Assiut J. of Agric. Sci., 38 (2) (111-126)

ENHANCEMENT OF BIOCONTROL OF ONION WHITE ROT USING ORGANIC SULPHIDES AND PLANT GROWTH PROMOTERS

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Abstract: Nine isolates of *Sclerotium cepivorum* obtained from different localities of Upper Egypt were able to infect Giza 6 onion cultivar and produced typical symptoms of white rot. The pathogenic potentialities of the tested isolates ranged from moderate to severe. *Trichoderma viride* and *T. harzianum* were capable to antagonize growth of the tested *S. cepviroum* isolates in *vitro*. They showed different inhibitory effect against growth of the pathogen isolates on PDA medium ranged from moderate to severe. Under greenhouse conditions treated infested soil with the pathogen isolates No. 5 and 7 with both antagonists at transplanting time, significantly reduced percentages of infection and white rot severity on Giza 6 onion cultivar and numbers of the pathogen sclerotia in soil. The least disease severity % and the lowest numbers of sclerotia in soil were achieved by *T. viride*. Treated Giza 6 onion plants with combined treatments of the biocide (BioHealth) and the plant growth promoters (Algarin and Inicium) caused significant reduction in incidence of onion white rot in greenhouse and in field plots (naturally or artificially infested with pathogen isolates). Soil treatment with single or combined treatments of *T. viride* or *T. harzianum* and the tested organic sulphides one week before transplanting date caused significant reduction in both disease incidence and numbers of the pathogen sclerotia in infested soil with the pathogen isolates. The lowest disease severity % and numbers of the pathogen sclerotia in soil were achieved by combined treatments (organic sulphides + antagonists) compared with single treatment with each of them alone.

Key words: biocontrol, onion white rot, organic sulphides, growth promoters.

Introduction

White rot of onion incited by the soil inhabiting fungus *Sclerotium cepivorum* Berk. causes an important economical loss in the main production areas of winter onion (*Allium cepa* L.) in Egypt (Abd El-Razik *et al.*, 1973) and around the world (Peŕez *et al.* 1994 and Tyson *et al.*, 2000). The base of infected bulbs are covered with a fluffy white mycelium and a large numbers of small, spherical, black sclerotia. Sclerotia formed on the rotting tissues can persist, as a dormant state, in the soil for long periods (more than 20 years) in absence of *Allium* species. The sclerotia break

dormancy, germinate and produce mycelium as a specific response to root exudates including sulphoxides, produced by *Allium* spp. and to the breakdown products of these compounds (Coley-Smith *et al.*, 1987).

Until now, traditional methods for controlling the disease by crop rotation, resistant varieties and fungicides are economically inefective (Fullerton and Stewart, 1991; Agrios, 1995; McLean and Stewart, 2000; Metcalf and Napier, 2002; Amin, 2003 and Clarkson *et al.*, 2006). Therefore, there is an urgent need for new effective methods to control the disease. Biocontrol of the disease is still partially effective (Abd El-Moity and Shatla, 1981, McLean and Stewart, 2000; Payghami *et al.*, 2001; Metcalf and Napier, 2002; Clarkson *et al.*, 2002; Amin, 2003, Sallam, Nashwa, 2004 and Clarkson *et al.*, 2006). The aim of this study was enhancement of the traditional biocontrol of the disease by plant growth promoters or by stimulants of germination of the pathogen sclerotia in soil, which may have a key role in the concept of control strategy of the disease.

Materials and Methods

Isolation and identification of *S. cepivorum* isolates:

Nine isolates of *S. cepivorum* Berk., the incitant of onion white rot, used in the current study were isolated from naturally infected onion plants (cultivar, Giza 6) showing typical symptoms of white rot and collected from different localities in El-Minea, Assiut and Sohag Governorates of Upper Egypt.

Infected onion plants with white rot were cut into small pieces, thoroughly washed with tap water, surface sterilized by immersing for 2-3 minutes in 2% sodium hypochlorite solution, rinsed several times in sterilized distill water and dried with sterilize filter papers. The surface sterilized plant pieces were plated on sterilized Potato Dextrose Agar medium (PDA) in Petri plates and incubated at 20°C. After 4-5 days incubation period, the developed fungal colonies were purified by hyphal tip technique on PDA medium at 20°C. Fungal isolates were identified by using the morphological features of mycelia and sclerotia as described by Domsch *et al.* (1980).

