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FIRST RECORD OF PLOND PSYLLIUM (*Plantago ovata* FORSK.) ROOT ROT AND WILT DISEASES IN EGYPT

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ABSTRACT

Surveying the plantation areas of blond psyllium (*plantago ovata*), as a new remunerative medicinal crop grown during 2005/2006 and 2006/2007 winter seasons, showed that the wilt and root rot diseases were recorded for the first time in Egypt. In the experimental area at Agronomy farm, plants showed symptoms of yellowing stunting and dried shoots, as well as xylem discoloration and a cortical root rot were noticed.

Isolation trials yielded *Fusarium incarnatum*, *F. oxysporum*, *F. solani*, *Rhizoctonia solani* and *Sclerotinia sclerotiorum*. Pathogenicity tests indicated that all isolates were equal in their significant effects in pre-emergence damping off symptoms. Where, *F. incarnatum*, *F. oxysporum* and *F. solani* were the most virulent in post-emergence damping off, while *R. solani* and *S. sclerotiorum* were the last ones.

From *Trichoderma viride*, *T. harzianum*, *T. hamatum*, *Gliocladium virens,* and *G. rosum*, *T. viride* and *T. harzianum* antagonized *F. incarnatum* and showed the maximum significant inhibition effect. The radial growth of *F. solani* was highly affected by *T. harzianum* and *G. virens*. Besides, *G. virens* exhibited the highest reduction of *Rhizoctonia solani* growth. The antagonistic behavior of *Trichoderma* and *Gliocladium* species resulted from growth of antagonistic fungi over all psyllium pathogens.

In pot experiment, the combined soil infestation with *G. virens* and *F. solani* recovered the highest percent of survival plants (75%) and maximized the plant height, number of leaves, spikes, fresh and dry weights plant^{-1}. In general, the presence of antagonistic fungi reduces the deleterious effect of wilt causative pathogens on growth parameters of psyllium plants.

Keywords: First record, *F. incarnatum*, *F. oxysporum*, *F. solani*, *R. solani* and *S. sclerotiorum*, *Plantago ovata*, antagonism, wilt, root rot.

INTRODUCTION

Plantago ovata Forsk (blond psyllium seed) belongs to the family *Plantaginaceae*. The suggested native for these species is Western Asia, but it was also found in the Arabian Peninsula and entered into India during Muslim settlement in middle ages (Morton, 1977). As a commercial crop, *P. ovata* is a winter annual in lower desert areas where temperatures rarely fall below -6.0^{o}C. Psyllium plants are grass-like in appearance, but are really classed as stemless herbs. Leaves are in rosettes, and seed scapes are produced which grow to a height of 20-30 cm (Russell, 1975).

P. ovata is produced commercially in several European countries, former Soviet Union, Pakistan, and India. The later dominates the world market in the production and export of psyllium (Hanson *et al*., 1992).

The economical value of these plants resulted from their possibility to be used as a successful plantation in desert regions. Moreover, their yield of

seeds and husks has been required for pharmacological purposes which they are considered mild and safe laxative particularly in cases of habitual constipation, chronic diarrhea and dysentery. Numerous studies confirm psyllium can lower total cholsterol levels (Bell *et al*., 1990 and Tunley *et al*., 1991) and reduction of the postprandial blood sugar increase. Chiej (1988) added that such seeds are used as cosmetically in face-masks to soften the skin because of its high mucilage content. Psyllium mucilage is also used as a natural dietary fiber for animals. The dehusked seed that remains after the seed coat is milled off is rich in starch and fatty acids and incorporated into breakfast cereals, ice cream, instant beverages, bakery products, and dietary products (Kalyanasundaram *et al*. 1984 and Chan & Wypiszyk 1988)**.** Also, is used in India as chicken and cattle feeds (Chevallier, 1996).

Unfortunately, losses due to the wilt-type disease have bean observed in growing areas in Egypt. There are reports of *Plantago ovata* wilt disease in USA (Russell, 1975) caused by *Fusarium oxysporum* grown in arid regions. Wilt symptoms were reported by Naresh-Mehta *et al*. (1985) in India caused by both *Fusarium oxysporum* and *F. solani*. Also, *Pythium ultimum* was reported as causative pathogen of both pre- and post-emergence damping-off in India (Rathore, 2000**)**. Symptoms of the disease were often clearly evident at the beginning of the flowering plant stage (60 days of sowing). Pre-emergence damping-off or a rapid wilting of plants including yellowing, stunting and dried shoots, as well as vascular discoloration and a cortical root rot were also observed during two successive winter seasons (2005/2006 & 2006/2007). As for the writers are aware, no study has been carried out on wilt disease in Egypt and very little knowledge are available all over the world.

