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CHARCOAL ROT DISEASE OF CUCURBITACEOUS PLANTS

M. A. Awad, M. M. Ammar and A. A. Bazanboor
Agric. Botany Dept., Faculty of Agric., Minufiya Univ., Shibin El-Kom, Egypt.
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ABSTRACT: *Cucurbit plants are widely grown in Egypt. They can be grown in different seasons throughout the year round in open fields and in protected cultivations. Cucumber an cantaloupe are considered two of the major vegetable crops for local consumption and export. Macrophomina phaseolina is the most soil borne pathogen on cucumber and cantaloupe plants causing charcoal rot disease and reducing the fruit yield. Isolation and identification of the causal pathogen were done using samples from different cucurbits growing areas from nine governorates in Egypt, as well as biological control agents. 13 M. phaseolina isolates were used in pathogenicity tests and revealed as pathogenic to cucumber and cantaloupe plants. Soil solarization of pots infested by two isolates of the pathogen; aggressive to cucumber plants; and another two isolates; aggressive to cantaloupe plants; were done for physical control of charcoal rot disease under field conditions. There were significant differences between polyethylene sheet treatments and non-treated pots; black sheet was the best in decreasing all disease parameters and increasing numbers of survival plants. Calcium salts in both tested concentrations decreased disease parameters and increased survivals in both infested cucumber and cantaloupe plants. Antioxidants great affected charcoal rot disease in cantaloupe and cucumber plants especially when applied with high concentrations. Biological control agents were minimized all disease parameters and maximized survivals. Applied bioagents were varied in controlling the disease.*

Key words: *Charcoal rot, cucurbits, Macrophomina phaseolina, disease control.*

INTRODUCTION

Cucurbitaccous plants i.e., *Cucumis* spp. (cucumber & melon), *Citrullus* spp. (watermelon) and *Cucurbits* spp. (squash & pumpkin) are widely grown in Eygpt. They can be grown in different seasons throughout the year round in Egypt, in open fields and in protected cultivations.

Cucumber (*Cucumis sativus* L.) and cantaloupe (*Cucumis melo* var. *reticulates*) are consider two of the major vegetable summer crops in commercial fields in Egypt. During the last few decades efforts were concentrated to grow these crops in protected system in greenhouses during autumn and winter seasons. The cultivated area of cucurbits is progressing at a relatively fast rate, especially in newly reclaimed desert lands.

Several fungal diseases attack cucumber and cantaloupe during different growth stages causing considerable losses in fruit yield. Soil borne diseases are economically very important and responsible of losses in fruit yield due to diseases infection. *Macrophomina phaseolina* is the most common pathogen on cucumber an cantaloupe plants causing charcoal rot and reducing the fruit yield (Yang and Navi, 2003). Charcoal rot disease was recorded as collar rot on squash plants in Brazil (Rego, 1994). Symptoms of charcoal rot included a brown spot spread around the stem or slightly aboveground level besides black lesions on the secondary roots; on necrotic roots, when epidermis peeled off, pycnidia could be observed, spore and pycnidia were the characteristic of *M. phaseolina* (Grezes-Besset *et al.,* 1996). *M. phaseolina* has a wide spread occurrence on many cultivars in their mature plants (Rego, 1994; Manici *et al.,* 1995 and Suchandra *et al.,* 2000). Many researchers reported about control of charcoal rot disease using different methods i.e., Lodha *et al.* (1997), Ahmed *et al.* (2000) and Ndiaye (2007) on soil solarization; Chang *et al.* (2007) on calcium salts; Khalifa (2003) and Abdou (2007) on antioxidants; Hussain *et al.* (1990), Ramakrishnan *et al.* (1994), Bandyoadhyaya and Cardwel (2002) and Ndiaye (2007) on biological control.

Therefore, this study was carried out to survey and frequent isolates of charcoal rot pathogen attacking cucumber and cantaloupe plants. Pathogenicity tests and evaluation of the common and commercial cultivars. Using physical control (soil solarization), biological control agents, calcium salts and antioxidants to control charcoal rot disease in cucumber and cantaloupe.