Pathogenicity tests:

Pathogenicity tests of *S. cepviroum* isolates on Giza 6 onion cultivar were carried out under greenhouse conditions in the Faculty of Agriculture, Assiut University in 2004/2005 onion growing season.

Inocula of the nine isolates of the fungus were prepared by growing isolates on autoclaved Sand-Corn meal medium (20 k coarse sand, 1 k ground maize and 4 liter water) in 500 ml milk bottles and incubated at 20°C for 30 days. Sclerotia of the

fungus isolates were recovered from the medium by screening through 0.0165 inch sieve and mixed with autoclaved clay soil at the rate of 5 sclerotia/g soil. Autoclaved pots were filled with infested soil. Non-infested soil was used as control.

Seeds of Giza 6 onion cultivar were surface sterilized by dipping in 5% sodium hypochloride solution for 3 min. followed by washing with sterilized water and seeded in autoclaved soil. After 45 days from seeding, onion transplants were uprooted and transplanted in infested or non infested soil (5 plants/pot 25 cm in diameter). Five replicates were used for each tested isolate. Percentage of infection and disease severity percentage of each isolate were recorded after 4 months from transplanting date. The arbitrary (0-4) disease scale described by **Abd-El-Razik *et al.* (1974)** was used to measure the disease severity percentage.

Preliminary tests for antagonistic capability of two species of *Trichoderma* against *S. cepivorum* isolates:

Trichoderma viride and *T. harzianum* isolates used in this study were obtained from culture collection of the Department of Plant Pathology, Faculty of Agriculture, Assiut University. Isolates of *Trichoderma* spp. and *S. cepivorum* were grown on PDA medium at 25°C and 20°C, respectively for 6 days and used as inocula. Disks (5 mm in diameter) from each isolate of *Trichoderma* spp. were inoculated on PDA medium in one site of Petri plate. However, the opposite site was plated by equal disks of the tested *S. cepivorum* isolates. Five replicates were used for each particular test and inoculated plates with *S. cepivorum* isolates only were used as control. Within 5 days incubation period at 20°C, linear growth of the tested *S. cepivorum* isolates was recorded and reduction percent of growth was calculated.

Effect of *Trichoderma viride* and *T. harzianum* on incidence of onion white rot and numbers of the pathogen sclerotia in soil, under greenhouse conditions:

Inocula of *T. viride* and *T. harzianum* were prepared by growing the antagonists on autoclaved Potato Dextrose Broth (PDB) medium. After 10 days incubation period at 25°C, contents of flasks were centrifugated at 5.200 x g. for 5 minutes, the supernatants were decanted, and the precipitates were suspended in sterile water to give concentration of $6x10^6$ CFU/ml. However, inocula of *S. cepviroum* isolates No. 5 (highly pathogenic) and No. 7 (moderately pathogenic) were prepared and mixed with the soil as described in pathogenicity tests. Autoclaved pots (25 cm in diameter) filled with autoclaved clay soil were infested by the tested isolates of *S. cepivorum* at the rate of 3 sclerotia/g soil, just before

transplanting. *Trichoderma viride* and *T. harzianum* inocula were applied to infested soil with the pathogen, at time of transplanting, at the rate of 50 ml from the previously prepared inoculum suspension/pot. Forty-five days old onion Giza 6 transplants were transplanted in the treated pots at the rate of 5 transplants/pot. Five replicates were used for each particular treatment and untreated soil with the tested bioagents was used as control.

After 4 months from transplanting date, disease severity % was recorded. The wet-screening technique described by **Adams (1979)** for recovery of sclerotia of *S. cepivorum* from the soil was used to assay sclerotia in soil samples collected, just after harvest, from each tested treatment. Size of each soil sample was equivalent in weight to 100 g of oven dry soil. Wet sieving weighted amounts of soil samples were entails on a 0.18 mm sieve and residues were examined microscopically to count sclerotia of the pathogen in the sample. Three subsamples are assayed for each treatment.

Effect of BioHealth® WSG and certain plant growth promoters in controlling white rot of onion:

BioHalth® WSG, composed of a selected *Trichoderma harzianum* strain (approx. 10%), Humic acids (approx. 75%), Seaweed extract (approx. 5%) and water (approx. 10%). It is manufactured by Humin® Tech Comp, Spain as a biocide formulation to soil born pathogens. Certain plant growth promoters Algarin (a mixture of organic algae powered on huomic acid) and Inicium (a mixture of amino acids 10% + P_2O_5, 6% w/v) were used in these experiments. They are produced by Bioiberica Company, Spain.