The aim of this work, therefore, was to investigate disease occurrence, identify the causal pathogens, and determine the fungal pathogenic capabilities and screening some antagonistic fungi for controlling wilt and root rot diseases under greenhouse conditions.

MATERIALS AND METHODS

1. Psyllium seeds and antagonistic fungi

The planted seeds were Müggenburg Firm, Hamburg, Germany Sourced. As the seeds are very small, light and average weight of 1000 seeds is about 1.50 g, they were drilled using a small manual driller at shallow depth of 0.5 cm.

Isolates of *Trichoderma viride* (No. SR 25)*, T. harzianum* (No. SR 78), *T. hamatum* (No. SR 15)*, Gliocladium virens* (No. MS 34) and *G. rosum* (No. MS 16) were obtained from Biological Central Dept., Plant Pathol. Res. Instit., Agric. Res. Center (ARC), Giza, Egypt.

2. Natural incidence of wilt and root rot diseases:

Plants showing wilt symptoms *i.e*., yellowing, stunting, dried shoots and root rot were recorded and the percentages were calculated during two successive winter seasons (2005/2006 and 2006/2007)). The fields of Tag-Ezz, Agric. Res. station, Agric. Res. Center and Fac. Agric., Mansoura Univ. (Dakahlia governorate, Egypt) were surveyed.

3. Isolation of the pathogens:

Naturally infected psyllium samples showed typical symptoms of wilt and root rot diseases were collected. Plant roots were washed in running tap water followed by sterile water. Using sterilized scalpel, each part were cut into small pieces (1cm^2). Surfaces were disinfested by dipping pieces of the sample in 2% sodium hypocholorite (NaOCl) solution for 3 min., washed and dried between sterilized filter papers. The dried pieces were then transferred using sterilized forceps into Petri dishes containing potato dextrose agar medium (PDA) supplemented with antibacterial agent (L-Chloramphenicol; 5 mg/L and Streptomycin Sulphate 5 mg/L). The dishes were then incubated at 25°C and checked for fungal growth three days after planting. Purification of the resulting isolates was done using the single spore and /or hyphal tip technique. Pure cultures of the isolated fungi were obtained and all isolates were maintained on slants of Potato-Carrot Agar (PCA) for further studies. The detected fungi were identified in consultation with Commonwealth Mycological Institute (CMI) description sheets, and Raper & Fennel (1965), Booth (1971), Domsch *et al*. (1980) and Moubasher (1993**)**.

4. Pathogenicity test:

Fungal inocula were prepared by transferring single spore or hyphal tip of each fungus to Potato Dextrose Agar (PDA) media in a Petri-dishes and incubated for 4 days in the dark at 25±2 ºC. When hyphal growth reached a diameter of 3 cm, four disks (0.5 cm diameter) was removed from each culture and transferred to autoclaved 500-ml size bottles containing sand-cornmeal (SCM) medium (96% quartz sand + 4% cornmeal, water to 20% v/w), then incubated at 25±2ºC for 14 days. The inoculum of each fungus was added to sterilized plastic pots (14 cm diameter X20 cm height), containing 2 Kg soil at the rate of 1 g inoculum/1000g soil. Pots containing soil mixed with the same amount of noninfested medium served as check. All pots were kept in greenhouse for seven days while the temperature was ranged from 20 to 24ºC to allow fungi to adapt before sowing seeds. During this period, soil was kept moistened by watering when needs.

Healthy seeds of psyllium were soaked in 70% ethyl alcohol solution for three minutes then placed on sterilized tissue paper until dryness at room temperature. The treated seeds were planted in pots at rate of 10 seeds/ pot. Ten replicates (pots) were used for each treatment. Percentages of pre-and post-emergence damping-off were recorded, 14 and 90 days after planting, respectively. Routine isolation from plants grown in artificially infested soil resulted in the recovery of the fungi from the vascular tissues of roots.