MATERIALS AND METHODS

Samples of diseased cucumber and cantaloupe plants showed charcoal rot symptoms were collected from different cucurbits growing areas in Egypt in different growing dates in 2007 season from nine governorates. These samples were used in pathogen isolation. The obtained pure cultures of the causal organism were examined and identified at Agric. Bot., Dept., Fac. of Agric., Minufiya Univ., according to the methods adopted by Barnett and Hunter (1972).

Isolation and identification of biological control agents from soil and rhizosphere of cucurbitceous fields were done according to Elad *et al.* (1982), Rifai (1969) and Bissett (1991).

Thirteen isolates of *M. phaseolina* were chosen for testing their virulence against the susceptible cultivars of cucumber "Beit Alpha" and cantaloupe "Ananas". Disease incidence was recorded as number and percentages of pre-emergence damping-off (2 weeks after sowing), post-emergence damping-off (4 weeks after sowing) and number of survival plants (70 days after sowing). Charcoal rot disease severity index was estimated at 70 days from planting according to Soliman *et al.* (1988), modified by Awad (2004).

Control of charcoal rot disease of cucurbits was done using fungal inocula of 4 selective *M. phaseolina* isolates i.e., 3, 4, 10 & 13 and different methods of control.

1. **Soil solarization** was carried out in black plastic pots under field conditions. Four polyethylene sheets were used i.e., transparent, red, green and black sheets. Inoculated pots were covered with one sort of plastic sheet and exposed to daily sunlight for one month (15 May to 15 June, 2007) and irrigated as usual during this period then, plastic sheets removed and cucumber and / or cantaloupe seeds were seeded in treated pots and disease date were estimate as usual.

2. Chemical control:

a) Four calcium salts i.e., sulphate, chloride, phosphate and carbonate were applied for controlling charcoal rot disease incidence in pots under greenhouse conditions. 200 and 400 ppm solutions were treated as soil drenches individually as irrigation treatment every 15 days intervals.

b) Antioxidants: ascorbic acid, hydroquinone, salicylic acid, sodium benzoate and ethylene diamine antioxidants at 12.5, 25, 50, 100 & 200 ppm were used for controlling the charcoal rot disease on cucumber and cantaloupe. Soil drenching with antioxidants solutions at different concentrations 2 weeks intervals.

3. Biological control:

Ten biological control agents that used in these experiments were 10 isolates of *Trichoderma* spp. i.e., *T. harzianum* (Tz_1 to Tz_6), *T. ressei* (Tr), *T. viride* (Tv) and *T. hamatum* (Tm_1 & Tm_2) against the same mentioned four pathogen isolates under greenhouse conditions. Inocula of bioagents were individually mixed thoroughly with sterilized field loamy soil at the rate of 3% of soil weight, the watered and left for one week for bioagents spread in pots soil, then inoculated with pathogen isolate individually and watered. Two days after seeds were sowed and disease parameters were calculated and recorded.

4. Statistical analysis:

All data obtained were subjected to the proper statistical analysis for each experiment using the Duncan's statistical software. Comparisons were made following Fishers LSD (0.05).

RESULTS AND DISCUSSION

Data illustrated in Table (1) indicated that significant differences were noticed between all tested isolates in disease parameters on both tested cultivars as compared with control treatment. Pre emergence damping-off was recorded highly significant values within cucumber "Beit alpha" genotype by isolates No. 9 followed by 7 & 10 and cantaloupe tested genotypes "Ananas" by isolates No. 5 and 10. Post-emergence on cantaloupe genotype was recorded the higher value by isolates 9 & 13, while it was recorded by isolates 3 & 6 on cucumber genotype. Disease severity index (DI) was recorded as highly significant values on cucumber by isolates 3 & 4, whereas by isolates 10 & 13 on cantaloupe genotype. These results confirmed those obtained by Baudry and Morzieres (1993), Mertely *et al.* (2005) and Zveibil and Freeman (2005).

