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Effect of *Trichoderma harzianum* as biocontrol agent on wheat damping-off disease

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Abstract:

Damping-off disease caused by certain Fusarium spp. and other fungal species is an important diseases of wheat. Isolations from infected wheat seedling revealed that presence of *Fusarium oxysporum*, *Fusarium chlamydosprum*, *Fusarium lateritium*, *Fusarium proliferatum*, *Fusarium equiseti*, *Rhizoctonia solani* and *Macrophomina phaseolina*. This pathogens were able to infect Giza-168 and Banyswif-1 wheat cultivars causing damping-off. The antagonistic capability of the isolated fungi against the pathogens revealed that 11 fungal isolates out of 53 tested isolates showed moderately and highly antagonistic effect against all tested pathogenic fungi. Two *Trichoderma harzianum* isolates gave over growth upon the mycelia growth of the pathogens. Soil treatment with *T.harzianum* gave significantly reduction in incidence of damping-off on Giza-168 and Banyswif-1 cultivars under greenhouse conditions.

Keywords: Biological control, Wheat, Damping–off, *Trichoderma harzianum*. *Fusarium*, *Macrophomina*, *Rhizoctonia solani*.

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Introduction

Wheat (*Triticum aestivum* L. em Thell.) is the most widely grown grain crop in the world. At least one-third of the world's population depends on wheat. The principle wheat food was used as bread, flour and pasta. It has numerous diseases problems, some of which have been eradicated by resistant wheat strains and new fungicides. There are still management problems for common diseases like *Fusarium* rots. Several species of *Fusarium* survive in fruiting bodies in the soil. The fungus causes root rot and damping-off. (Stephen and klien, 1998). Damping-off disease caused by *F. oxysporum F. lateritium, F.equiseti, F. chlamydosporum, F. proliferatum, R. solani* and *M. phaseolina* is an important diseases of wheat (Chen *et al.*, 1996; Hajieghrari, 2009 and Saremi *et al.* 2011). Fungicides may lead to the appearance of new resistant strains of pathogens. Biological control of plant disease especially soil borne plant pathogens by using microorganisms has been considered a more natural and environmentally acceptable alternative to the existing chemical treatment methods (Barker and Panlitz, 1996 and Eziashi *et al.*, 2007). The mycoparasite ability of *Trichoderma* species against some economically important aerial and soil borne plant pathogens was studied by several investigators (Papavizas, 1985; Elad *et al.*, 1993; Elad, 2000; Freeman *et al.*, 2004 and Dubey *et al.*, 2007). *Trichoderma* species reduces the incidence of soil borne plant pathogenic fungi under natural conditions (Sivan and Chet, 1986 and Calvet *et al.*, 1990).

Trichoderma harzianum is an efficient biocontrol agent that is commercially produced to prevent development of several soil pathogenic fungi. Different mechanisms have been suggested as being responsible for their biocontrol activity, which include competition for space and nutrients, secretion of chitinolytic enzymes, mycoparasitism and production of inhibitory compounds (Hassan,1992 and Howell, 2003).

The objectives of this investigation were to reduce wheat damping off disease caused by certain *Fusarium* spp., *Rhizoctonia solani* and *Macrophomina phaseolina* .

Materials and Methods

1- Pathogens isolation and identification

One hundred and two fungalisolates were collected from different wheat fields located at Assiut, Sohag and Aswan governorates. Pathogen isolates were isolated from infected wheat showing damping off according the methods described by Hajieghrari, (2009). Isolated fungi were purified using single spore and hyphal tip techniques and identified on basis of morphological and culture characteristic according to Domsch *et al.* (2007). Then confirmed by Assiut University, Mycological Center (AUMC).

2 - Pathogenicity testes:

Twenty isolates were distinguished on basis of primary pathogenic capability tests and used for pathogenicity tests on Giza-168 and Banyswif-1 cultivars under greenhouse conditions during growing season 2010 / 2011. Pathogens inocula were prepared on barley medium as described by Ahmed *et al.*, (2009). The sterilized barley grains were inoculated with 5 mycelial blocks (0.5-mm in diameter) 5- days old. The inoculated flasks were kept under room temperature for 3 weeks. The flasks were shaken with hand every alter-

nate day. The autoclaved pot (25 cm in diameter) filled with autoclaved soil and inoculated with fungal inoculums at the rate of 70 g/kg of soil. Three pots were used as replicates. Ten seeds of Giza- 168 and/ or Banyswif -1 cultivars were surface sterilized by dipping in 3% sodium hypo chlorite solution for 3 min. followed by washing with sterilized water and seeded in every pot containing infested and non infested soil. Pre and post damping-off were recoded after 15 and 30 days respectively.