5. Dual culture test:

Plates of PDA were inoculated with a 5 mm diameter disk from each of psyllium pathogens 10 mm from the edge of the plate. A 5 mm diameter disk of the antagonistic fungi being tested was placed 10 mm from the edge of the plate and opposite to each of the disks of pathogens. PDA plates inoculated with only 5 mm diameter disk of either pathogenic or antagonistic fungus 10 mm from the edge of the plates were used as control. Five replicates of each treatment were used. Plates incubated at 25±2°C under alternating luminosity (12 h light/ 12 h dark) for 6 days. The inward linear

growth was measured and the size of the inhibition zone and amount of overgrowth of each *Trichoderma* and *Gliocladium* species on pathogens were used to measure the interaction between the two fungal groups (Desai *et al*., 2002).

6. Greenhouse evaluation of the biological control agents against psyllium pathogens:

6.1. Preparation of inocula:

The inocula of antagonistic and pathogenic fungi were carried out using the same method as described in pathogenicity test. The inoculum of each fungus was added to sterilize plastic pots and classified as follow:
1) Soil infested individually with the either antagonistic or pathogenic fungi (positive check), 2) soil infested with both antagonistic and pathogenic fungi (1 g for each/1000 g soil), and 3) soil without any treatment (negative check).

6.2. Disease incidence and psyllium growth parameters of psyllium:

Data of disease incidence were recorded after 14 days of sowing for pre-emergence damping-off and after 90 days for post-emergence damping-off. At the end of the cultivation period (90 days), plant height (cm), root length (cm), number of leaves and number of spikes plant^{-1}, fresh weight and dry weight plant^{-1} (g) were determined.

7. Statistical Analysis

Data were analyzed as complete randomized plot design with the statistical analysis software, CoStat (2005). All data were first subjected to analysis of variance (ANOVA). Means were compared using Duncan's multiple range test (Duncan, 1955) at P= 0.05.

RESULTS

1. Description of psyllium wilt and root rot symptoms

Symptoms of wilt and root rot diseases under field conditions began to appear in circular areas of wilted and dried plants of 40-60 days old. The disease is noticed either as a pre-emergence damping-off, or as a late season wilt of plants. They start to produce seed scapes followed by wilting, which first occurs in the outer leaves with changes in leaf color from green to silver. Roots of wilting plants always show a black discoloration that gradually progresses from the root tip towards the crown (Plate, 1C).

A cortical root rot accompanies cortical discoloration and malformation with few or absence of lateral roots. Isolates were most easily obtained from surface-disinfested roots, and to a lesser extent from above-ground parts. In case of *S. sclerotiorum* infection (as detected later), abundant white fungal mycelium covered the plants foliage were noticed in field. Mycelium and sclerotia usually appear on the lower surface of the outer leaves, but under moist conditions the fungus invades the plant completely and causes it to rot, producing a white, fluffy, mycelial growth over the entire plant (Plate, 1A and B).

2. Screening of fungi associated with psyllium wilt symptoms

Different frequency percentages of many parasitic and saprophytic fungal species were found to be associated with the infected psyllium plants

(Table 1). Among *Fusarium* genus, *Fusarium solani* recorded the highest frequency (22.6%). Moreover, *Rhizoctonia solani* and *Sclerotina sclerotiorum*, recorded 19.5 and 6.5%, respectively. On the other hand, the remaining saprophytic fungal species varied in their frequencies, where they ranged 3% for *Mucor* species up to 35% for *Stemphylium* species.

Table 1: Fungi associated with infected psyllium root samples.

Fungus	Occurrence %
Acremonium sp.	6.50 g
Alternaria alternate	29.00 b
Cladosporium sp.	9.70 f
Epicoccum sp.	6.50 g
Fusarium incarnatum	12.90 e
Fusarium oxysporum	19.50 d
Fusarium solani	22.58 c
Mucor sp.	3.00 h
Rhizoctonia solani	19.50 d
Sclerotina sclerotiorum	6.50 g
Stemphylium sp.	35.50 a
Verticillium sp.	6.50 g

Values of means within a column followed by the same letter are not significantly different at *P*=0.05.

3. Pathogenicity test of important soilborne fungi on psyllium

Data in Figure (1) reveal that soil infested with pathogenic fungi showed maximum equal significant effect on pre-emergence damping off compared with the other treatments. Pre-emergence damping-off was observed as rotted seeds which were covered by mycelium and spores of the tested pathogens. *F. incarnatum*, *F. oxysporum* and *F. solani* were found to be more significant in post-emergence damping off (19, 26 and 17%, respectively) and survivals (16, 17 and 21%, respectively) compared with the other pathogenic fungi.