Efficiency of BioHealth and the tested plant growth promoters in the biocontrol of onion white rot was tested under greenhouse and field conditions. In greenhouse experiments, preparation of inocula of the tested isolates of the pathogen and soil infestation with the pathogen sclerotia were done as described in pathogenicity tests. However, in field tests, the experiment was carried out under natural infection in field plots (with back history of white rot) or artificial soil infestation with the tested isolates. The tested pathogen isolate inocula were placed in soil adjacent to onion transplants at time of transplanting. Uninfested and nontreated soil with BioHealth were used as control and 4 plots were used as replicates. The tested treatments were as following:

1- Dipping Giza 6 onion transplants in BioHealth (0.5 %) before transplanting followed by twice applications at the rate of $^1/_2$ kg biocide / feddan to irrigation water after 42 & 63 days from transplanting date. The growth promoters (Algarin $^1/_2$ kg and Inicium 1.5 L / feddan)

were applied to irrigation water after 21 & 63 days from transplanting date.

2- Dipping Giza 6 onion transplants in BioHealth (1 %) before transplanting followed by twice applications at the rate of $^1/_2$ kg biocide / feddan to irrigation water after 42 & 63 days from transplanting date. The growth promoters (Algarin $^1/_2$ kg and Inicium 1.5 L / feddan) were applied to irrigation water after 21 & 63 days from transplanting date.

3- The same treatment without BioHealth service as control.

Marketable bulb yield Kg / feddan was determined in every treatment after 4 months from transplanting.

Corn flower was mixed with the biocide solution 6 hrs before dipping of transplant at the rate of 1 and 2 % for treatment No. 1 and 2, respectively.

Effect of certain organic sulphides on incidence of onion white rot and biocontrol of the disease by *Trichoderma* spp.:

This experiment was carried out under greenhouse conditions in season 2006/2007. The pathogen isolates No. 5 and 7, the bioagents *T. viride* and *T. harzianum* as well as the two organic sulphides N-allyl sulphide and N-propyl sulphide were used in this study. The procedures previously mentioned for preparing pathogen and bioagent inocula in pathogenicity tests and biocontrol experiments were adopted. The autoclaved clay soil in autoclaved pots (30 cm in diameter) was infested by the tested pathogen isolates at the rate of 3 sclerotia/g soil. The infested pots were divided into nine treatments {(*T. viride* (TV), *T. harzianum* (TH), N-allyl sulphide (AS), N-propyl sulphide (PS), TV + AS, TV + PS, TH + AS, TH + PS and Untreated (control)}. Five replicates were used for each treatment Infested soil was treated with the tested sulphide compounds at the concentration 200 ppm, (300 ml/pot) and bioagents inocula ($6x10^6$ CFU, 50 ml/pot) 7 days before transplanting with 45 days old Giza 6 onion seedlings (7 transplants/pot). After 4 months from transplanting date, percentage of infection and disease severity were recorded and soil samples were collected for assaying numbers of sclerotia in soil.

Results

Pathogenicity tests

Results presented in Table (1) indicate that, the tested isolates of *S. cepivorum* were able to infect onion plants with different degrees of infection and disease severity causing white rot disease. Isolates No. 1, 3, 4, and 5 caused the highest percentage of infection and disease severity followed by isolates No. 2, 6 and 9 and finally isolates No. 7 and 8.

Table(1): Pathogenicity tests of *S. cepivorum* isolates on Giza 6 onion cultivar:

Isolate No.	Infection %	Disease severity %
1	100.0	78.0
2	96.0	68.0
3	96.0	72.0
4	100.0	79.0
5	100.0	90.3
6	92.0	68.0
7	71.9	63.5
8	76.4	68.8
9	78.3	74.2
Control	0.0	0.0
L.S.D. at 5 % :	11.8	20.3

Biological control of onion white rot:

A-Preliminary tests for antagonistic capability of two species of *Trichoderma* against growth of *S. cepviroum* in *vitro*:

Data in Table (2) indicate that the tested antagonists inhibited similarly growth of *S. cepivorum* on PDA medium. *T. viride* and *T. harzianum* caused 52.4% and 51.9% of inhibition to the pathogen growth, respectively.