3- Isolation and Identification of the antagonistic:

Isolation of fungal antagonistis from wheat rhizospher were carried out by using method described by Al-Mahareeq (2005). Antagonistic isolates were purified using single spore and hyphal tip techniques and identified on basis of morphological and culture characteristic according to Domsch *et al.* (2007).The identification was confirmed by Assiut University, Mycological Center (AUMC).

4- Evaluation of antagonistic activity in dual culture technique *(in vitro)* :

The antagonistic capability of fifty three fungal isolates isolated from wheat rhizosphere was tested against the tested pathogenic fungi in dual culture *in vitro*. The highly pathogenic isolates of *F. chlamydosporium* (Isolate No. III), *R. solani* (Isolate No. II) and *M. phaseolina* (Isolate No. I) were selected for this study. Petri dishes (9 cm. in diameter) each containing 10 ml of PDA medium, were seeded with 5-mm equal disks of the tested fungi obtained from 4 days old cultures grown on PDA medium at 25±1°C.

A disc (5 mm in diameter) of *F. chlamydosporium, R. solani* and *M. phaseolina* were inoculated at equal distance of the opposite side of Petri dish. Plates inoculated with pathogenic fungi only were used as control and three replications were used for each test. The inoculated plates were incubated at 25±1°C.

Observation on antagonism and/ or mycoparasitism of the tested fungi were recorded when the growth of the pathogenic fungi completely covered the plate surface in control treatments. The following arbitrary scale index was used to estimate antagonism or mycoparasitism of the tested fungi (Hassan, 1992; Azza and Allam, 2004; AbdulKareem, 2011) as follows : 0 = No antagonism, 1 = slightly antagonism, 2 = moderately antagonism , 3 = highly antagonism and 4 = over growth (mycoparasitism).

5- Culture filtrate (nonvolatile) and early volatile metabolites testes:

Culture filtrates of 6 tested fungi showed over growth upon pathogen mycelium were tested by growing fungi in conical flasks (250 ml) each contained 100 ml of Czapek's solution agar liquid medium at 25 ±1°C. After 14 days incubation period, mycelia mats was discarded by filtration through filter papers and culture filtrates were sterilized by passing through sterilized Schleicher and Schuell filter (0.2 µm. , 7 bar max.). Sterilized culture filtrates were added to autoclaved Potatoes Dextrose Agar (PDA) medium to get 10% concentration (v/v) before dispensing medium in Petri dishes, when the temperature of the medium was about 50°C. PDA medium without addition of culture filtrates of antagonists was used as controls. Disks 5-mm. in diameter, from 5-days old cultures of each pathogenic fungus were transferred to the center of dishes and incubated at

25±1°C. Three replicates were used for each treatment. After 6 days incubation period, liner growth of tested pathogens was recorded. Percent growth inhibition rate was calculated according to the formula used by Moubarak and Abdel-Monaim (2011) as follows:

PI= [(C- T) /C] X 100

Where; PI= Percent growth inhibition rate

C= Radial growth of the pathogen in control plates

T = Radial growth of the pathogen in treated plates

6- Biological control of wheat damping-off by *Trichoderma harzianum* under greenhouse conditions.

The effect of *T. harzianum* on incidence of damping-off of wheat were carried out under greenhouse condition at the Assiut University Greenhouse during growing seasons 2010/2011 and 2011/2012. Salty loam soil with 1% organic maters was used. Completely randomize designs with three replicates were accomplished in greenhouse. Inoculum of pathogens and *T. harzianum* were prepared as above mentioned described in pathogenicity test.

Five isolates from *Fusarium* and one isolate of R. *solani* , one isolate of *M. phaseolina,* two isolates of *T.harziznum* were used in this study.