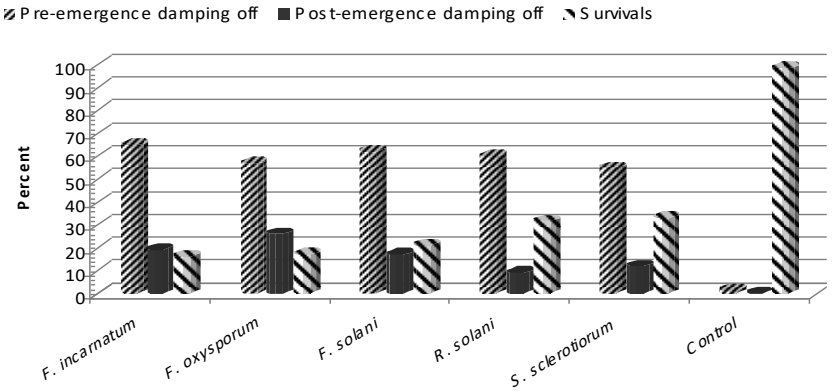

Fig. (1): Pre- and post-emergence damping-off and survivals of psyllium plants infected with different pathogenic fungi.

(L.S.D. at *P*=0.05 for pre-emergence=10.849; post-emergence=11.775 and survivals=12.883).

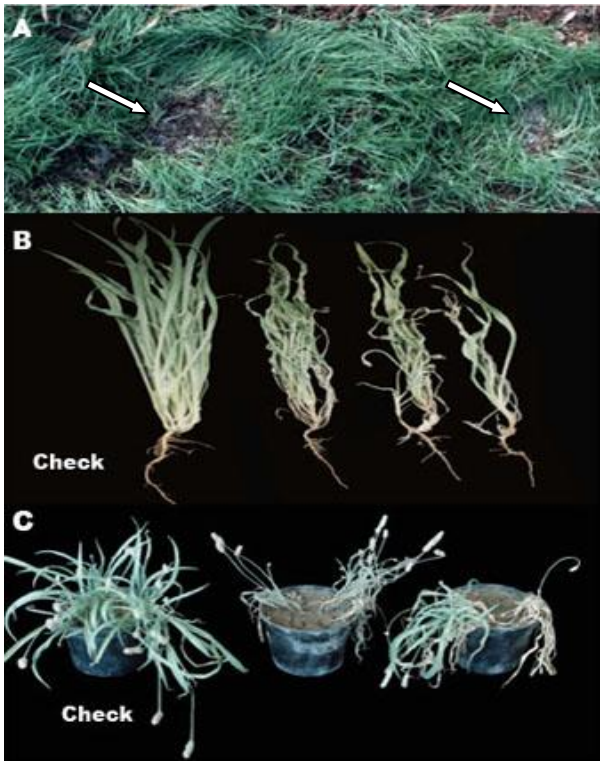

Plate (1): Naturally infected blond psyllium seedlings in field with *S. sclerotiorum* showing abundant white cottony mycellium covered the plants foliage (arrows)(A), Sclerotinia mold on psyllium plants (B) and Ninety days old blonde psyllium plants showing wilt symptoms due to infested soil with *Fusarium* spp. (C).

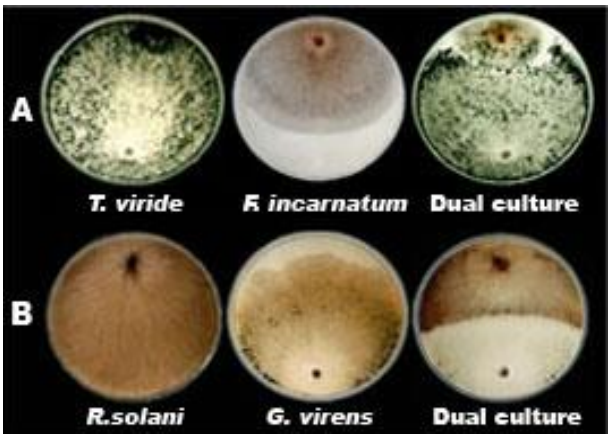

Plate (2): The antagonistic effect of *T. viride* against *F. incarnatum* (A) and *G. virens* against *R. solani* (B) after 6 days .

Infected seedlings showed lesions on lower stems near the soil surface and thread like roots of typical damping-off symptoms. Also wilt accompanied with yellowing, discoloration of the internal tissues, stunting and dried shoots were rated on seedlings (Plate, 1C).

