Biological and Chemical Control of the Sudden Wilt Disease of Cantaloupe in Egypt

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Abstract: The possible biological control of the sudden wilt disease-associated fungi, *Fusarium solani, Macrophomina phaseolina, Pythium aphanidermatum*, and *Rhizoctonia solani*. All cucurbit hosts were infected with the causal organisms of sudden wilt. Capritop and Topsin M were the most effective fungicides in controlling the tested pathogens of sudden wilt disease. *Trichoderma* species was investigated. *In vitro* experiment revealed that all the tested *Trichoderma spp.* inhibited the growth of all pathogens under study to a limited extent. *T ressei* inhibited fungal growth significantly higher than the rest of isolates (*T. pseudokoningii* and *T. hamatum* or *T. viride* and *T. harizanum*). *F. solani* and *R. solani* were generally the most sensitive fungi to the tested *Trichoderma spp*., while, *P. aphanidermatum*, *M. cannonballus* and *M. phaseolina*, were less sensitive in this respect.
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1.Introduction

Melon (*Cucumis melo*) is considered one of the major summer and nili vegetable crops in commercial fields and under protected cultivation during winter in Egypt. It is considered a major source of essential nutrients such as vitamins, minerals, carbohydrates, antioxidants and anti-carcinogenic substances, which are important to human nutrition and health (**Joseph, 1994**). The cultivated area of melons during 2010 was 146211 feddans yielded about 8-10 tons / feddan of fruits.

Melon is liable to attack by several soil-borne fungal pathogens during different growth stages resulted in considerable losses in fruits yield. The most important diseases, however, are damping-off, Fusarium wilt (**Zitter *et al*., 1996**), Sudden wilt (**Cohen *et al*., 1996, Pivonia *et al*., 1997, Cohen *et al*., 2000**) and Monosporascus root rot/vine decline (**Uematsu *et al*., 1985, Osada and Sasahara 1996, Stravato *et al*., 2002**).

In Egypt, sudden wilt disease on melon has been frequently occurred causing severe losses during the growing seasons. Therefore, the present investigation aims to study the disease occurrence, causal pathogen, and pathogenic capabilities of the isolated fungi, and different means of control management.

2.Materials and Methods

Isolations:

Isolation was made from twenty plants per field. Five fields represented each location showing disease symptoms. The diseased plants were uprooted then the roots were washed under running tap water to remove the soil particles. The tap and lateral roots were excised into small pieces (0.5-0.8 cm). The root pieces were disinfested in 0.5% sodium hypochlorite then 70% ethanol for two minutes each. Then, they were rinsed in sterilized distilled water and dried between two sterilized filter papers. The surface sterilized samples were plated onto Potato Dextrose Agar (PDA) medium and incubated at 25˚C until the recovery of the fungal colonies. The recovered fungi were microscopically examined, counted and the frequency of each fungus was determined.

Identification:

Purification of the isolated fungi was carried out using hyphal tip and/or single spore techniques (**Dhingra and Sinclair, 1985**). The developed fungal colonies were kindly identified by the staff members of the "Mycology and Plant Diseases Survey Department" in Plant Pathology Research Institute. They used the morphological and microscopically characteristics of the recovered fungi according to (**Barnett 1960**, **Pollack and Uecker 1974** and **Nelson *et al*., 1983**). The identified fungi were sub-cultured on PDA slants and kept at 5^{o}C for further studies.

Isolates of *Trichoderma* spp. isolated from the rhizosphere and rhizoplane of cantaloupe were identified after growing them on 20% malt extract agar then incubated for two days at 25°C according to (**Rifai, 1969 and Bissett, 1991**).

Pathogenicity Test:

Inoculum:

The fungi used in this study were grown on cornmeal-sand medium. A mixture of 2-5 g corn meal and 95-98 g fine sand previously washed was

transferred in a 250 ml glass bottle. Then each bottle received 50 ml of distilled water and plugged with a cotton stopper. The bottled medium was autoclaved at 121°C for 1 hr. The tested fungi were grown on PDA and incubated for 7 days at 25°C. A 4-mm disc of agar with mycelium from 7 days old culture of each fungus used in this study was transferred to the surface of the bottled medium. Five bottles were served for each fungus. All bottles were incubated at 25°C for 15 days and were daily shaken to spread the fungal inoculums through the medium.