Table (1). Pathogenicity of thirteen isolates of *Macrophomina phasolina* on charcoal rot incidence of Cantaloupe cv. Ananas and Cucumber cv. Beit Alpha under greenhouse conditions.

Isolate No.	Cantaloupe				Cucumber			
	Disease Parameters %			S.P %	Disease Parameters %			S.P %
	Pre	Post	D.I		Pre	Post	D.I	
1	15.78^{b}	31.57^{ab}	89.47^{b}	47.36^{a}	41.17^{a}	11.76^{a}	82.35^{ab}	47.05^{a}
2	47.36^{a}	15.78^{a}	92.10^{a}	36.84^{a}	35.29^{a}	0.00^{b}	83.82^{b}	58.82^{a}
3	31.57^{a}	15.78^{a}	82.89^{b}	52.63^{a}	41.17^{a}	35.29^{a}	95.58^{a}	29.40^{a}
4	36.83^{ab}	5.26^{b}	61.84^{b}	57.89^{a}	35.28^{ab}	11.76^{b}	98.52^{a}	58.82^{a}
5	57.89^{a}	5.26^{b}	86.84^{b}	36.83^{ab}	41.17^{a}	17.64^{a}	92.64^{b}	47.05^{a}
6	36.84^{a}	21.05^{a}	78.94^{b}	42.10^{a}	41.17^{a}	35.29^{a}	72.05^{b}	29.40^{a}
7	42.10^{a}	15.78^{a}	90.78^{a}	42.10^{a}	58.81^{a}	17.64^{b}	70.58^{b}	29.40^{ab}
8	10.52^{b}	15.78^{b}	59.21^{b}	68.41^{a}	52.93^{a}	0.00^{b}	89.70^{a}	52.93^{a}
9	21.05^{b}	42.10^{a}	84.21^{b}	36.83^{a}	64.69^{a}	11.76^{b}	91.17^{a}	29.40^{b}
10	57.89^{a}	26.31^{ab}	94.73^{a}	10.52^{b}	58.81^{a}	5.88^{b}	86.76^{a}	35.28^{ab}
11	31.57^{ab}	10.52^{b}	88.15^{b}	57.89^{a}	29.40^{b}	5.88^{b}	77.94^{b}	70.58^{a}
12	26.31^{a}	21.05^{ab}	93.42^{a}	52.63^{a}	29.40^{b}	23.52^{a}	75.00^{b}	47.05^{a}
13	31.57^{a}	42.10^{a}	97.36^{a}	26.31^{a}	47.05^{ab}	0.00^{b}	85.29^{a}	58.81^{a}
Mean	32.22	20.64	84.61	43.71	44.33	13.57	84.72	45.69
Control	0.00	0.00	10.00	100.00	0.00	0.00	15.00	100.00

Within columns, means followed by a common letter do not differ significantly by Duncan's multiple range test ($P \leq 0.05$).

Key: Pre = Pre-emergence damping off. D. I. = Disease index.
Post = Post-emergence damping off. S.P = Survival Plants.

Data in Table (2) indicated that covering of inoculated pots decreased all disease parameters in comparing to control (non-covered) treatment. The least pre-emergence damping-off was recorded by covering inoculated pots with black sheets in case of isolate 3 against cucumber plants, while this treatment was recorded the higher percentage of post-emergence. Noticeable differences were noticed between covered treatments and non covered control. The least disease index value was recorded by black sheet, followed by transplant sheet in comparing to control.

The highest survival plants number was recorded by covering with black sheet incase of isolate 3. Isolate (4) of *M. phaseolina* was strongly affected by soil solarization and the disease parameters were recorded on cucumber plants at the same trend of isolate (3) as shown in Table (2).

Table (2). Effect of soil solarization on charcoal rot incidence of cucumber genotype Beit alpha incited by two aggressive isolates of *Macrophomina phaseolina* under greenhouse conditions.