In greenhouse tests, Pots (25-cm in diameter) were filled with sterilized clay loam soil. *Trichoderma* inoculum was added at the same time of soil infestation with pathogens inoculum at rate 70 g /kg. Pots seeded with 10 seeds of poth Giza-168 and Banyswif-1 wheat cultivars. Pots irrigated and fertilized as recommended, three pots was used as replicates and three replicates without treatment was used as control. Pre and post emergence damping-off were recorded after 15 and 30 days, respectively.

7- Statically analysis:

Data was subjected to statistical analyses of variance was carried out using the MSTATC computer program. Means were compared using L.S.D tests at P≤ 0.05 according to Gomez and Gomez (1984).

Experimental Results

1-Isolation, identification and isolates sources of the causal pathogen:

Twenty fungal isolates were isolated from wheat seedling showing symptoms of damping-off collected from different localities of Assiut, Aswan and Sohag governorate. The isolated fungi were identified on basis of morphological and culture characteristic according to Domsch *et al.* (2007). Identification of the isolated fungi presented in Table (1) revealed that the fungal isolates were 6 isolates of *F. oxysporum* (Schlecht. Exfr. Emend. Snyder&Hansen), three isolates of *F. chlamydosprum* (Wollenw.&Reinking). three isolates of *F. lateritium* (Nees), one isolate *F. proliferatum* (Matsushima) Nirenberg, one isolate of *F. equiseti* (Corda) Sacc, four isolates of *Rhizoctonia solani* (Kuhn), and two isolates of *M. phaseolina* (Tassi) Goidanich.

2- Pathogenicity testes:

Isolated fungi were tested for their pathogenic capability during growing season 2010/ 2011 on Giza-168 and Banyswif-1wheat cultivars. Data presented in Table (2) indicate that all the tested fungal isolates were able to infect wheat plants causing damping-off with varied degrees. Data indicated that, generally in both wheat cultivars *R. solani* (II) and *F. oxysporum* (II and IV) caused the highest total infection and signifi-

cantly incident of disease. While *Rhizoctonia solani* (III), *F. oxysporum* (VI), *M. phaseolina* (I and II) , *F. chlamydosprium* (I and II), *F. lateritium* (II and III) caused the lowest total infection. The rest of tested isolates are in between.

Rhizoctonia solani(II), *F. chlamydosprium* (III), *F. oxysporum* isolate (II), *F.lateritium* (I), *F. proliferatum* (I), *F. equiseti* (I) and *M. phaseolina* (I) were selected for further studies.

3- Preliminary test for antagonistic capability of the fungal isolates:

Fifty three fungal isolates isolated from rhizosphere of wheat were tested against the caused pathogens of wheat damping-off *in vitro*. The highly pathogenic fungal isolates was selected for this study.

Data presented in Table (3) indicated that only 11 fungal isolates out of 53 tested isolates exhibited moderately and highly antagonistic effect against all tested pathogenic fungi. However, 6 fungal isolates showed over growth upon tested pathogenic fungi. The rest of fungal isolates gave negative or slight antagonistic effect.

4- Effect of culture filtrate on radial growth of the tested Pathogen fungi *in vitro* .

Culture filtrate of *Trichoderma harzianum*, *T. longibrachiatum*, *T. atroviride* and *Aspergillus flavus* obtained from rhizoshere of wheat were tested *in vitro* against *Rhizoctonia solani*(II), *F. chlamydosprium* (III), *F. oxysporum* (II), *F.lateritium* (I), *F. proliferatum* (I), *F. equiseti* (I) and *M. phaseolina* (I), the causal pathogens of wheat damping-off . In general Data in Table (4) indicate that either *T. harzianum*, *T. longibrachiatum*, *T. atroviride* and *Aspergillus flavus* or its filtrate inhibited the growth of the causal pathogens. Data also indicate that treatment with culture filtrate of these antagonistics reduce significantly the liner growth of the tested pathogens compared with control *in vitro*.