4. Influence of antagonistic fungi on radial growth of wilt causative psyllium pathogens.

All antagonistic fungi reduced significantly the radial growth of pathogens (Table, 2). *Trichoderma viride* and *T. harzianum* showed the maximum significant reduction in radial growth of *F. incarnatum* (86.7 and 88%, respectively) **(**Plate, 2A), *F. oxysporum* (92.3 and 90.7%, respectively) and *S. sclerotiorum* (92 and 95%, respectively). *G. virens* came in the next order and recorded 81.7, 80.7 and 85.7% reduction for *F. incarnatum*, *F. oxysporum* and *S. sclerotiorum*, respectively. The radial growth of *F. solani* was highly affected by both *T. harzianum* and *G. virens* with 86.7 and 87.3%, reduction, respectively. *G. virens*, followed by *T. viride* and *T. harzianum* exhibited the highest reduction of *Rhizoctonia solani* growth *i.e*., 88.3%, 80 and 77.3%, respectively (Plate, 2B). On the other hand, *T. hamatum* and *G. rosum* showed the lowest significant reduction of mycelial growth of all tested pathogens.

5. Impact of soil infestation with antagonistic fungi on the incidence of psyllium diseases.

There is no any significant difference in wilt incidence among psyllium plants cultivated in soil infested individually with each of antagonistic fungi (Table, 3). The combined infestation with *G. virens* and *F. solani* into the soil recovered the highest percent of survival plants (75%) and the lowest percent of pre-emergence (23%) and post-emergence (2%) damping-off. Soil infestation with *T. viride* or *T. harizanum* in combined with *S. sclerotiorum* recorded the second high survivals of psyllium plants (*ca*, 56%). It is of especial important to note that, there is no significant effect on survival plants when soils infested with *T. harizanum* or *T. viride* in combined with any of the tested pathogenic fungi.

Table 2: Antagonistic effect of *Trichoderma* and *Gliocladium* species against psyllium wilt pathogens.

Pathogen / Antagonist	Inhibition %[1]				
	F. incarnatum	***F. oxysporum***	***F. solani***	***R. solani***	***S. sclerotiorum***
T. viride	86.67 a[2]	92.30 a	78.67 b	80.00 b	92.00 a
T. harzianum	88.00 a	90.67 a	86.67 a	77.30 b	95.30 a
T. hamatum	66.67 c	62.67 c	52.67 c	67.30 c	49.30 c
G. virens	81.67 b	80.67 b	87.30 a	88.30 a	85.67 b
G. rosum	52.00 d	64.30 c	48.30 c	48.30 d	34.30 d
Check	0.00 e	0.00 e	0.00 e	0.00 e	0.00 e

[1] Inhibition % represents the distance between disk's center and the margin of the colony.
[2]Each value represents the mean of 5 replicates, values within a column followed by the same letter are not significantly different (*P*=0.05).

Table 3: Effect of antagonistic fungi on disease incidence of psyllium wilt under greenhouse.

Treatment			Damping-off (%)		Survivals (%)
			Pre-emergence	Post-emergence	
Antagonist	*T. viride*		11.0 e[1]	5.8 g-j	83.2 ab
	T. harzianum		10.0 e	4.0 h-j	86.0 ab
	G. virens		10.0 e	6.0 f-j	84.0 ab
Combination	*T. viride*	*F. incarnatum*	40.0 ab	18.2 b-d	41.8 c-e
		F. oxysporum	48.2 a	12.0 d-h	39.8 de
		F. solani	33.0 b-d	28.2 a	38.8 e
		R. solani	47.5 a	15.0 c-g	37.5 e
		S. sclerotiorum	30.0 b-d	13.6 c-h	56.4 c
	T. harzianum	*F. incarnatum*	32.0 b-d	25.0 ab	43.0 c-e
		F. oxysporum	31.8 b-d	13.2 c-h	55.0 cd
		F. solani	40.0 ab	13.2 c-h	46.4 c-e
		R. solani	25.0 cd	22.4 a-c	52.6 c-e
		S. sclerotiorum	36.2 bc	7.5 e-j	56.3 c
	G. virens	*F. incarnatum*	34.0 b-d	10.0 d-j	56.0 c
		F. oxysporum	38.2 ab	10.0 d-i	51.8 c-e
		F. solani	23.0 d	2.0 i-j	75.0 b
		R. solani	33.0 b-d	17.0 b-e	50.0 c-e
		S. sclerotiorum	37.5 ab	16.5 b-f	46.0 c-e
Check			5.4 e	0.0 j	94.6 a

[1]Each value represents the mean of 10 replicates, values within a column followed by the same letter(s) are not significantly different (*P*=0.05).