Black plastic bags (30 cm in diameter each), served as pots, and were filled with a mixture of sterilized sand and clay (1:1, v/v). This mixture was mixed thoroughly with fresh fungal inocula of the individual isolates of the pathogens and/or with the different possible fungal combinations at the rate of 3% of soil (w/w) one week before sowing. The infested pots were watered and covered with plastic sheets under greenhouse condition for a week.

Ten seeds of the cantaloupe cv Ideal (Galia type), were sown in each pot, and five pots were served as replicates for each isolate. Pots were irrigated and fertilized as recommended.

Assessment:

Plants suffered from pre - and post - emergence damping - off disease were assessed two and four weeks after sowing respectively. Sudden wilt disease incidence was recorded as of the fruit setting stage of cantaloupe plants. Plants were uprooted washed carefully, and the disease incidence was determined. Disease severity was determined using an improved grading system for measuring plant diseases described by (**Horsfall and Barratt, 1945**). Also, plant growth parameters such as root and shoot length and root and shoot fresh weight were recorded.

Biological control:

***In vitro* Experiment:**

One of the highest virulent fungal species represented each fungal genera were chosen in this study. The fungi used were *Fusarium solani* (isolate No. 6), *Rhizoctonia solan* (isolate No. 4), *Macrophomina phaseolina* (isolate No. 7), *Pythium aphanidermatum* (isolate No. 1) and *Monosporascus cannonballus* (isolate No.1).

The studied *Trichoderma* species were isolated from the rhizosphere and rhizoplane of cantaloupe roots. The mentioned *Trichoderma* species were microscopically identified according to their morphological features (**Almassi, *et al.*, 1991**). The antagonistic ability of five *Trichoderma* spp*, i.e. T. harzianum*, *T. hamatum*, *T. koningii*, *T. reesei* and *T. viride* was tested on each of the different pathogens of cantaloupe: (*F. solani*, *R. solani*, *M. phaseolina*, *P. aphanidermatum* and *M. cannonballus*) in dual culture according to the method of (**Fokkema, 1973**). One week old PDA culture of each pathogen was used as a source of inoculum. A 4 mm disc of each species of *Trichoderma*, 7 days old grown on PDA, was placed 20 mm apart from the edge of PDA plate (90 mm). A 4 mm disc of the known pathogen was placed across from the *Trichoderma* disc and was 20 mm away from the other side of the plate edge. Check plate was inoculated with 4 mm the known pathogen alone. Cultures were incubated in the dark at 25°C until the growth of the pathogen covered the check plates. The inhibition of each pathogen growth was taken as an index of antagonistic ability, which was calculated by comparing radial growth of the pathogen colony directly to the biocontrol fungi colony with maximum radial growth according to (**Zhou and Reeleder, 1990**);

Percentage of inhibition = (R1 – R2) / R1 x 100 where,

R1 = is the maximum radius of the pathogen colony.

R2= is the radius of that part of the pathogen colony directly opposite the biocontrol agent colony.

Greenhouse Experiment:

The biological control potential of each of the previously mentioned five *Trichoderma* spp. was tested against each of the different pathogens of cantaloupe, *i.e. F. solani*, *R. solani*, *M. phaseolina*, *P. aphanidermatum* and *M. cannonballus*, in the greenhouse.

Preparation of the inocula required from the pathogens and *Trichoderma spp*. was done as mentioned before. Two weeks before sowing, inoculum of each *Trichoderma* species was thoroughly mixed with non-sterilized field sandy loamy soil at the rate of 3% of soil weight (w/w). Soil was infested with each pathogen alone or in combinations one week before sowing. Infested soil only with each pathogen alone was considered as a check treatment. The infested soil was watered and covered with plastic sheet under greenhouse conditions.