5- Effect of *Trichoderma harzianum* on incidence of damping-off on two wheat cultivars under greenhouse conditions during growing seasons 2010/2011 and 2011/2012

The effect of two isolates of *T. harzianum* on incidence of wheat damping-off on two wheat cultivars during two successive growing seasons were carried out under greenhouse conditions. The results of this study are presented in Tables (5-8). I n general soil treatments with two *T. harzianum* (I and II) during both seasons 2010/2011 and 2011/2012 reduced the total infection *F. oxysporum*, *F. lateritium*, *F.chlamydosporum R. solani* and *M. phaseolina* compared with the untreated soil of wheat cultivar Giza-168 and Banyswif-1.

A- On Giza-168 wheat cultivar:

Data in Tables (5 and 6) show that *T.harzianum* (I and II) significantly reduce the total infection of *F.chlamydosporum* during two growing seasons compared with the untreated. Data also indicate that *T.harzianum* (I and II) significantly reduced total infection of *F. lateritium*, *M. phaseolina* and *F. oxysporum* and gave less effect against other fungi during growing season 2011/2012 whereas no significantly effect against pathogens during growing season 2010/2011. Data also indicate that *T.harzianum* (I and II) were not significantly effected on *F. equiseti* and *F. proliferatum* during two

growing seasons compared with the untreated.

B- On Banyswif-1 wheat cultivar: Data presented in Tables (7 and 8) indicated that *T. harzianum* (I and II) significantly reduced total infection of *M. phaseolina, F. oxysporum* and *F. chlamydosporum* during growing season 2011/2012 compared with the untreated. Data also indicate that *T.harzianum* (I and II) were not significantly effected on *F. equiseti* and *F. proliferatum* during two growing seasons compared with the untreated.

Discussion

Damping-off disease caused by certain Fusarium spp. and other fungal species is an important diseases of wheat. Isolations from infected wheat seedling revealed that presence of F. *oxysporum, F. chlamydosprum, F. lateritium, F. proliferatum, F. equiseti, R. solani* and *M. phaseolina* . According to the available literature, the pathogens were isolated from wheat and caused damping-ff on wheat cultivars (Fouly *et al.* 1996; Moubarak and Abdel-Monaim, 2011 and Abo-Elnaga, 2012). Pathogenicity tests of isolated fungi was carried out on two wheat cultivars (Giza-168 and Banyswif -1) and reviled that all the tested fungal isolates were able to infect wheat plants causing damping-off. Data reported herein indicate that *R. solani, F. chlamydosprium, F. oxysporum, F. proliferatum, F. lateritium* and *F. equiseti* caused the highest total infection and significantly incident of disease. These results are in harmony with those reported by Hashem and Hamada (2002), Fernandez and Chen (2005), Atef (2008) and Moubarak and Abdel-Monaim (2011). Isolation of antagonistic fungal from rhizosphere of wheat soil resulted in 53 antagonistis isolates. Testing antagonistic capability of the isolated fungi pathogen in bicultural studies revealed that 11 fungal isolates out of 53 tested isolates showed moderately and highly antagonistic effect against all tested pathogenic fungi. However, 6 fungal isolates showed over growth upon tested pathogenic fungi. Such results are in agreement with those reported by Hassan(1992), Azza and Allam (2004) and AbdulKareem (2011). The selected antagonistis varied in their inhibitory effect on radial growth of the tested pathogen. *T. harzianum* has been reported as the best antagonists for damping-off disease caused by *F. oxysporum, F. chlamydosprum, F. lateritium, F. proliferatum, F. equiseti, R. solani* and *M. phaseolina* under laboratory condition. *T. harzianum* completely over grew on the colony of the pathogens fungi. The results are agree with those reported by El-Nashar *et al.*(2001), Atef (2008), Hajieghrari *et al.*(2008), Waheed and Khilare (2010), Hassan Dar *et al.*(2011), and Abo-Elnaga (2012). They reported similar results in their studies on one or more of the tested microorganisms. Soil treatment with *T. harzianum* demonstrated reduction in incidence of damping-off in Giza-186 and Banyswif -1 cultivars under greenhouse conditions. These results are in accordance with those reported by Atef (2008), Moubarak and Abdel-Monaim (2011), Haggag and Mohamed (2011), Abo-Elnaga (2012) and El-Bramawy and El-Sarag (2012). The highest significant values of *T.harzianum* for suppressing *F. chlamydosporium and F. oxysporium and M. phaseolina.*

Tricoderma species may be very useful in biological control against wheat and toxygenic Fusarium species to reduced their inoculums and to

prevent Fusarium mycotoxin accumulation in plant tissues (Busko *et al.*, 2007 and Abo-Elnaga, 2012).