Table(2): Antagonistic effect of two species of *Trichoderma* against growth of *S. cepivorum* isolates, in *vitro*:

Isolate No.	Antagonists		$\overline{X}$
	T. viride	*T. harzianum*	
1	57.4*	47.4	52.4
2	53.7	56.3	55.0
3	48.5	62.2	55.4
4	51.1	60.0	55.6
5	53.7	48.1	50.9
6	46.3	45.5	45.9
7	55.6	49.3	52.4
8	54.1	45.5	49.8
9	50.8	52.6	51.7
$\overline{X}$	52.4	51.9	-

* Inhibition % in growth of *S. cepivorum*

L.S.D.at 5%

Antagonists (A)	:	2.1
Pathogen isolates (B)	:	4.5
(A) x (B)	:	6.4

The highest inhibitory effect to growth of the tested isolates of the pathogen was displayed by *T. viride* to isolate No. 1 (57.4%) and *T. harzianum* to isolates No. 2 (56.3%), 3 (62.2%) and 4 (60%). However, the lowest inhibitory effect was obtained by *T. viride* to the pathogen isolates No. 3, 4, 6 and 9 (46.3-51.1%) and *T. harzianum* to isolates No. 1, 5, 6, 7 and 8 (45.5 - 49.3%).

B -Effect of *T. viride* and *T. harzianum* on incidence of onion white rot and numbers of *S. cepivorum* sclerotia in soil:

Data in Table (3) indicate that application of *T. viride* and *T. harzianum* to infested soil with the tested pathogen isolates at time of transplanting, have significantly reduced disease severity percent of white rot on Giza 6 onion cultivar. The least percentage of disease severity was obtained by application of the tested antagonists to soil infested with the pathogen isolate No. 5. However, *T. viride* antagonist caused the least percentage of disease severity with the pathogen isolate No. 7. Data also indicate that the pathogenic capability of the tested isolates of the pathogen was approximately similar.

Data also indicate that treated infested soil with *S. cepviroum* isolates (No. 5 and No. 7) and the tested antagonists (*T. viride* and *T. harzianum*) with Giza 6 onion cultivar reduced significantly numbers of the pathogen sclerotia in soil compared with untreated soil (control). *T. viride* caused the least numbers of sclerotia in soil formed by the tested isolates of the pathogen followed by *T. harzianum*.

Data also indicate that isolate No. 7 of the pathogen produced higher numbers of sclerotia in soil than isolate No. 5.

Table(3): Effect of *T. viride* and *T. harzianum* on incidence of onion white rot and numbers of *S. cepivorum* sclerotia in soil, under greenhouse conditions:

Antagonists	Disease severity %			No. of sclerotia/100g soil		
	Pathogen isolates			Pathogen isolates		
	No. 5	No. 7	$\bar{X}$	No. 5	No. 7	$\bar{X}$
T. viride	24.6	44.3	34.47	81.3	105.6	93.5
T. harzianum	30.6	50.6	40.62	106.3	131.0	118.7
Control	82.6	75.12	78.86	325.3	471.7	398.5
$\bar{X}$	45.96	56.67	-	171.0	236.1	-

L.S.D. at	**5%**	
Pathogen isolates (A)	12.3	26.4
Antagonists (B)	15.1	32.3
A x B	21.4	45.6

C-Effect of the biocide BioHealth and certain plant growth promoters on incidence of onion white rot under greenhouse and field conditions:

Data in Table (4) indicate that, under greenhouse conditions, dipping Giza 6 onion transplants for 30 minutes before transplanting in the tested solutions of BioHealth (0.5 and 1%) followed by twice applications of the biocide (at rate $^1/_2$ kg/feddan) and the growth promoters ($^1/_2$ kg Algerin/feddan + 1.5 L Inicium/ feddan) to irrigation water caused significant reduction in percentage of disease severity compared with the control (0 level) of BioHealth. However, insignificant differences were existed among disease severity of plants treated by 0.5 or 1% rates of BioHealth and infected with the tested pathogen isolates.