Ten seeds of cantaloupe, Ideal cv. were sown in each pot and each treatment was replicated 4 times. Percentages of infected plants were recorded by the end of the growing period.

Chemical control:

***In vitro* Experiment:**

In vitro experiment was carried out by growing the studied fungi on PDA medium supplemented with different concentrations (10, 12, 14, 16, 18 and 20 ppm) of each of the tested fungicides (Table 1). Sterile distilled water was used for preparing stock suspension of the formulations of the tested fungicides. Discs, 5 mm in diameter, obtained from the outer margins of the tested fungi grown on PDA medium, were transferred to the center of PDA plates supplemented with the different concentrations of

each fungicide. Check plates had no fungicides. Each treatment was replicated five times. Plates were incubated at 25°C. The average diameter of each fungus was measured when the fungal growth in any one of the check plates covered the surface of the medium. However, the incubation period for *F. solani*, *P. aphanidermatum*, *M. phaseolina* and *R. solani* was 7 days and 28 days for *M. cannonballus.*

Greenhouse Experiment:

The best concentration of each fungicide tested in *in vitro* experiment and significantly reduced the radial growth of the tested pathogen was chosen for greenhouse experiment. The sterilized potted clay soil was infested with each of the studied fungi at the rate of 3% of soil weight. The infested pots were left for one week before sowing. Then, seven seeds of cantaloupe Ideal cv. were sown in each pot and five pots were used per each treatment. The potted soil were drenched with each fungicide one week after sowing and repeated every 15 days. Data were recorded at fruit setting (70 days of sowing). Inoculated pots and non-treated with fungicides were used as check treatment.

Table (1): Commercial, common names, active ingredients and the rate of application of the tested fungicides.

Commercial name	Common name	Active ingredients	Application rate / Liter
Cabritop WG 60%	**Pyraclostrobin**	MethylN-{2-[1-(4-chlorophenyl)pyrazol-3-yloxymethyl]phenyl}(N-methoxy) carbamate.	2 g
Previcur N SL 72.2%	**Propamocarb hydrochloride**	Propyl (3-dimethyl amino) propyl carbamate monohydrochloride.	2.5 ml
Moncut WP 30%	**Flutolanil**	N-(3-(1- methylethoxy)phenyl)-2-(trifuoromethyl)benzamide.	2 g
Rizolex T WP 50%	**Telcolofos-methyl/ thiram**	20% Telcolofos-methyl (0,2,6 dichloro-4-methyl-phenyl 0,0 dimethyl phosphoro thioate) and 30% thiram.	3 g
Shirlan SC 50%	**Fluazinam**	3-chloro-N-(3-chloro-5-trifluoromethyl-2-pyridyl)-ά,ά,ά-trifluoro-2,6-dinitro-p-toluidine	0.5 ml
Topsin M WP 70%	**Thiophanate-methyl**	(Dimethyl[1,2-phenylene])-bis(iminocabonothioyl)bis [carbamate]).	1.5 g

3. Results

Isolation and Identification:

Five genera of pathogenic fungi, *i.e. Fusarium*, *Monosporascus*, *Rhizoctonia*, *Pythium* and *Macrophomina* were isolated from diseased roots of cantaloupe. They were identified to their species level as *F. solani*, *Macrophomina phaseolina*, *Monosporascus cannonballus*, *Rhizoctonia solani* and *Pythium aphanidermatum*.

Pathogenicity test:

Pathogenicity test of different fungal isolates was conducted in order to confirm their virulence and to define the most aggressive fungal isolates causing serious damage on cantaloupe plants. Thirty six fungal isolates were selected out of one hundred and forty three fungal isolates.

The most pathogenic isolates of the studied fungi were chosen to perform the pathogenicity test as follows: Eight isolates of *F. solani*, *P. aphanidermatum*, and *M. phaseolina*, seven isolates of *R. solani* and 5 isolates of *M. cannonballus* were chosen to perform the pathogenicity test.

Data presented in **Tables** (2, 3) indicated that all the tested isolates were pathogenic on cantaloupe Ideal cv. since they significantly increased root diseases parameters.