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Table 1. The fungal isolates and their Sources

No.	Isolates		Source of isolates
1	*Fusarium oxysporum*	(I)	Abnoub
2	*Fusarium oxysporum*	(II)	Dirout
3	*Fusarium oxysporum*	(III)	Sahel Sleem
4	*Fusarium oxysporum*	(IV)	Manfalout
5	*Fusarium oxysporum*	(V)	Manfalout
6	*Fusarium oxysporum*	(V1)	Manfalout
7	*Fusarium chlamydosprum*	(I)	Assiut
8	*Fusarium chlamydosprum*	(II)	Sahel Sleem
9	*Fusarium chlamydosprum*	(III)	Aswan
10	*Fusarium lateritium*	(I)	Albadari
11	*Fusarium lateritium*	(II)	Sohag
12	*Fusarium lateritium*	(III)	Aswan
13	*Fusarium proliferatum*	(I)	Alfath
14	*Fusarium equiseti*	(I)	Sohag
15	*Rhizoctonia solani*	(I)	Dirout
16	*Rhizoctonia solani*	(II)	Dirout
17	*Rhizoctonia solani*	(III)	Sahel Sleem
18	*Rhizoctonia solani*	(IV)	Sohag
19	*Macrophomina phaseolina*	(I)	Elkosya
20	*Macrophomina phaseolina*	(II)	Aswan

Table 2. Pathogenic capability of 20 fungal isolates on "Giza -168" and "Banyswif -1" wheat cultivars under greenhouse condition.

Cultivar / Pathogen		Giza-168			Banyswif-1		
		Damping-off (%)			Damping-off (%)		
		Pre	Post	Total	Pre	Post	Total
F. oxysporum	(I)	13.3	10.0	23.3	36.7	10.0	46.7
F. oxysporum	(II)	40.0	10.0	50.0	43.3	3.30	46.6
F. oxysporum	(III)	23.3	00.0	23.3	43.3	13.3	56.6
F. oxysporum	(IV)	26.7	00.0	26.7	43.3	3.30	46.6
F. oxysporum	(V)	16.7	10.0	26.7	23.3	3.30	26.6
F. oxysporum	(V1)	16.7	03.3	20.0	23.3	3.30	26.6
F. chlamydosprum	(I)	16.7	03.3	20.0	23.3	10.0	33.3
F. chlamydosprum	(II)	20.0	03.3	23.3	13.3	20.0	33.3
F. chlamydosprum	(III)	43.3	06.7	50.0	13.3	6.70	20.0
F. lateritium	(I)	26.7	06.7	33.4	20.0	13.3	33.3
F. lateritium	(II)	16.7	03.3	20.0	13.3	3.30	16.6
F. lateritium	(III)	13.3	10.0	23.3	30.0	0.00	30.0
F. proliferatum	(I)	26.7	10.0	36.7	16.7	0.00	16.7
F. equiseti	(I)	23.3	06.7	30.0	20.0	13.3	33.3
R. solani	(I)	23.3	06.7	30.0	23.3	10.0	33.3
R. solani	(II)	53.3	03.3	56.6	56.7	3.30	60.0
R. solani	(III)	23.3	00.0	23.3	26.7	0.00	26.7
R. solani	(IV)	16.7	06.7	23.4	36.7	6.70	43.4
M. phaseolina	(I)	10.0	03.3	13.3	13.3	10.0	23.3
M. phaseolina	(II)	0.00	06.7	6.70	23.3	3.30	26.6
Control		0.00	00.0	0.00	0.00	0.00	0.00
L.S.D. at 5 %		23.7	12.8	25.4	36.4	15	37

Table 3. Antagonistic effect of isolated fungi against growth of Fusarium chlamydosporum, Rhizoctonia solani and Macrophomina phaseolina in vitro