Data presented in Table (5) indicate that, under field conditions, the tested BioHealth treatments significantly reduced percentage of infection by white rot and increased marketable bulb yield of onion in both naturally and artificially infested field plots with mixture of *S. cepivorum* isolates (No. 5 and No. 7). The lowest percentage of infection with the disease and the highest marketable bulb yield were obtained by application of BioHealth at the rate of 1% in both naturally and artificially infested field plots. Data also indicate that the disease caused a drastic reduction on marketable bulb yield of onion Giza 6 grown in naturally and artificially infested soil with the pathogen isolates.

Table(4): Effect of BioHealth and certain plant growth promoters (Algarin & Inicium) on incidence of onion white rot, in greenhouse:

Treatments	Infection %			Disease severity %		
	Pathogen isolates			Pathogen isolates		
	No. 5	No. 7	$\overline{X}$	No. 5	No. 7	$\overline{X}$
Dipping in BioHealth (0.5%) followed by twice applications of the biocide + growth promoters to irrigation water	63.1	45.7	54.4	29.3	31.4	30.4
Dipping in BioHealth (1 %) followed by twice applications of the biocide + growth promoters to irrigation water	60.0	57.4	55.7	25.0	21.4	23.2
Untreated control (0 level)	82.1	86.9	84.5	64.6	75.1	69.9
$\overline{X}$	68.4	61.3		39.63	42.7	

L.S.D. at 5%

Pathogen isolates (A)	:	10.4	10.1
BioHealth conc. (B)	:	12.8	12.4
A x B	:	18.7	17.5

Table (5): Effect of BioHealth and certain plant growth promoters (Algarin & Inicium) on incidence of onion white rot and bulb yield in artificially and naturally infested soil with *S. cepivorum* under field conditions:

Treatments	Infection % in soil infested		Marketable bulb yield kg/feddan in soil infested	
	Artificially*	Naturally	Artificially*	Naturally
Dipping in BioHealth (0.5%) followed by twice applications of the biocide + growth promoters to irrigation water	90.2	19.0	552.5	7290.6
Dipping in BioHealth (1 %) followed by twice applications of the biocide + growth promoters to irrigation water	84.5	9.3	1566.4	8646.5
Untreated control (0 level)	96.5	20.5	362.1	7230.7
Uninfested soil	0	-	12544.0	-
L.S.D. at 5%	4.7	9.2	92.8	85.3

*Infection with mixture of isolates 5 and 7.

Effectiveness of certain germination stimulants of *S. cepivorum* sclerotia on incidence of onion white rot and biocontrol of the disease:

Data in Table (6) indicate that application of the tested organic sulphides and antagonists, in single or in combination, to infested soil with the pathogen isolates, one week before transplanting, reduced significantly disease severity % from 73.9 in control treatment to 11.5 - 40.6 in treated ones.

Application of the tested organic sulphides and antagonists together to infested soil with the pathogen caused the lowest percentage of disease severity compared with application of each one alone.

Data also show that, in general, application of the tested organic sulphides and antagonists in single or in combination one week before transplanting date caused significant decrease in numbers of sclerotia in infested soil with the pathogen isolates (from 388.5 sclerotia/100 g soil in control treatment to 31.5 - 118.3 sclerotia/100 g soil in treated ones). The lowest numbers of sclerotia in soil was found in infested soil treated with both tested organic sulphides and bioagents without significant differences among treatments. Data also indicate that, isolate No. 7 of the pathogen formed the higher numbers of sclerotia in soil (128.0 sclerotia/100g soil) than that formed by isolate No. 5 (89.7 sclerotia/100g soil).

Table(6): Effect of soil treatment with N-allyl sulphide, N-propyl sulphide and the bioagents *T. viride* and *T. harzianum*, one week before transplanting, on incidence of onion white rot and number of *Sclerotium cepivorum* sclerotia in soil, under greenhouse conditions:

Treatments	Disease severity %			No. of sclerotia/100 g soil		
	Pathogen isolates			Pathogen isolates		
	No. 5	No. 7	$\overline{X}$	No. 5	No. 7	$\overline{X}$
T. viride (TV)	24.6	40.7	32.7	81.3	105.7	93.5
T. harzianum (TH)	30.6	50.6	40.6	106.3	130.3	118.3
N-allyl sulphide (AS)	20.7	35.7	28.2	105.7	58.2	82.0
N-propyl sulphide (PS)	24.8	17.9	21.4	56.7	130.0	93.4
TV + AS	2.9	25.0	13.9	32.3	57.0	44.7
TV + PS	8.8	16.4	12.6	31.3	31.7	31.5
TH + AS	17.3	5.7	11.5	32.7	32.3	32.5
TH + PS	20.7	6.4	13.6	32.0	55.7	43.8
Untreated (control)	78.6	69.1	73.9	325.3	451.7	388.5
$\overline{X}$	25.4	29.7	-	89.7	128.0	-

