*, 2009).

Effect of *Trichoderma* species on the yield quantity of sunflower under field conditions:

Data presented in (Table 4) reveale that all the different application methods with *Trichoderma* bioagents significantly increased head diameter over control in the two seasons. Both of *T.viride* and *T.harzianum* at foliar spray gave the highest number of head diameter, which recorded 31 and 30 cm, respectively in the first season. Mean while the treatments of *T.reesei* as foliar spray and soil drench recorded 29.3cm. On the other hand, the lowest head diameter number resulted from the treatments of *T.koningii* as foliar spray which gave 22.6 in second season viz. 16 and 19.3 cm for untreated control during the seasons 2015and2016, respectively. This increase in the number of head diameter might be due to either three different application methods or *Trichoderma* bioagents isolates which showed important ecological increase the growth of head sunflower diameter (Begum *et al.*, 2010).

As shown in (Table 4) the data reveale that the different three application methods with all *Trichoderma* treatments significantly increased the stem diameter over control in both seasons. The highest number of stem diameter resulted from the treatment with *T.harzianum* and *T.viride* as foliar spray, which gave 4.51 and 4.11 cm., respectively in the first season. While the same treatments showed 4.45 and 4.28 cm at the same application method in the second season viz. 2.60 and 2.71 cm for untreated control plants in both seasons. This increase of stem diameter might be due to that

Trichoderma bioagents production hydrolytic enzymes like chitinase and B- 1.3. glucanase. These hydrolytic enzymes led to an increase in growth stem of sunflower plants (Begum *et al.*, 2010).

It is obvious from data in (Table 4) also that all treatments at the three application methods gave the wide leaves than control in both seasons. The widest leave area resulted from plants sprayed with *T. viride* as soil drench followed by *T. harzianum* as foliar spray, which gave 972 and

960.6 cm², respectively in the first season. However in the second season, the *T. harzianum* treatment recorded 941.3, 906.6 and 863.6 cm² respectively at three application method viz. 526.6 and 546.3 cm² for the control in the two seasons respectively. This result showed that treatments with *Trichoderma* bioagents as foliage spray on leaves of sunflower with the different application methods induced significant protection and led to an increase in growth of the leaves of sunflower plants (Chander *et al.*, (2010).

Table 4. Effect of *Trichoderma* species with different application methods on yield of sunflower plants under field condition, during growing season (2015-2016)

Bioagents	Foliar spray						Foliar spray + soil drench						Soil drench					
	HE. DI.		ST. DI.		LE. Ar.		HE. DI.		ST. DI.		LE. Ar.		HE. DI.		ST. DI.		LE. Ar.	
	(cm.)	(cm.)	(cm.)	(cm.)	(cm.2)	(cm.2)	(cm.)	(cm.)	(cm.)	(cm.)	(cm.2)	(cm.2)	(cm.)	(cm.)	(cm.)	(cm.)	(cm.2)	(cm.2)
<i>T. harzianum</i>	30.0a	29.0a	4.51a	4.45a	960.0a	941.3a	27.3a	27.3a	3.50a	3.61a	866.6ab	906.6a	26.6a	27.0ab	3.30b	3.20c	898.6ab	863.6ab
<i>T. viride</i>	31.0a	27.3ab	4.11b	4.28a	915.6a	899.3ab	27.0a	29.0a	3.49a	3.52b	903.0a	813.0a	25.6a	28.0a	3.40a	3.30b	972.0a	900.6a
<i>T. hamatum</i>	28.3a	27.0ab	3.50c	3.79b	706.0b	775.3b	26.0a	27.6a	3.32b	3.24cd	677.3c	765.6b	29.3a	27.3ab	3.30b	3.20d	763.3bc	678.6bc
<i>T. koningii</i>	26.0a	22.6bc	3.58c	3.00c	649.0b	673.3c	26.0a	27.6a	3.30b	3.20d	731.0bc	717.6b	30.3a	29.0a	3.20c	3.20cd	687.3c	737.6abc
<i>T. reesei</i>	26.6a	28.6a	3.62c	3.58b	938.0a	852.3ab	27.6a	29.3a	3.41ab	3.46b	866.6ab	811.0ab	28.0a	26.3ab	3.30b	3.40a	848.6ab	739.0abc
<i>T. polysporum</i>	26.3a	26.6ab	3.50c	3.38b	853.0a	811.0b	27.6a	26.6a	3.17c	3.30c	805.0abc	720.6b	27.3a	27.0ab	3.30ab	3.20cd	833.6ab	822.0ab
control	16.0b	19.3c	2.60d	2.71c	526.6c	546.3d	16.0b	19.3b	2.60d	2.71e	526.6d	546.3c	16.0b	19.3b	2.60d	2.71e	526.6d	546.3c
Mean	26.33	25.70	3.63	3.59	792.60	785.54	25.38	26.71	3.25	3.27	768.0	762.9	26.19	26.28	3.25	3.21	790.04	755.3
L.S.D at 5%	6.31	3.86	0.12	0.34	79.83	95.06	5.99	4.45	0.10	0.06	117.22	81.6	5.94	5.59	0.05	0.04	105.03	146.9