6. Effect of antagonistic fungi on growth parameters of psyllium plants recovered from wilt pathogens infested soil.

Data in Table (4) show that the infestation of pathogenic fungi alone into the soil markably decreased all growth parameters of psyllium plants. In contrast, the sole infestation of antagonistic fungi led to obvious improvement in the studied parameters.

The combined infestations with pathogenic and antagonistic fungi show various degrees of effects. These effects were found to be better than or worth than those of control *i.e*., *G. virnes+R. solani* recorded the highest plant height (16.13 cm), number of leaves (19.13), number of spikes (6.63), fresh weight (2 g) and dry weight (0.72 g). On the other hand, the combined infestation *T. viride+F. solani* recorded the lowest values of psyllium growth parameters *i.e*., plant height (11.25 cm), number of leaves (12.13), root length (3.75 cm), number of spikes (3.75), fresh weight (0.86 g) and dry weight plant^{-1} (0.29 g). Between the previous highest and lowest values of the growth parameters, the combined infestation with *G. virens+F. oxysporum* or *T. harizanum+F. incarnatum* recorded moderate values of the studied parameters. In general, the presence of antagonistic fungi reduces the deleterious effect of wilt causative pathogens on growth parameters of psyllium plants.

Table 4: Effect of antagonistic fungi on growth parameters of psyllium rising from soil infested with wilt pathogens.

Treatment			Plant height (cm)	Root length (cm)	No. of leaves plant^{-1}	No. of spikes plant^{-1}	Fresh weight (g)	Dry weight (g)
Pathogen	*F. incarnatum*		12.06	6.65	14.25	3.63	1.04	0.47
	F. oxysporum		12.34	6.96	14.38	4.13	1.02	0.39
	F. solani		12.41	7.23	15.63	3.88	1.10	0.28
	R. solani		12.34	6.39	14.00	3.88	1.07	0.32
	S. sclerotiorum		12.04	6.41	13.13	3.88	1.02	0.28
Antagonist	*T. viride*		13.15	5.44	17.63	5.50	1.66	0.49
	T. harzianum		11.73	6.31	15.88	4.13	1.22	0.39
	G. virens		12.86	7.06	15.75	4.00	1.24	0.34
Combination	*T. viride*	*F. incarnatum*	12.90	6.66	15.88	4.88	1.21	0.43
		F. oxysporum	12.83	7.06	15.13	4.75	1.32	0.42
		F. solani	11.25	3.75	12.13	3.75	0.86	0.29
		R. solani	12.45	6.50	15.75	5.13	1.16	0.40
		S. sclerotiorum	13.18	7.50	16.38	4.63	1.48	0.41
	T. harzianum	*F. incarnatum*	13.64	8.50	18.50	5.13	1.50	0.46
		F. oxysporum	12.09	6.94	14.63	4.00	1.09	0.27
		F. solani	12.84	6.89	17.13	4.75	1.48	0.44
		R. solani	14.15	6.83	13.75	3.5	1.29	0.38
		S. sclerotiorum	11.98	6.38	14.50	4.00	1.12	0.31
	G. virens	*F. incarnatum*	13.79	6.88	15.88	4.25	1.67	0.43
		F. oxysporum	13.96	7.13	18.13	5.75	1.62	0.43
		F. solani	11.75	6.38	12.50	3.88	0.95	0.30
		R. solani	16.13	7.31	19.13	6.63	2.00	0.72
		S. sclerotiorum	12.95	7.05	14.88	4.50	1.00	0.35
Check			13.45	6.38	14.75	4.38	1.15	0.38
L.S.D. at *P*=0.05			1.129	0.988	2.683	0.935	0.448	0.112

DISCUSSION

Symptoms of wilt and root rot diseases with serious effect on growth were noticed on psyllium plants during growing winter seasons of 2005/2006 and 2006/2007. From the isolation trials, five causal pathogenic fungi were obtained from infected roots and identified as *F. incarnatum*, *F. oxysporum*, *F. solani*, *R. solani* and *S. sclerotiorum*. Koch's postulates were applied in isolation and identification of the pathogens. The previous isolated fungi are well known as causative pathogens of many wilt and root rot diseases on several plants (Domsch *et al*., 1980 and Strange, 2003).