Treatments	Fungal isolates							
	Isolate (3)				Isolate (4)			
	Disease parameters %			S.P %	Disease Parameters %			S.P %
	Pre	Post	D.I		Pre	Post	D.I	
Transparent sheet	31.37^{a}	23.52^{ab}	68.43^{a}	47.05^{ab}	23.51^{a}	47.05^{a}	71.50^{a}	31.37^{c}
Red sheet	13.05^{bc}	22.28^{ab}	69.12^{a}	4.17^{b}	23.03^{a}	14.38^{b}	68.12^{a}	35.20^{c}
Green sheet	19.66^{b}	26.96^{ab}	70.28^{a}	52.24^{ab}	26.12^{a}	16.28^{b}	63.66^{ab}	48.66^{b}
Black sheet	7.84^{c}	31.37^{a}	60.58^{b}	62.74^{a}	7.84^{b}	15.68^{c}	58.82^{b}	86.27^{a}
Mean	17.98	260.3	67.10	51.55	20.12	23.34	65.52	50.37
Control (Non-covered)	15.68	47.05	94.10	39.21	31.37	39.21	92.15	23.52
P value (sig.)	0.101	0.643	0.073	0.374	0.116	0.116	0.145	0.069

(1) Within columns, means followed by a common letter do not differ significantly by Duncan's multiple range test ($P \leq 0.05$).

(2) *P* value (sig) [≤ 0.05*, 0.01** and 0.001***] between:

Key: Pre = Pre-emergence damping-off
Post = Post-emergence damping-off.
D.I. = Disease index.
S.P = Survival Plants.

Sheets = 0.744
Isolates = 0.813
Sheets × Isolates = 0.938

Data in Table (3) recorded the effect of soil solarization on charcoal rat incidence on cantaloupe plants "Ananas" genotype incited by two aggressive isolates of *M. phaseolina* (10 & 13) under greenhouse conditions. Soil solarization great affected disease parameters on cantaloupe plants infested with both pathogen isolates. There were significant differences between polyethylene sheet treatments and non-covered treatment. Black sheet was the best treatment in decreasing pre-, post-emergence and DI. Also, survival plants were at higher level in black sheet treatment (91.22%). Similar results were reported by (Ndiaye, 2007).

Table (3). Effect of soil solarization on charcoal rot incidence of cantaloupe genotype Ananas incited by two aggressive isolates of *Macrophomina phaseolina* under greenhouse conditions.

Treatments	Fungal isolates							
	Isolate (10)				Isolate (13)			
	Disease parameters %			S.P %	Disease Parameters %			S.P %
	Pre	Post	D. I		Pre	Post	D. I	
Transparent sheet	14.03^{a}	56.14^{a}	61.22^{ab}	21.05^{b}	7.01^{b}	0.00^{c}	42.42^{a}	91.22^{a}
Red sheet	15.34^{a}	12.14^{bc}	64.36^{ab}	19.28^{b}	10.03^{a}	6.08^{b}	42.33^{a}	77.38^{b}
Green sheet	16.28^{a}	20.22^{b}	73.03^{a}	18.23^{b}	14.82^{a}	12.23^{a}	40.43^{a}	73.22^{b}
Black Sheet	14.03^{a}	0.00^{c}	35.08^{b}	91.22^{a}	0.00^{c}	14.03^{a}	37.10^{ab}	94.22^{a}
Mean	14.92	22.12	58.42	37.44	7.96	8.08	40.57	84.01
Control(Non-covered)	21.05	49.12	94.73	35.08	28.07	35.00	96.49	42.10
P value (sig)	1.00	0.039	0.062	0.019	0.374	0.374	0.452	0.678

(1) Within columns, means followed by a common letter do not differ significantly by Duncan's multiple range test ($P \leq 0.05$).

(2) *P* value (sig) [≤ 0.05*, 0.01** and 0.001***] between:

Key: Pre = Pre-emergence damping-off
Post = Post-emergence damping-off.
D.I. = Disease index.
S.P = Survival Plants.