HE- DI: Head diameter

ST- DI: stem diameter

LE- AR: leaf area

Mean with the same letters within each column are not significant differences at 0.05 level of probability.

Yield components:

Data in (Tables 5) show the relationship between the tested *Trichoderma* spp. bioagents and yield components *i.e.*, seed number per head, 1000 seed weight and seed yield per plant compared with untreated plants with the different application method on sunflower plants during the two seasons 2015/2016. Data also showed significant differences between treatments, applying *T. harzianum* with foliar spray led to the highest number of seed per head in the first season, as gave 1711 seed per head followed by *T. viride* with the soil drench method in the second season, which gave 1636.3 seed per head compared with untreated control 707.6 and 730.3 seed per head in both season 2015/16. On the other hand,

weight of 1000 seed and seed yield per plant were recorded, the highest values of both weight character found with the two bioagents *T. harzianum* and *T. viride* followed by *T. reesei* in both seasons 2015 and 2016. This increase of weight might be due to increases of plants oxygen uptake capacity with an associated increase in chlorophyll II production an increase in the permeability of plant membranes and improving the uptake of nutrient. Although, data indicated that specific strains of fungi in the genus *Trichoderma* colonize and penetrate plant head tissues and initiate a series of morphological and biochemical changes in the plant. (Janisiewicz and Korsten (2002).

Table 5. Effect of *Trichoderma* species with different application methods on yield of sunflower plants under field conditions, during growing season (2015-2016)

Bioagents	Foliar spray						Foliar spray+ soil drench						Soil drench					
	(No.) of seeds /head		Weight of 1000 seed		Seed yield/ plants		(No.) of seeds/ head		Weight of 1000 seed		Seed yield/ plant		(No.) of seeds/ head		Weight of 1000 seed		Seed yield/ plant	
	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016
<i>T. harzianum</i>	1711.0a	1595.3a	90.5a	90.6a	139.6a	139.0a	1103.3b	1122.3bc	90.2a	83.9ab	104.3a	130.0a	1665.0a	1204.6b	77.3b	83.0ab	134.3a	121.3ab
<i>T. viride</i>	1370.0ab	1570.0a	90.1a	87.0a	131.3a	127.0ab	1418.6a	1308.6b	77.3ab	87.1ab	112.6a	129.3a	1458.3ab	1636.3a	90.3a	90.6a	136.6a	132.0a
<i>T. hamatum</i>	1296.6ab	1252.6ab	76.8b	77.1a	110.6b	101.6c	1079.3b	1003.0c	80.3ab	77.1ab	105.3a	109.6a	1053.3c	1040.6b	73.93b	77.1b	100.3c	103.0b
<i>T. Koningii</i>	953.3bc	1043.0ab	73.5b	80.3a	93.3b	104.3c	1182.0ab	1107.3bc	73.8b	77.0ab	109.6a	113.6a	1245.6bc	1150.0b	73.7b	73.8b	114.0bc	109.3ab
<i>T. reesei</i>	1190.0ab	1243.3ab	87.16a	87.0a	109.6b	102.3c	1414.3a	1599.3a	90.5a	90.53a	126.0a	131.3a	1393.3b	1223.0b	90.1a	83.9ab	124.6ab	116.0ab
<i>T. Polysporum</i>	1496.0ab	1180.0ab	73.7b	76.9a	105b	111.3bc	1181.3ab	1171.3bc	77.1ab	74.1b	109.6a	118.0a	1316.0bc	1186.3b	77.2b	80.5ab	119.3ab	121.0ab
Control	707.6c	730.3b	60.1c	57.0b	67.0c	69.0d	707.6c	730.3d	60.1c	57.1c	67.0b	69.0b	707.6d	730.3c	60.1c	57.1c	67.0d	69.0c
Mean	1246.38	1230.6	78.86	79.44	109.64	107.8	1155.2	1148.90	78.45	78.1	104.95	114.42	1262.8	1167.3	77.54	78.12	113.7	110.23
L.S.Dat 5%	392.14	475.6	8.18	10.60	16.79	15.67	192.02	174.8	9.23	9.27	17.31	19.76	216.2	204.48	6.90	8.35	13.90	15.73