No. of isolates	Pathogenic fungi			Mean
	F. chlamydosporum	*R. solani*	*M. Phaseolina*	
1-22	0	0	0	0.00
23-28	4	4	4	4.00
29	0	0	1	0.33
30	1	0	1	0.66
31	0	4	0	1.33
32	0	4	0	1.33
33	2	2	3	2.33
34	0	1	0	0.33
35	0	3	0	1.00
36	0	0	4	1.33
37	3	4	3	3.33
38	0	3	0	1.00
39	0	1	1	0.66
40	4	3	2	3.00
41	4	0	2	2.00
42	4	0	4	2.66
43	4	3	0	2.33
44	4	3	3	3.33
45	0	3	0	1.00
46	4	3	3	3.33
47	0	0	2	0.66
48	4	1	3	2.66
49	1	0	1	0.66
50	4	3	0	2.33
51	4	0	0	1.33
52	4	4	0	2.66
53	2	2	0	1.33

Arbitrary antagonism scal:
0 = No antagonism
1 = Slightly antagonism
2 = Moderately antagonism
3 = High antagonism
4 = Over growth

Table 4. Effect of culture filtrate of 6 antagonists on radial growth of the 7 tested pathogenic fungi in vitro

Antagonists isolates	*F.lateritium*		*R. solani*		*M. phaseolina*		*F.equiseti*		*F.oxysporum*		*F.chlamydosporum*		*F.proliferatum*	
	Liner growth (mm)	Inhi-bition (%)	Liner growth (mm)	Inhi-bition (%)	Liner growth (mm)	Inhi-bition (%)	Liner growth (mm)	Inhi-bition (%)	Liner growth (mm)	Inhi-bition (%)	Liner growth (mm)	Inhi-bition (%)	Liner growth (mm)	Inhi-bition (%)
T.harzianum I	32.0	42.2	16.7	67.6	52.2	38.7	33.7	39.9	32.7	42.0	41.7	23.8	34.7	37.0
T.longibrachiatum	27.0	51.2	21.7	55.1	51.7	39.2	27.0	51.8	23.7	57.9	25.0	54.2	25.0	54.5
T.harzianum II	11.7	78.9	17.5	66.2	11.5	86.5	13.3	76.2	13.0	76.9	19.2	64.8	15.0	72.7
T.atroviride I	51.7	06.6	40.5	21.6	83.7	1.60	51.3	08.3	51.0	09.5	50.3	7.90	51.3	06.7
T.atroviride II	36.0	34.9	36.7	29.3	21.7	74.5	34.7	38.1	33.3	40.8	23.3	57.3	34.7	37.0
Aspergillus flavus	52.7	04.8	37.8	26.7	66.2	22.0	52.7	06.0	52.7	06.5	50.3	7.90	52.3	04.9
Control	55.3	0.00	51.7	0.00	85.0	0.00	56.0	0.00	56.3	0.00	54.7	0.00	55.0	0.00
L.S.D at 5%	2.2	4.1	8.60	16.6	8.30	9.80	1.6	2.8	2.4	4.1	4.60	8.10	2.6	4.8

Table 5. Effect of Trichoderma harzianum on incidence of damping-off on " Giza-168" wheat cultivar under greenhouse conditions during growing season 2010/2011

Pathogenic fungi	Treatment	Damping-off (%)		
		Pre-emergence	Post-emergence	Total
Fusarium lateritium	*T.harzianm* I I	16.67	03.33	20.00
	T.harzianm I	10.00	03.33	13.33
	Control	26.67	03.33	30.00
	Mean	17.78	3.33	21.11
Rizoctonia solani	*T.harzianm* I I	10.00	13.33	23.33
	T.harzianm I	10.00	00.00	10.00
	Control	36.67	06.67	43.34
	Mean	18.89	06.67	25.56
Macrophomina phaseolina	*T.harzianm* I I	16.67	06.67	23.34
	T.harzianm I	06.67	03.33	10.00
	Control	23.33	06.67	30.00
	Mean	15.56	5.56	21.11
Fusarium equiseti	*T.harzianm* I I	13.33	10.00	23.33
	T.harzianm I	10.00	13.33	23.33
	Control	10.00	16.67	26.67
	Mean	11.11	13.33	24.44
Fusarium oxysporum	*T.harzianm* I I	26.67	03.33	30.00
	T.harzianm I	00.00	10.00	10.00
	Control	33.33	06.67	40.00
	Mean	20.00	06.67	26.67
Fusarium chlamydosporum	*T.harzianm* I I	06.67	16.67	23.34
	T.harzianm I	20.00	03.33	23.33
	Control	60.00	03.33	63.33
	Mean	28.89	07.78	36.67
Fusarium proliferatum	*T.harzianm* I I	03.33	03.33	06.66
	T.harzianm I	13.33	06.67	20.00
	Control	33.33	03.33	36.66
	Mean	16.67	04.44	21.11
L.S.D. 5% for:	Fungi (A)	17.20	5.96	15.33
	Treatment (B)	19.79	6.86	17.65
	A x B	34.03	14.53	31.10