Sheets = 0.852
Isolates = 0.945
Sheets × Isolates = 0.674

The four calcium salts in both tested concentrations decreased disease parameters that decreased insignificantly in comparing to control plants (infested) on cucumber plants that infested by both aggressive *M. phaseolina* isolates 3 & 4. Significant differences were noticed between all treatments and control in case of isolate 4 (Table 4).

Table (4). Effect of four calcium salts on charcoal rot incidence of cucumber genotype Beit Alpha incited by two aggressive isolates of *Macrophomina phaseolina* under greenhouse conditions.

Calcium Salts	Concentration (PPM)	Fungal isolates							
		Isolate (3)				Isolate (4)			
		Disease parameters %			S.P %	Disease Parameters %			S.P %
		Pre	Post	D. I		Pre	Post	D. I	
Calcium Sulphate	200	7.84^{a}	23.52^{a}	70.17^{a}	70.58^{a}	15.68^{a}	23.52^{a}	79.53^{a}	78.43^{a}
	400	7.84^{a}	23.52^{a}	70.17^{a}	70.58^{a}	23.52^{a}	31.37^{a}	70.17^{a}	39.21^{c}
Calcium Chloride	200	23.52^{a}	15.68^{a}	71.92^{a}	70.58^{a}	7.84^{a}	23.52^{a}	64.91^{a}	70.58^{ab}
	400	15.68^{a}	23.52^{a}	75.43^{a}	62.74^{a}	7.84^{a}	39.21^{a}	65.49^{a}	47.05^{bc}
Calcium Phosphate	200	7.84^{a}	15.68^{a}	72.51^{a}	86.27^{a}	15.68^{a}	31.37^{a}	74.85^{a}	47.05^{bc}
	400	15.68^{a}	31.37^{a}	65.49^{a}	54.90^{a}	0.00^{a}	15.68^{a}	49.67^{a}	94.11^{a}
Calcium Carbonate	200	15.68^{a}	31.37^{a}	82.45^{a}	62.74^{a}	7.84^{a}	31.37^{a}	70.17^{a}	70.58^{ab}
	400	15.68^{a}	23.52^{a}	63.15^{a}	62.74^{a}	7.84^{a}	39.21^{a}	65.49^{a}	70.58^{ab}
Mean		13.72	23.52	71.41	67.63	10.78	29.40	67.36	64.69
Control (infested)		23.52	47.05	92.15	23.52	31.37	54.90	94.11	7.84
P value (sig)		0.965	0.989	0.934	0.828	0.905	0.769	0.613	0.002

(1) Within columns, means followed by a common letter do not differ significantly by Duncan's multiple range test ($P \leq 0.05$).
(2) *P* value (sig) [≤ 0.05*, 0.01** and 0.001***] between:

Key: Pre = Pre-emergence damping- off. Isolates = 0.723
Post = Post-emergence damping -off. Calcium salts = 0.973
D.I. = Disease index. Isolate × Calcium salts = 0.971
S.P = Survival Plants.

Data in Table (5) indicated that on cantaloupe plants that infested with both aggressive *M. phaseolina* isolates (10 & 13), calcium salts decreased disease parameters and increased number of survival plants in comparing control plants (infested only). Significant differences were noticed between calcium treatments and control in DI in case of isolate 13, and in number of survival plants under stress of both isolates 10 & 13. the obtained results are confirmed those obtained by El-Bana *et al.* (2006) and Chang *et al.* (2007).

Table (5). Effect of four calcium salts on charcoal rot incidence of cantaloupe genotype Ananas incited by two aggressive isolates of *Macrophomina phaseolina* under greenhouse conditions.