Data given in **Table** (2) reveal that *F. solani* (isolate No. 6), *P. aphanidermatum* (isolate No. 1), and *M. phaseolina* (isolate No. 7) were the most aggressive isolates than the others with significant differences between them. These aggressive isolates, however, showed the highest significant percentages of plant mortality.

Data presented in **Table** (3) show that *R. solani* (isolate No. 4) and *M. cannonballus* (isolate No. 1) were the most aggressive isolates than the other ones of these fungi with significant differences between these isolates and the others. These aggressive isolates showed the highest significant percentages of plant mortality.

The five virulent isolates of *F. solani* (isolate No. 6), *M. phaseolina* (isolate No. 7), *P. aphanidermatum* (isolate No. 1), *R. solani* (isolate No. 4) and *M. cannonballus* (isolate No. 1), were chosen for performing biological and chemical control measures of sudden wilt disease on cantaloupe.

Table (2): Virulence of eight isolates per each one of *F. solani*, *P. aphanidermatum* and *M. phaseolina* against the susceptible cantaloupe. (cv. Ideal), under greenhouse conditions.

Fungus/ Isolates	Disease parameters				Survival plants (%)
	Pre-emergence (%)	Post-emergence (%)	Vine decline (%)	Mortality (%)	
F. solani					
Isolate 1	34.16*	18.55	12.69	47.31	42.69
2	29.70	15.94	11.06	42.69	47.31
3	26.56	16.38	11.06	36.77	53.53
4	36.70	16.38	12.69	47.31	42.69
5	25.69	18.55	9.00	40.27	49.62
6	30.08	21.25	15.94	**49.62**	**40.27**
7	21.25	15.94	9.00	35.62	57.34
8	21.25	16.38	5.31	33.21	56.02
Control	3.69	3.69	0.71	5.31	76.6
L. S. D. at 5%	3.80	2.25	0.85	2.90	5.44
P. aphanidermatum					
Isolate **1**	33.21	16.38	9.00	**42.69**	**47.31**
2	24.22	3.69	5.31	29.09	57.34
3	25.69	12.69	12.69	38.03	52.07
4	25.69	24.64	5.31	41.44	48.46
5	15.94	15.94	12.69	34.16	56.06
6	15.94	18.55	9.00	34.16	56.06
7	18.55	12.69	9.00	29.70	58.72
8	16.38	9.00	15.94	29.70	58.72
Control	3.69	3.69	0.71	5.31	76.6
L. S. D. at 5%	2.08	1.22	0.90	1.06	3.66
M. phaseolina					
Isolate 1	15.94	12.69	5.31	29.09	60.78
2	25.69	15.94	12.69	40.27	49.62
3	16.38	12.69	16.38	29.70	58.72
4	15.94	11.25	11.25	33.21	56.02
5	16.38	9.00	5.31	26.56	72.00
6	9.00	11.25	6.00	26.56	72.00
7	25.69	15.94	18.55	**42.69**	**49.16**
8	16.38	16.38	15.94	34.16	56.06
Control	3.69	3.69	0.71	5.31	76.6
L. S. D. at 5%	2.10	1.85	0.88	1.33	2.92

*Mean of five replicates.
Percentages data were arcsine-transformed before carrying out the analysis of variance.

Table (3): Virulence of *R. solani* and *M. cannonballus* isolates against the susceptible cantaloupe cv. Ideal, under greenhouse conditions.

Fungus/ Isolates	Disease parameters				Survival plants (%)
	Pre-ergence(%)	Post-emergence (%)	Vine decline (%)	Mortality (%)	
R. solani					
Isolate 1	18.55	16.38	9.00	33.21	56.02
2	23.49	16.38	12.69	37.98	52.07
3	15.94	23.49	18.55	42.69	47.31
4	29.70	23.49	15.94	**50.87**	**39.13**
5	24.22	15.94	16.38	33.21	56.02
6	25.69	9.00	12.69	36.77	53.53
7	30.55	9.00	15.94	41.44	48.46
Control	3.69	3.69	0.71	5.31	76.6
L. S. D. at 5%	1.66	1.35	0.68	0.98	2.25
M. cannonballus					
Isolate **1**	29.22	29.70	(12.69)	**50.87**	**39.13**
2	23.31	23.31	(9.00)	37.98	52.07
3	26.56	26.56	(12.69)	43.80	46.15
4	29.22	23.31	(9.00)	42.69	47.31
5	33.21	24.22	(16.38)	49.62	40.27
Control	3.69	0.71	(3.69)	5.31	76.6
L. S. D. at 5%	0.57	0.88	0.56	1.48	3.66