Means with the same letters within each column are not significant differences at 0.05 level of probability.

Data in Table (6) indicate that *T.harzianum* showed the highest values of yield per plot and total yield per feddan with the foliar spray method which gave [8.38 k.g- 8.34 k.g] and [1.952 (ton)- 1.958 (ton)] respectively in both seasons 2015 and 2016 compared with the other treatments.

Meanwhile the lowest values of seed yield per plot was observed with *T.hamatum* which gave the lowest values in yield per plot with the different application methods being (6.10, 6.58 and 6.18 k.g) per plot in the second season, compared with untreated control, being (4.14 k.g) in the

same season. On the other hand, the total yield per feddan gave the same trend with the highest values of yield per feddan with all *Trichoderma* isolates compared with untreated control plants (Dange *et al.*, (2005)). Although, data in Tables (6) represent the effect of applying bioagents on yiled component, the applying of *Trichoderma* bioagents *i.e* *T.harzianum* as foliar spray method followed by *T.viride* and *T. reesei* resulted in the highest weight of seed per plot and also the highest yield per feddan in both seasons 2015/2016 compared with untreated control plants. This increase of

weight might be due to the major mechanisms involved in the biocontrol activity of *Trichoderma* spp. *i.e* competition for space and nutrients, production of diffusible or volatile antibiotics and hydrolytic enzymes like chitinase and B- 1,3-glucanase. These hydrolytic enzymes partially degrade the pathogen cell wall and lead to its parasitization (Kubicek *et al.*, 2001). Many recent findings suggest that plant development and biochemistry are strongly affected by *Trichoderma* strains (Lorito *et al.*, 2010).

Table 6. Effect of *Trichoderma* species with different application methods on total yield, under field conditions during growing season (2015 and 2016)