Calcium Salts	Concentration (PPM)	Fungal isolates							
		Isolate (10)				Isolate (13)			
		Disease parameters %			S.P %	Disease Parameters %			S.P %
		Pre	Post	D. I		Pre	Post	D. I	
Calcium	200	7.01^{a}	49.12^{a}	88.88^{a}	42.10^{bc}	14.03^{a}	49.12^{a}	84.21^{b}	42.10^{a}
Sulphate	400	14.03^{a}	49.12^{a}	88.88^{a}	42.10^{bc}	21.05^{a}	28.07^{a}	66.66^{c}	56.14^{a}
Calcium	200	14.03^{a}	21.05^{a}	86.54^{a}	70.17^{ab}	7.01^{a}	35.08^{a}	86.54^{b}	63.15^{a}
Chloride	400	42.10^{a}	35.08^{a}	91.22^{a}	28.07^{c}	7.01^{a}	42.10^{a}	95.90^{a}	56.14^{a}
Calcium Phosphate	200	7.01^{a}	35.08^{a}	79.53^{a}	63.15^{ab}	0.00^{a}	42.10^{a}	86.54^{b}	63.15^{a}
	400	7.01^{a}	14.03^{a}	56.14^{b}	84.21^{ab}	14.03^{a}	35.08^{a}	84.21^{b}	56.14^{a}
Calcium Carbonate	200	14.03^{a}	28.07^{a}	74.85^{a}	63.15^{ab}	21.05^{a}	0.00^{a}	86.54^{b}	84.21^{a}
	400	14.03^{a}	42.10^{a}	87.71^{a}	49.12^{bc}	7.01^{a}	35.08^{a}	63.15^{c}	63.15^{a}
Mean		14.91	34.20	81.71	55.25	11.40	33.33	81.71	60.52
Control (infested)		42.10	39.08	98.24	21.05	28.07	56.07	91.22	7.01
P value (sig)		0.478	0.604	0.02	0.017	0.715	0.499	0.000	0.681

(1) Within columns, means followed by a common letter do not differ significantly by Duncan's multiple range test ($P \leq 0.05$).

(2) *P* value (sig) [≤ 0.05*, 0.01** and 0.001***] between:

Key: Pre = Pre-emergence damping- off.
Post = Post-emergence damping -off.
D.I. = Disease index.
S.P = Survival Plants.

Isolates = 0.93
Calcium salts = 0.98
Isolate × Calcium salts = 0.951

Soil drenching with antioxidants solutions to pots filled with soil infested with isolates 3 & 4 and planted with Beit alpha cucumber plants were effective in disease parameters as compared to control plants. Isolate 3 lost their efficiency to infect cucumber plants as pre & post-emergence in cases of all antioxidants at higher concentrations except ascorbic acid. Significant differences were noticed between all antioxidant treatments in pre-and post-emergence in case of isolate 4 on cucumber plants. DI was recorded significant differences between all antioxidant treatments in both Isolates 3 & 4. Also, significant differences were noticed between all antioxidant treatments in number of survival plants and these numbers were increased significantly (Table 6).

Table (6). Control of charcoal rot disease on cucumber plants genotype Beit Alpha with antioxidants by soil drenching under greenhouse conditions.