*Mean of five replicates. Percentages data were arcsine-transformed before carrying out the analysis of variance.

Control methods:

Biological control:

In vitro experiment showed that all the tested *Trichoderma* spp. **Table** (4), inhibited the growth of all pathogens under study to a limited extent. *T. ressei* was significantly the best bioagent used, while there was no significant difference between either *T. pseudokoningii* and *T. hamatum* or *T. viride* and *T. harizanum*. *F. solani* and *R. solani* were generally the most sensitive fungi to the tested *Trichoderma spp.*, while, *P. aphanidermatum*, *M. cannonballus* and *M. phaseolina*, were less sensitive in this respect.

Table (4): Effect of five species of *Trichoderma* on the causal agents of sudden wilt growth *in vitro*.

Fungi	*Mean inhibition zones (mm)*					
	T. pseudo-koningii	*T. viride*	*T. harzianum*	*T. ressei*	*T. hamatum*	*Mean*
F. solani	8.00	8.00	8.00	10.00	10.0	8.80[a]
M. phaseolina	4.00	2.00	4.00	6.00	8.00	4.80[a]
P. aphanidermatum	1.00	1.00	1.00	1.00	2.00	1.20[b]
R. solani	8.00	8.00	6.00	10.00	1.00	8.40[b]
M. cannonballus	4.00	2.00	2.00	2.00	4.00	2.80[b]
Mean	5.00[b]	4.20[c]	4.20[c]	5.80[a]	5.00[b]	-

Standard deviation between *Trichoderma spp*. means = 0.67
Standard deviation between fungi means = 3.5
Numbers in column or row followed by the same letter are not significantly different.

Table (5) showed that *Trichoderma harizanum* reduced the infection of cantaloupe plants to 10 % for *F. solani*, 5% for *M. phaseolina*, *P. aphanidermatum*, *R. solani* and *M. cannonballus* in the greenhouse experiment.

Table (5): Effect of five species of *Trichoderma* on sudden wilt disease infection (%) on cantaloupe, (cv. Ideal), under greenhouse condition.

Pathogens	*Mean percentage of infection*						
	T. pseudo-koningii	*T. viride*	*T. harzianum*	*T. ressei*	*T. hamatum*	*Control**	*Mean*
F. solani	**35**	**25**	**10**	**20**	**25**	**60**	**29.2**
M. phaseolina	**15**	**10**	**5**	**15**	**40**	**50**	**22.5**
P. aphanidermatum	**30**	**10**	**5**	**10**	**20**	**40**	**19.2**
R. solani	**20**	**15**	**5**	**15**	**15**	**40**	**18.3**
M. cannonballus	**35**	**20**	**5**	**10**	**20**	**25**	**19.2**
Mean	**27**	**16**	**6**	**14**	**24**	**43**	**-**

*Control = Control without bioagent
LSD at 5% for Fungi (F) = 2.4
Bioagents(B) = 2.9
F x B = 3.1

Chemical control:

a. *In vitro* Experiment:

Data in **Table** (6) show that the used fungicides differed in their effect on the linear growth of each pathogen under this study as follows:

In general, all the best concentration used for the tested fungicides was 20 ppm. For example Capritop, Moncut, Topsin M, and Shirlan at 20 ppm reduced the linear growth of *M. phaseolina* when compared to the control. In addition, Capritop, Moncut, Rizolex T, and Shirlan at 20 ppm reduced the linear growth of *M. cannonballus* when compared to the control. Also, Capritop, and Shirlan were the most effective in reducing the linear growth of *F. solani* at 20 ppm followed by Topsin M at the same concentration. Linear growth of *R. solani* was significantly reduced by Shirlan followed by Capritop, Moncut, and Topsin M at 20 ppm. Also, the linear growth of *P. aphanidermatum* was reduced by Shirlan followed by Previcur N.