Soilborne fungi that cause disease are dispersed as spores or sclerotia, which remain in a quiescent state until the appearance of a host plant which stimulates germination (Katan, 1996). The initiation of infection depends on signals that induce spore germination (Deacon, 1996). Nutrients such as sugars and amino acids leaking from roots have been presumed to serve as nonspecific signals for germination because such compounds are present in root exudates and also can stimulate germination under controlled conditions (Lockwood, 1988).

The pathogenicity test confirmed that all five isolates are causative agents of wilt and root rot diseases causing pre- and post-emergence damping-off. *F. incarnatum* was the most aggressive pathogens in this respect. As far as we know, it is the first record for *F. oxysporum* and *F. solani* in Egypt and first record for *F. incarnatum*, *R. solani* and *S. sclerotiorum* as soilborne pathogens worldwide.

F. oxysporum and *F. solani* were previously recorded as wilt causative fungi on *Plantago ovata* plants in both USA and India **(**Russell, 1975 and Naresh-Mehta *et al*., 1985). Elwakil and Ghoneem (1999) recorded *F. incarnatum*, *F. oxysporum* and *F. solani* as seed-borne pathogens on psyllium seeds in Egypt.

The vascular wilt fungi *F. oxysporum* and *F. solani* are soilborne facultative parasites that cause economically important losses in a wide variety of crops. The pathogens survive as non-motile resting spores in the soil. After germination, the fungus contacts the root and colonizes its surface, growing and branching, forming a network of hyphae. The pathogen will penetrate epidermal walls of root directly and subsequently colonize the tissue by intra- and intercellular growth. That growth is accompanied with polygalacturonase activities, and polygalacturonate lyase activities, the later is correlated with virulence, then grow through or between the root cells to reach the vascular system of the root (Mendgen *et al*., 1996).

Sclerotinia sclerotiorum, the causative agent of stem rot or white mold, is a non-host-specific, necrotizing pathogen that attacks a wide range of cultivated and wild plant species including canola (oilseed rape), mustard, alfalfa, soybean, field-bean, lentil, field pea, and sunflower resulting in crop lodging and severe yield losses (Li *et al*., 2004). The fungus deploys two main pathogenicity determinants, the secretion of oxalic acid and hydrolytic enzymes. Oxalic acid exerts a toxic effect on the host tissue by acidifying the immediate environment and by sequestering calcium in the middle lamellae leading to loss of plant tissue integrity (Godoy *et al*., 1990).

Rhizoctonia solani is distributed worldwide, exhibits physiologic strains, wide host range causing different symptoms on the same host depending on the time of infection. Since the pathogen has a propensity for attacking juvenile tissues, bedding plants grown from seed are especially vulnerable to pre-emergence damping-off (Lewis and Lumsden 2001).

Results of dual culture reveled that, *T. viride* and *T. harizanum* showed the maximum significant reduction in growth of *F. incarnatum*, *F. oxysporum* and *S. sclerotiorum*, while *G. virens* was more effective on the growth of *R. solani*.

Members of the genus *Trichoderma* and *Gliocladium* are potent mycoparasites against a number of plant pathogenic fungi and therefore are used as biocontrol agents as a promising alternative to chemical pesticides (Harman & Bjorkman, 1998 and Hjeljord & Tronsmo, 1998). It has been known for many years that *Trichoderma* spp. inhibit the fungal growth by three mechanisms: competition (for space and nutrients), parasitism (deriving nutrients from the host); and antibiosis (production of an inhibitory metabolite or antibiotic) (Harman, 2006). While one mechanism may predominate, this does not exclude the possibility that one or both of the other two mechanisms may also play a role in the antagonistic behavior. It may occur as an independent phenomenon or in combination with other mechanisms (Benitez *et al*., 2004).

Since overgrowth is very clear on plates of dual culture test (Plate, 2), we suggest the predominant of mycoparasitism as a mechanism for reduction in growth of pathogenic fungi. As mycoparasitism by *Trichoderma* requires the degradation of the host's cell wall and as chitin together with β-(1,3)-glucan is the major constituent of the fungal cell walls, hydrolytic enzymes, particularly glucanases and chitinases, play a major role in this process (Reithner *et al*., 2005). In addition to amylase, it has been reported that *Trichoderma* species also produce extracellular cellulase and pectinase that are capable of hydrolyzing the cell walls of other fungi (Marco *et al*., 2003). The concept of enzyme biosynthesis as a mechanism of biocontrol has been expanded to include synergism between enzymes and antibiotics **(**Schirmbock *et al*., 1994).