L.S.D. at 5%

Pathogen isolates (A)	:	6.3	22.5
Treatments (B)	:	13.3	47.8
A x B	:	18.8	67.6

Discussion

White rot of onion incited by the soil inhabiting fungus *Sclerotium cepivorum* Berk. is one of the most important diseases affecting winter onion productivity in Egypt (Abd El-Razik *et al.*, 1973). Nine isolates of *S. cepivorum* were isolated from different localities in Upper Egypt. Pathogenic potentialities of the tested isolates, on Giza 6 onion cultivar, were different and ranged from moderate to severe. Such results are in agreement with those reported by Abd El-Razik *et al.*, 1974 and Sallam, Nashwa, 2004 and confirms the idea that *S. cepivorum* is a variable pathogen and consists of a heterogenous group of strains that might vary in morphology,

virulence, physiology and genetic diversity (Sallam, Nashwa, 2004).

Testing in *vitro*, antagonistic capability of *Trichoderma viride* and *T. harzianum* against *S. cepivorum* isolates showed similar inhibitory effect against growth of the pathogen. Tolerance of the tested pathogen isolates to the antagonistic effect of both antagonists was different and ranged from weak to moderate. Such results are in agreement with results of Abd El-Moity *et al.* (1982), Mousa *et al.* (1987), Hassan (1992), Amin (2003) and Sallam, Nashwa (2004).

In greenhouse experiment, application of *T. viride* and *T. harzianum* to infested soil with *S. cepviroum* isolates No. 5 and No. 7 at transplanting time, significantly reduced percentage of disease severity of white rot on Giza 6 onion cultivar and reduced numbers of the pathogen sclerotia in soil compared with untreated treatment. In general, *T. viride* showed the greatest reduction in disease incidence and the least numbers of the pathogen sclerotia in soil followed by *T. harzianum*. Such results are in accordance with those reported by Abd El-Moity and Shatla (1978), Mohamed and Fahmy (1988), Hassan (1992), Payghami *et al.* (2001), Clarkson *et al.* (2002 and 2004), Sallam, Naswha (2004), Clarkson *et al.* (2006), Coventry *et al.* (2006) and Garcia *et al.* (2006). Elad (1986) suggested that the antagonistic microorganisms can operate in biocontrol of plant diseases by rapid colonization in advance of the pathogen or subsequent competition or combat may lead to niche exclusion. Antibiotics may also produced or they may be mycoparasitis or lysis of the pathogen. In addition, some microorganisms may act simply making the plant grow better, so that even the disease is not cured its symptoms at least partly masked.

Dipping onion transplants in the tested biocide (BioHalth® WSG) before transplanting and plant growth promoters which mixed with irrigation water significantly reduced infection percent with white rot in greenhouse and field plots (naturally or artificially infested with pathogen isolates No. 5 and No. 7). Raising the tested rate of the bioside from 0.5% up to 1% had no effect on infection % of white rot in greenhouse, however, caused the highest decrease in infection % and the highest marketable bulb yield in the field. Such results are in line with those reported by Sallam, Nashwa (2004), McLean *et al.* (2005) and Garcia *et al.* (2006) who enhanced the biocontrol of onion white rot by applying the effective bioagents, to infested soil with the pathogen, in formulation forms. The detectable increase in marketable bulb yield may be due to the effect of the applied biocide and the tested

growth promoters on the pathogen and plant growth, respectively.

Efficacy of germination stimulants of sclerotia of *S. cepivorum* (certain organic sulphides possessed by *Allium* spp.) for management of white rot of onion and garlic have been studied by Coley-Smith and Parfitt (1986), Abd El-Razik *et al.* (1988), Tyson *et al.* (2000) and Davis *et al.* (2007). They showed that applying organic sulphides to infested soil with pathogen sclerotia in absence of an *Allium* crop cause death of the sclerotia of the pathogen after they germinate and exhaust nutrient reserves, as well as subsequently reduce population of sclerotia in soil and white rot percentage.