b. Greenhouse Experiment:

The same fungicides used in *in vitro* experiment were used in greenhouse experiment at 20 ppm to control the sudden wilt disease of cantaloupe. Data in **Table** (7) showed that Capritop and Topsin M were the most effective fungicides in controlling the tested pathogens in the greenhouse when used at the rate of 20 ppm. Topsin M and Capritop were the best fungicides in controlling *F. solani* and *M. phaseolina* respectively. Also, Rizolex T, Previcur N, and Shirlan were the most effective fungicides in controlling *R. solani*, *P. aphanidermatum*, *and M. cannonballus* respectively.

Table (6): Effect of six concentrations (ppm) of six fungicides on the linear growth (cm) of sudden wilt pathogens 6 days after of incubation under laboratory conditions.

Fungi	Fungicide	Concentrations (ppm)/ Linear growth (cm)						Mean fungicides	Mean Fungi
		10	12	14	16	18	20		
M. phaseolina	Cabriotop	6.0	4.4	1.8	1.6	1.2	0.8	2.6	3.9
	Moncut	7.4	6.8	6.0	5.2	4.0	0.8	5.0	
	Topsin M	1.8	1.6	1.4	1.2	1.0	0.8	1.3	
	Rizolex T	6.0	5.0	4.4	3.6	2.0	1.8	3.8	
	Previcur N	6.8	5.8	4.2	4.6	2.8	2.4	4.4	
	Shirlan	1.8	1.0	1.0	1.2	1.0	0.8	1.1	
	Control	9.0	9.0	9.0	9.0	9.0	9.0	9.0	
Mean		5.0	4.1	3.1	2.9	2.0	1.2		
M. cannonballus	Cabriotop	1.2	1.0	0.8	0.6	0.4	0.2	0.7	2.8
	Moncut	4.0	2.4	2.0	1.2	1.0	0.8	1.9	
	Topsin M	1.2	1.0	0.8	0.6	0.4	0.2	0.7	
	Rizolex T	1.2	1.0	0.8	0.6	0.4	0.2	0.7	
	Previcur N	7.8	7.6	6.0	5.8	5.6	4.0	6.1	
	Shirlan	1.2	1.0	0.8	0.6	0.4	0.2	0.7	
	Control	9.0	9.0	9.0	9.0	9.0	9.0	9.0	
Mean		3.7	3.3	2.9	2.9	2.5	2.1		
F. solani	Cabriotop	1.6	1.4	1.2	1.0	0.8	0.6	1.1	4.1
	Moncut	7.2	7.0	6.6	6.0	5.8	5.6	6.4	
	Topsin M	4.0	2.6	1.6	1.2	1.0	0.8	1.9	
	Rizolex T	5.0	3.0	2.0	1.4	1.2	1.0	2.3	
	Previcur N	7.8	7.6	7.4	7.2	7.0	6.8	7.3	
	Shirlan	1.0	1.4	1.2	1.0	0.8	0.6	1.0	
	Control	9.0	9.0	9.0	9.0	9.0	9.0	9.0	
Mean		5.1	4.6	4.1	3.8	3.7	3.5		
R. solani	Cabriotop	6.6	5.6	5.4	5.2	4.8	4.0	4.5	5.4
	Moncut	7.6	6.8	6.0	5.8	5.4	5.0	5.2	
	Topsin M	7.6	6.8	6.0	5.8	5.4	5.0	5.2	
	Rizolex T	7.6	7.2	6.8	6.0	5.8	4.6	5.4	
	Previcur N	7.0	6.0	5.8	5.4	5.0	5.6	5.0	
	Shirlan	5.0	5.0	4.0	3.8	3.4	3.0	3.5	
	Control	9.0	9.0	9.0	9.0	9.0	9.0	9.0	
Mean		7.2	6.6	6.1	5.9	5.5	5.2		
P. aphanidermatum	Cabriotop	7.8	7.6	7.0	5.0	4.8	4.0	6.0	5.9
	Moncut	7.2	5.8	5.0	4.5	3.6	3.7	5.0	
	Topsin M	7.8	6.8	5.8	5.4	4.4	3.8	5.7	
	Rizolex T	7.8	7.6	7.0	5.8	4.6	3.8	6.1	
	Previcur N	7.8	7.4	6.8	5.6	5.2	2.6	5.9	
	Shirian	6.0	4.8	3.8	3.2	3.8	2.0	3.9	
	Control	9.0	9.0	9.0	9.0	9.0	9.0	9.0	
Mean		7.6	7.0	6.3	5.5	5.1	4.1		
L. S. D. at 5% for Fungi (F) = 1.2 F X Fu = 1.7 Fungicides (Fu) = 0.9 F X C = 2.5 Concentrations (C) = 1.5 FU X C = 2.2 F X Fu X C= 2.9									