Table 6. Effect of Trichoderma harzianum on incidence of damping-off on " Giza-168" wheat cultivar under greenhouse conditions during growing season 2011/2012

Pathogenic fungi	Treatment	Damping-off (%)		
		Pre-emergence	Post-emergence	Total
Fusarium lateritium	*T.harzianm* I I	03.33	06.67	10.00
	T.harzianm I	00.00	10.00	10.00
	Control	13.33	13.33	26,66
	Mean	05.55	10.00	15.55
Rizoctonia solani	*T.harzianm* I I	00.00	16.67	16.67
	T.harzianm I	03.34	23.33	26.67
	Control	13.33	13.33	26.66
	Mean	05.56	17.78	23.33
Macrophomina phaseolina	*T.harzianm* I I	03.33	10.00	13.33
	T.harzianm I	00.00	13.33	13.33
	Control	10.00	16.67	26.67
	Mean	04.44	13.33	17.78
Fusarium equiseti	*T.harzianm* I I	03.33	13.33	16.66
	T.harzianm I	03.33	16.67	20.00
	Control	13.33	10.00	23.33
	Mean	06.66	13.33	19.99
Fusarium oxysporum	*T.harzianm* I I	00.00	06.67	06.67
	T.harzianm I	00.00	10.00	10.00
	Control	10.00	20.00	30.00
	Mean	03.33	12.22	15.56
Fusarium chlamydosporum	*T.harzianm* I I	00.00	10.00	10.00
	T.harzianm I	00.00	09.00	09.00
	Control	10.00	13.33	23.33
	Mean	03.33	110.78	14.11
Fusarium proliferatum	*T.harzianm* I I	00.00	16.67	16.67
	T.harzianm I	03.33	10.00	13.33
	Control	06.67	16.67	23.34
	Mean	03.33	14.44	17.78
L.S.D. 5% for:	Fungi (A)	7.17	11.76	10.79
	Treatment (B)	8.25	13.54	12.43
	A x B	7.76	11.86	12.68

Table 7. Effect of Trichoderma harzianum on incidence of damping-off on " Banyswif-1" wheat cultivar under greenhouse conditions during growing season 2010/2011

Pathogenic fungi	Treatment	Damping-off (%)		
		Pre-emergence	Post-Emergence	Total
Fusarium lateritium	*T.harzianm* I I	06.67	06.67	13.34
	T.harzianm I	26.67	13.33	40.00
	Control	50.00	00.00	50.00
	Mean	27.78	06.67	34.45
Rizoctonia solani	*T.harzianm* I I	16.67	06.67	23.34
	T.harzianm I	06.67	10.00	16.67
	Control	46.67	03.33	50.00
	Mean	23.34	06.67	30.00
Macrophomina phaseolina	*T.harzianm* I I	23.33	00.00	23.33
	T.harzianm I	20.00	10.00	30.00
	Control	33.33	06.67	40.00
	Mean	25.55	05.56	31.11
Fusarium equiseti	*T.harzianm* I I	13.33	13.33	26.66
	T.harzianm I	26.67	06.67	33.34
	Control	40.00	10.00	50.00
	Mean	26.67	10.00	36.67
Fusarium oxysporum	*T.harzianm* I I	20.00	13.33	33.33
	T.harzianm I	13.33	13.33	26.66
	Control	36.67	10.00	46.67
	Mean	23.33	12.22	35.55
Fusarium chlamy-dosporum	*T.harzianm* I I	23.33	13.33	36.66
	T.harzianm I	23.33	03.33	26.66
	Control	50.00	00.00	50.00
	Mean	32.22	05.55	37.77
Fusarium proliferatum	*T.harzianm* I I	03.33	10.00	13.33
	T.harzianm I	26.67	00.00	26.67
	Control	13.33	20.00	33.33
	Mean	14.44	10.00	24.44
L.S.D. 5% for:	Fungi (A)	20.77	8.60	20.15
	Treatment (B)	23.90	9.90	23.20
	A x B	29.14	14.44	28.83