Soil treatment with the tested organic sulphides alone or in combination with *T. viride* or *T. harzianum* (one week before transplanting) have significantly reduced percentages of disease severity of white rot and numbers of the pathogen sclerotia in infested soil (with isolates No. 5 and No. 7). The highest reduction in disease severity % and in numbers of sclerotia in soil was achieved by the combined treatments (organic sulphides + antagonists) compared with single treatments with each one alone. Such results are in line with results of Smolinska *et al.* (2005) who reported that integrated control of white rot with seed treatments with fungicides, *Trichoderma* spp. and plant materials including onion waste improved the control of the disease, biological and chemical properties of the soil and increased growth of onion plants.

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تحسين المقاومة الحيوية للعفن الأبيض فى البصل باستخدام مركبات الكبريت العضوية ومنشطات النمو النباتية

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يعتبر مرض العفن الأبيض فى البصل المتسبب عن الفطر *Sclerotium cepivorum* من أهم الأمراض الفطرية التى تصيب محصول البصل الشتوى فى مصر وفى عديد من دول العالم الأخرى مسببا خسائر كبيرة فى المحصول .

ونظرا لقلة فعالية استخدام الطرق التقليدية فى مقاومته (الدورة الزراعية – التشميس – غمر التربة بالمياه – المقاومة الحيوية – المقاومة الكيميائية) بجانب عدم توافر أصناف مقاومة للمرض حتى الآن ، فإن الأمر يستلزم البحث عن وسائل جديدة للمقاومة مع تحسين ما هو متاح منها حتى يمكن تقليل الخسائر الناتجة عن الإصابة . لذلك هدفت هذه الدراسة إلى تحسين المقاومة الحيوية للمرض باستخدام منشطات إنبات الأجسام الحجرية للفطر المسبب (ذات الدور الهام فى حياة الفطر المسبب ودورة المرض وإنتشاره) ومنشطات نمو النباتات بالإشتراك مع الكائنات المضادة للطفيل فى المقاومة الحيوية للمرض

تبين من اختبار القدرة المرضية لتسعة عزلات للفطر المسبب للمرض تم عزلها من حقول البصل المصابة بالوجه القبلى قدرتها على إحداث مرض العفن الأبيض على نباتات البصل صنف جيزه (٦) بدرجات مختلفة من الحدة المرضية تراوحت بين شديدة ومتوسطة.

بينت التجارب المعملية قدرة الفطرين *Trichoderma viride* و *T. harzianum* على التضاد مع عزلات الفطر المسبب للمرض بدرجات متباينة تراوحت بين شديدة وضعيفة ولقد اختلفت باختلاف عزلات الفطر الممرض .

أدت معاملة التربة الملوثة بعزلتى الفطر الممرض (رقم ٥ ، ٧) قبل الزراعة (تحت ظروف الصوبة) بكل من الفطرين *Trichoderma viride* و *T. harzianum* إلى خفض شدة الإصابة بالمرض على صنف البصل جيزه (٦) وكذلك أعداد الأجسام الحجرية للفطر الممرض فى التربة.

ولقد كانت كفاءة الفطر *Trichoderma viride* أعلى من الفطر *T. harzianum* فى خفض الإصابة بالمرض وخفض أعداد الأجسام الحجرية فى التربة .

أدت المعاملات المختبرة من المبيد الحيوى بيوهيلث تحت ظروف الصوبة والظروف الحقلية وتحت ظروف العدوى الصناعية (بعزلتى الفطر الممرض رقم ٥ ، ٧) والعدوى الطبيعية بغمر الشتلات ثم إضافة المواد المنشطة لنمو نباتات البصل إلى ماء الرى إلى خفض معنوى فى نسبة الإصابة بالمرض على الصنف جيزه ٦ فى الصوبة والحقل وزيادة معنوية فى محصول الأبصال الصالحة للتسويق مقارنة بالنباتات غير المعاملة فى الحقل.

ولقد كانت معاملة التربة المشتركة بمنشطات انبات الأجسام الحجرية للفطر الممرض وكل من الفطرين *Trichoderma viride* و *T. harzianum* قبل الشتل بأسبوع إلى احداث أقل نسبة اصابة بالمرض المتسبب عن عزلتى الفطر وأقل أعداد للأجسام الحجرية فى التربة مقارنة بالمعاملة المنفردة لكل منها .