Bioagents	Foliar spay				Foliar spray +soil drench				Soil drench			
	Yield / plot(kg)		Yield (ton)/ feddan		Yield / plot(kg)		Yield (ton) Feddan		Yield/plot (Kg)		Yield(ton)/ feddan	
	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016
<i>T.harzianum</i>	8.38a	8.34a	1.952a	1.958a	6.46a	7.80a	1.502a	1.871a	8.06a	7.28ab	1.943a	1.746ab
<i>T. viride</i>	7.88ab	7.62ab	1.891a	1.828ab	6.76a	7.76a	1.622a	1.862a	8.20a	7.92a	1.967a	1.900a
<i>T.hamatum</i>	6.64bc	6.10c	1.593ab	1.463c	6.32a	6.58a	1.516a	1.578a	6.02c	6.18b	1.444c	1.483b
<i>T.koningii</i>	5.60c	6.26c	1.343b	1.505c	6.58a	6.82a	1.579a	1.636a	6.84bc	6.56ab	1.641bc	1.573ab
<i>T. reesei</i>	6.58bc	6.14c	1.579ab	1.473c	7.56a	7.88a	1.814a	1.891a	7.48ab	6.96ab	1.794ab	1.670ab
<i>T. polysporum</i>	6.30bc	6.68bc	1.511ab	1.602bc	6.58a	7.08a	1.597a	1.699a	7.16ab	7.26ab	1.718ab	1.742ab
Control	4.02d	4.14d	964.6c	993.3d	4.02b	4.14b	964.6b	993.3b	4.02d	4.14c	964.6d	993.3c
Mean	6.48	6.46	1.547	1.546	6.32	6.86	1511	1647.42	6.82	6.61	1637.80	1587
L.S.D at 5%	1.30	0.98	321.5	235.9	1.07	1.21	249.34	284.71	0.83	0.94	200.02	226.76

Means with the same letters within each column are not significant differences at 0.05 level of probability

Recently, using biological control agents led to the reduction in the disease components and increased the weight of yield per plot as well as weight yield per feddan.

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فاعلية بعض أنواع لجنس التريكوثيرما على مكافحة عفن القرص في عباد الشمس

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يهدف هذا البحث الي:- دراسة تاثير التضاد الحيوى لأنواع مختلفة من التريكوثيرما من خلال اضافاتها بطرق تطبيقية متنوعة على التقليل من عفن القرص لنبات عباد الشمس وذلك لأهم الفطريات المسببة للإصابة وهي بوترايتس سينيريا وفطر الريزوكتونيا سولاني وكذلك فطريات اسبراجلس فلافس وفيوزاريوم مونيليفورم وريزوبس استولينييفر. وذلك تحت الظروف التطبيقية للحقل. فتحت ظروف المعمل أختبرت القدرة التضادية لجميع أنواع التريكوثيرما المستخدمة على الفطريات المسببة لعفن القرص في عباد الشمس. بينما تحت ظروف الحقل فقد تم عمل انتخاب للأنواع المختلفة المستخدمة وهي تريكوثيرما هاريزيانم وفيردي وهامتم وكونجبي وريسي وترايكوديرما بولي سبوروم خلال موسمي 2015، 2016م في محطة ايتاي البارود بمحافظة البحيرة. حيث تم الرش بالمعاملات الحيويه بثلاث طرق تطبيقية مختلفة وهي اما رشا للنباتات او اضافة المحلول للتربة او المعاملة المزدوجة بهما وذلك خلال ثلاث فترات زمنية مختلفة هي 30، 45، 60 يوما بعد الزراعة. وقد تم تقدير تأثير المعاملات وطرق الرش المختلفة على نسبة الإصابة وكذلك على مكونات المحصول وكميته الكلية مقارنة بالكنترول (النباتات الغير معاملة). - وقد اوضحت النتائج: أن كل المعاملات المستخدمة مع وسائل الاضافة المختلفة كانت فعالة في تقليل كلا من شدة الإصابة وزيادة الانتاجية لجميع الصفات المحصولية للمحصول جنباً الى جنب مع كمية البذور المتحصل عليها من المحصول من حيث الكم ونوعية المحصول وجودته وذلك مقارنة بالنباتات الغير معاملة (الكنترول). كما اوضحت النتائج ان أعلى تاثير تضادي تحت ظروف الحقل كان مع الفطر ترايكوديرما هاريزيانم عندما طبقت في صورة رشا للنباتات حيث اعطت اعلى نسبة في خفض المرض واعلى زيادة في كمية المحصول وذلك مقارنة بالكنترول خلال الموسمين 2015 – 2016 وتلي التأثير الفعال لهذا النوع ترايكوديرما فيردي عندما طبقت كإضافة للتربة ثم النوع ترايكوديرما ريسي عندما طبقت في حالة المعاملة المزدوجة (كإضافة للتربة و رشا للنباتات معاً) على الترتيب.