Table 8. Effect of Trichoderma harzianum incidence of damping-off on "Banyswif-1" wheat cultivar under greenhouse condition during growing season 2011/2012

Pathogenic fungi	Treatment	Damping-off (%)		
		Pre-emergence	Post-emergence	Total
Fusarium lateritium	*T.harzianm* I I	10.00	13.33	23.33
	T.harzianm I	03.33	16.67	20.00
	Control	13.33	13.33	26.66
	Mean	08.89	14.44	23.33
Rizoctonia solani	*T.harzianm* I I	10.00	06.67	16.67
	T.harzianm I	00.00	13.33	13.33
	Control	16.67	10.00	26.67
	Mean	08.89	10.00	18.89
Macrophomina phaseolina	*T.harzianm* I I	03.33	06.67	10.00
	T.harzianm I	03.33	06.67	10.00
	Control	16.67	13.33	30.00
	Mean	07.78	08.89	16.67
Fusarium equiseti	*T.harzianm* I I	00.00	20.00	20.00
	T.harzianm I	03.33	26.67	30.00
	Control	10.00	23.33	33.33
	Mean	04.44	23.33	27.78
Fusarium oxysporum	*T.harzianm* I I	03.33	13.33	16.66
	T.harzianm I	03.33	06.67	10.00
	Control	00.00	36.67	36.67
	Mean	02.22	18.89	21.11
Fusarium chlamydosporum	*T.harzianm* I I	03.33	23.33	26.66
	T.harzianm I	00.00	10.00	10.00
	Control	20.00	10.00	30.00
	Mean	07.78	14.44	22.22
Fusarium proliferatum	*T.harzianm* I I	00.00	23.33	23.33
	T.harzianm I	03.33	23.33	26.66
	Control	00.00	30.00	30.00
	Mean	01.11	25.55	26.66
L.S.D. 5% for	Fungi (A)	8.16	7.64	8.49
	Treatment (B)	9.39	8.79	9.78
	A x B	35.71	15.85	15.20

تاثير الفطر تريكودرما هارزيانم كعامل مكافحة حيوية لمرض الذبول المفاجئ لبادرات القمح

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مرض موت البادرات المفاجئ الناجم عن بعض انواع الجنس فيوزاريم وبعض الفطريات الاخرى يعتبر من أهم أمراض البادرات فى القمح. أمكن العزل من بادرات القمح المصابه والمتحصل عليها من مناطق مختلفة من محافظات أسيوط وسوهاج وأسوان كل من الفطريات

Fusarium oxysporum, F.chlamydosprum F.lateritium, F.proliferatum, F.equiseti, Rizoctonia solani and Macrophomina phaseolina

وتبين من اختبار القدرة المرضيه لها ان جميع المسببات المرضية المذكوره كانت قادرة على اصابة صنفى القمح جيزه ١٦٨ وبنى سويف ١ مسببة مرض الموت المفاجئ للبادرات. كما تبين من اختبارات التضاد لعدد ٥٣ عزلة فطرية تم عزلها من منطقة جذور القمح واختبار قدرتها على تضاد المسببات المرضية. تبين ان ١١ عزلة منها أظهرت تضاد عالى أو متوسط الدرجة للفطريات الممرضة المختبره . كما أظهرت ٦ عزلات فطرية نمو فوق ميسليوم الفطريات الممرضة منها عزلتين من الفطر *Trichoderma harzianum*، ثم تم معاملة التربة بهما واختبار تاثيرهما على مكافحة المرض تحت ظروف الصوبة الزجاجية وتبين من الدراسة انخفاض معنوى فى حدوث مرض موت البادرات المفاجئ على صنفى القمح جيزه ١٦٨ وبنى سويف ١.