In pot experiment, the application of *G. virens* with *F. solani* as soil infestation minimized the percentage of infected plants. It is well known that *F. incarnatum*, *F. oxysporum*, *F. solani*, *R. solani* and *S. sclerotiorum* are active fungi in producing cellulalytic, pectinolytic and chytinolytic enzymes. These enzymes are responsible for appearance and development of pre- and post-emergence damping off disease symptoms, this may be explained by softening of the outer plant tissues (especially roots), so, help in easy penetration by the pathogen into plant (Egmond and de Vlieg, 2000; Garcia-Maceira *et al*., 2001 and Li *et al*., 2004). The reduction in pre- and post-emergence damping off in the presence of *G. virens* may explained by the work of Shiomi *et al*. (2000) who isolated a new chitinase inhibitor enzyme; Argifin from the culture fluid of the biocontrol fungus *Gliocladium* sp. FTD-0668, which inhibit the action of the previous chitinase enzymes on plant tissues.

The results of growth parameters attained statistical significance between presence and absence of antagonistic fungi. The improvement in growth parameters of the infected psyllium plants in the presence of antagonistic fungi is expected, since these antagonists fungi stopped or minimized the deleterious action of the pathogenic ones. Recent researches suggest that *Trichoderma* spp. can also induce systemic and localized resistance as well as directly attacking or inhibiting the growth of plant pathogens (Lo *et al*., 2000 and Harman *et al*., 2004). In addition, certain *Trichoderma* strains have substantial influence on plant growth and development (Hedge & Hofreiter, 1962 and Abdel-Fattah *et al*., 2007) by production of growth-regulating factors which increased the rate of seed germination and dry weight of shoots and stems (Windham *et al*., 1986)**.**

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أول تسجيل لمرضى عفن الجذور و الذبول على نباتات بذر القطونة فى مصر

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سُجل مرضى عفن الجذور و الذبول لأول مرة في مصر خلال موسمى النمو 2005/2006 و 2006/2007 وذلك أثناء حصر لزراعات نبات بذر القطونة, والذي يُعد أحد النباتات الطبية ذات العائد الاقتصادى. وقد اشتملت الأعراض في المناطق المصابة على حدوث اصفرار و تقزم و جفاف للمجموع الخضري بالإضافة إلى تلون أوعية الخشب و كذلك القشرة الخارجية للمجموع الجذري.

عُزلت فطريات *Fusarium incarnatum* و *F. oxysporum* و *F. solani* و *Rhizoctonia solani* و *Sclerotinia sclerotiorum* من النباتات المصابة. وقد تبين من اختبارات العدوى الصناعية , أن جميع الفطريات المعزولة كانت متساوية فى قدرتها على إحداث موت للبادرات تحت سطح التربة, بينما كان *F. oxysporum* و *F. solani* هما الأكثر ضراوة في إحداث موت للبادرات فوق سطح التربة , في حين احتل *Rhizoctonia solani* و *Sclerotinia sclerotiorum* المرتبة الأخيرة.

باستخدام الفطريات المضادة *Trichoderma viride* و *T. harizanum* و *T. hamatum* و *Glicladium virens* و *G. rosum* , أظهر *Trichoderma viride* و *T. harizanum* أعلى تأثير مثبط لنمو *F. incarnatum* كما تأثر نمو *F. solani* بكل من *T. harizanum* و *G. virens*. وقد كان *G. virens* الأعلى تثبيط لنمو *R. solani*. وبصفة عامة فقد كان التاثير التثبيطى يرجع إلي علاقة فرط التطفل .

في اختبارات تجارب الأصص, وجد أن إضافة فطر *G. virens* الي التربة المعداه بفطر *F. solani* أدى إلي زيادة عدد النباتات الحية (٧٥%) , كما انعكس ذلك على زيادة طول النبات وعدد الأوراق والسنابل وكذلك الوزن الغض والجاف للنباتات الناتج . وعموما فان وجود الفطريات المضادة قلل من التأثير الضار لمسببات أمراض الذبول علي صفات النمو لنباتات بذر القطونة.