Table (7): Effect of the most effective concentration (20ppm) of studied fungicides against the causal agents of sudden wilt on cantaloupe cv. Galia in greenhouse.

Pathogen	Fungicides / % Infection						
	Cabritop	Moncut	Topsin M70	Rizolex T	Previcur N	Shirilan	Check treatment
F. solani	45	50	25	60	70	75	79
M. phaseolina	25	40	40	30	60	60	70
P. aphanidermatum	30	50	45	60	20	25	63
R. solani	40	50	40	25	70	40	76
M. cannonballus	45	35	40	50	50	25	58
Mean	37^{c}	45^{bc}	38^{c}	45^{bc}	54^{b}	45^{bc}	69.2^{a}

Standard deviation between fungicides means = 11.05
Means followed by the same letters are not significantly different at 0.05 of probability.

4. Discussion

In the past two decades, a destructive disorder of cantaloupe (*Cucumis melo* L.), characterized by sudden (commonly within 2 weeks to harvest) and generally uniform collapse of entire fields has plagued the cantaloupe industry in warmer climatic production regions (**Reuveni *et al*., 1983, Krikun, 1985 Eyal and Cohen, 1986, Uematsu *et al*., 1992, Martyn and Miller, 1996, Pivonia *et al*., 1996a,b , Stanghellini *et al*., 1996 and Pivonia *et al*., 1997**) Common names of the disorder include crown blight, collapse, vine decline, quick decline and sudden wilt (**Reuveni *et al*., 1983, Eyal and Cohen, 1986, Ucko *et al*., 1992, Garcia-Jimenez, *et al*. 1994, Stanghellini *et al*., 1995, Cohen *et al*., 1996, Bruton and Miller, 1997, Pivonia *et al*., 1998a and b, Cohen *et al*., 1999 and Edelstein *et al*., 1999**).

In the present study, the sudden wilt symptoms were recorded on cantaloupe plants. Soilborne fungi belonged to genera. *F. solani*, *M. phaseolina*, *M. cannonballus*, *P. aphanidermatum* and *R. solani* were the species most frequently isolated from root systems of sudden wilted cantaloupe plants

Pathogenicity tests were conducted in order to establish the relative importance of the various fungi isolated from wilted plants to the sudden wilt syndrome of cantaloupe in the greenhouse. Eight isolates of *F. solani*, *P. aphanidermatum*, and *M. phaseolina*, seven isolates of *R. solani* and 5 isolates of *M. cannonballus* were chosen for pathogenicity test. All the tested isolates were pathogenic on cantaloupe since they significantly increased root diseases parameters. *F. solani* (isolate No. 6), *P. aphanidermatum* (isolate No. 1), and *M. phaseolina* (isolate No. 7), *R. solani* (isolate No. 4) and *M. cannonballus* (isolate No. 1) were the most aggressive isolates than the others with significant differences between them.