Antioxidants	Concentrations (ppm)	Fungal isolates							
		Isolate (3)				Isolate (4)			
		Disease parameters %			S.P %	Disease Parameters %			S.P %
		Pre	Post	D.I		Pre	Post	D.I	
Ascorbic acid	12.5	0.00^{b}	47.05^{a}	94.11^{a}	47.05^{ab}	47.05^{a}	23.52^{ab}	94.11^{a}	23.52^{c}
	25	23.52^{ab}	23.52^{a}	88.23^{abc}	47.05^{ab}	23.52^{ab}	47.05^{a}	94.11^{a}	23.52^{c}
	50	23.52^{ab}	0.00^{a}	86.27^{abc}	70.58^{ab}	23.52^{ab}	0.00^{b}	75.76^{bc}	70.58^{b}
	100	23.52^{ab}	0.00^{a}	82.35^{abcd}	70.58^{ab}	0.00^{b}	23.52^{ab}	70.58^{cd}	70.58^{b}
	200	23.52^{ab}	0.00^{a}	75.76^{de}	70.58^{ab}	0.00^{b}	0.00^{b}	47.05^{f}	94.11^{a}
Hydroqui-none	12.5	47.05^{a}	23.52^{a}	94.11^{a}	23.52^{b}	47.05^{a}	23.52^{ab}	94.11^{a}	23.52^{c}
	25	23.52^{ab}	47.05^{a}	94.11^{a}	23.52^{b}	47.05^{a}	0.00b	70.58^{cd}	23.52^{c}
	50	0.00^{b}	47.05^{a}	82.35^{abcd}	47.05^{ab}	0.00^{b}	23.52^{ab}	82.35^{ab}	70.58^{b}
	100	0.00^{b}	23.52^{a}	47.05^{f}	70.58^{ab}	0.00^{b}	0.00^{b}	58.82^{e}	94.11^{a}
	200	0.00^{b}	0.00^{a}	23.52^{g}	94.11^{a}	0.00^{b}	0.00^{b}	23.52^{h}	94.11^{a}
Salicylic acid	12.5	23.52^{ab}	23.52^{a}	94.11^{a}	47.05^{ab}	23.52^{ab}	23.52^{ab}	94.11^{a}	47.05^{bc}
	25	0.00^{b}	47.05^{a}	90.38^{ab}	47.05^{ab}	0.00^{b}	23.52^{ab}	86.27^{ab}	70.58^{b}
	50	0.00^{b}	23.52^{a}	70.58^{de}	70.58^{ab}	23.52^{ab}	0.00^{b}	70.58^{cd}	70.58^{b}
	100	0.00^{b}	0.00^{a}	54.90^{f}	70.58^{ab}	0.00^{b}	0.00^{b}	35.29^{g}	94.11^{a}
	200	0.00^{b}	0.00^{a}	47.05^{f}	70.58^{ab}	0.00^{b}	0.00^{b}	23.52^{h}	94.11^{a}
Sodium benzoate	12.5	47.05^{a}	23.52^{a}	94.11^{a}	23.52^{b}	23.52^{ab}	23.52^{ab}	86.27^{ab}	47.05^{bc}
	25	23.52^{ab}	47.05^{a}	94.11^{a}	23.52^{b}	23.52^{ab}	23.52^{ab}	62.74^{de}	47.05^{bc}
	50	23.52^{ab}	23.52^{a}	70.58^{de}	70.58^{ab}	0.00^{b}	23.52^{ab}	82.35^{ab}	70.58^{b}
	100	0.00^{b}	0.00^{a}	47.05^{f}	94.11^{a}	0.00^{b}	0.00^{b}	47.05^{f}	70.58^{b}
	200	0.00^{b}	0.00^{a}	23.52^{g}	94.11^{a}	0.00^{b}	0.00^{b}	23.52^{h}	94.11^{a}
Ethylene diamine	12.5	23.52^{ab}	23.52^{a}	88.23^{abc}	47.05^{ab}	47.05^{a}	23.52^{ab}	94.11^{a}	23.52^{c}
	25	0.00^{b}	23.52^{a}	82.35^{abcd}	70.58^{ab}	47.05^{a}	23.52^{ab}	94.11^{a}	23.52^{c}
	50	0.00^{b}	23.52^{a}	75.76^{dc}	70.58^{ab}	23.52^{ab}	47.05^{a}	94.11^{a}	23.52^{c}
	100	0.00^{b}	0.00^{a}	58.82^{ef}	94.11^{a}	23.52^{ab}	0.00^{b}	70.58^{cd}	70.58^{b}
	200	0.00^{b}	0.00^{a}	47.05^{f}	94.11^{a}	0.00^{b}	0.00^{b}	47.05^{f}	70.58^{b}
Mean		12.23	18.81	72.29	62.11	16.93	14.11	68.90	60.22
Control – (infested)		23.52	47.05	92.15	23.52	31.37	54.90	94.11	7.84
P value (sig)		0.007	0.110	0.000	0.001	0.002	0.117	0.000	0.000

(1) Within columns, means followed by a common letter do not differ significantly by Duncan's multiple range test ($P \leq 0.05$).