In vitro experiment, using several species of *Trichoderma* as biological control agents against the tested sudden wilt pathogens in dual culture revealed that *Trichoderma* spp. inhibited the growth of all pathogens under study to a limited extent. *T. ressei* was significantly the best bioagent used, while there was no significant difference between either *T. pseudokoningii* and *T. hamatum* or *T. viride* and *T. harizanum*. *F. solani* and *R. solani* were generally the most sensitive fungi to tested *Trichoderma* spp., while, *P. aphanidermatum*, *M. cannonballus* and *M. phaseolina*, were less sensitive in this respect. However, *T. harizanum* reduced the infection of cantaloupe plants to 10 % for *F. solani*, 5% for *M. phaseolina*, *P. aphanidermatum*, *R. solani* and *M. cannonballus* in the greenhouse experiment. To date few reports on biological control of the sudden wilt causal agents were indicated. However, several methods of biological control have shown promise. Inoculation with a gliotoxin producing strain of *Trichoderma virens* has produced a significant reduction in Monosporascus disease on muskmelon roots (**Zhang *et al*., 1999**) and hypovirulent isolates of *M. cannonballus* show potential for reducing the infection by *Monosporascus* on cantaloupe (**Batten *et al*., 2000**). However, neither has been used on a commercial basis.

The fungicidal activity of each of six fungicides, Cabriotop, Moncut, Topsin M, Rizolex T, Previcur N, and Shirlan on the vegetative mycelial growth of the tested pathogens was studied *in vitro*. Each fungicide was used at seven concentrations 0, 10, 12, 14, 16, 18, and 20 ppm. Results showed that the most effective concentration for the tested fungicides was 20 ppm. Capritop, Moncut, Topsin M, and Shirlan at 20 ppm reduced the linear growth of *M. phaseolina* when compared to the control. Also, Capritop, Moncut, Rizolex T, and Shirlan at 20 ppm reduced the linear growth of *M. cannonballus* when compared to the control. Capritop, and Shirlan were the most effective in reducing the linear growth of *F. solani* at 20 ppm followed by Topsin M at the same concentration as well. Likewise, linear growth of *R. solani* was significantly reduced by Shirlan followed by Capritop, Moncut, and Topsin M at 20 ppm. Also, the linear growth of *P. aphanidermatum* was reduced by Shirlan followed by Previcur N.

However, in greenhouse the same fungicides were applied as soil drench to control the tested pathogen of cantaloupe at 20 ppm. Capritop and Topsin M were the most effective fungicides in controlling the tested pathogens. Topsin M and Capritop were the best fungicides in controlling *F. solani* and *M. phaseolina* respectively. Likewise, Rizolex T, Previcur N, and Shirlan were the most effective fungicides in controlling *R. solani*, *P. aphanidermatum*, and *M. cannonballus*, respectively. Traditionally, methyl bromide has been effective for controlling fungal infections such as Monosporascus. In fields infested with *M. cannonballus*, the fumigation of melon beds before planting with methyl bromide effectively reduces stunting of the plants and increases melon yield (**Martyn and Miller, 1996**). However, its impending phase out for use in developed countries presents a challenge to the established agriculture scientific community to develop effective alternatives that are environmentally acceptable (**Cohen *et al*., 2000**). Such alternatives may include manipulating irrigation, grafting onto more resistant genotypes, good drainage, breeding for resistance, improved soil solarization, fungicides, crop rotation, and biological

control. Since no single method is currently available to replace methyl bromide, a combined management approach will likely be most effective. Continuous monitoring of fields for early detection of pathogen escape will be necessary (**Cohen *et al*., 2000**). In comparison with fumigants, soil fungicides are usually more cost effective. Another advantage is that fungicides make up and application often allow for targeting of specific organisms with less likelihood of having detrimental effects on microorganisms in the soil. Fluazinum and kresoxim-methyl are two of the most effective fungicides tested; both inhibit *M. cannonballus* growth at concentrations of 10 µg *a.i.* /ml. Because Fluazinum also inhibited *P. aphanidermatum*, which may be involved in melon sudden wilt, and kresoxim-methyl did not inhibit this pathogen, Fluazinum was chosen for field application (**Cohen *et al*., 2000**).

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