(2) P value (sig) [≤ 0.05*, 0.01**and 0.001***] between: Isolates = 0.405 Concentrations = 0.00 Isolate × concentration = 1.00

Key: Pre = Pre-emergence damping-off. Post = Post-emergence damping-off. D.I. = Disease index. S.P = Survival Plants.

Data in Table (7) indicated that antioxidants greatly reduced charcoal rot disease in cantaloupe plants that infested with two aggressive isolates 10 & 13. Significant differences were noticed between all tested antioxidants with their applied concentrations on disease parameters i.e., pre-, post-emergence and disease severity index (DI). These disease parameters were decreased significantly as well as increasing significantly of number of survival plants in comparing to control plants as shown in Table (7). These results are inacordance with results obtained by Galal *et al.* (2003), Abdou *et al.* (2004) and Abdel-Rahim (2007).

Table (7). Control of charcoal rot disease on cantaloupe plants genotype Ananas with antioxidants by soil drenching under greenhouse conditions.

Antioxidants	Concent-rations (ppm)	Fungal isolates							
		Isolate (10)				Isolate (13)			
		Disease parameters %			S.P %	Disease Parameters %			S.P %
		Pre	Post	D.I		Pre	Post	D.I	
Ascorbic acid	12.5	28.07^{abcd}	49.12^{a}	98.24^{a}	21.05^{g}	63.15^{a}	28.07^{abc}	95.90^{ab}	14.03^{de}
	25	28.07^{abcd}	14.03^{cd}	94.73^{ab}	63.15^{bcdef}	35.08^{bcde}	42.10^{ab}	89.47^{abc}	14.03^{de}
	50	0.00^{e}	14.03^{cd}	63.15^{ef}	91.22^{ab}	14.03^{def}	35.08^{abc}	98.24^{a}	56.14^{abc}
	100	7.01^{de}	21.05^{bcd}	86.54^{abcd}	77.19^{abcd}	35.08^{bcde}	28.07^{abc}	95.90^{ab}	42.10^{bcd}
	200	7.01^{de}	21.05^{bcd}	60.8^{1f}	77.19^{abcd}	35.08^{bcde}	7.01^{c}	87.71^{abc}	63.15^{ab}
Hydroqui-none	12.5	21.05^{cde}	28.07^{abc}	78.94^{d}	35.08^{fg}	28.07^{bcdef}	49.12^{a}	84.21^{bc}	7.01^{e}
	25	7.01^{de}	28.07^{abc}	77.19^{d}	49.12^{defg}	42.10^{abcd}	21.05^{abc}	87.71^{abc}	35.08^{bcde}
	50	0.00^{e}	14.03^{cd}	63.15^{ef}	84.21^{abc}	35.08^{bcde}	14.03^{bc}	84.21^{bc}	56.14^{abc}
	100	7.01^{de}	28.07^{abc}	59.64^{f}	63.15^{bcdef}	7.01^{ef}	14.03^{bc}	63.15^{d}	84.21^{a}
	200	14.03^{cde}	0.00^{d}	52.63^{fg}	84.21^{abc}	21.05^{cdef}	14.03^{bc}	52.63^{e}	63.15^{ab}
Salicylic acid	12.5	42.10^{ab}	7.01^{cd}	88.88^{abcd}	56.14^{cdef}	63.15^{a}	14.03^{bc}	92.98^{ab}	21.05^{de}
	25	42.10^{ab}	21.05^{bcd}	81.87^{d}	21.05^{g}	14.03^{def}	35.08^{abc}	79.53^{c}	42.10^{bcd}
	50	0.00^{e}	7.10^{cd}	51.46^{fg}	98.24^{a}	56.14^{ab}	14.03^{bc}	95.90^{ab}	35.08^{bcde}
	100	21.05^{bcde}	7.01^{cd}	59.64^{f}	70.17^{abcde}	35.08^{bcde}	42.10^{ab}	91.22^{abc}	21.05^{de}
	200	0.00^{e}	21.05^{bcd}	60.81^{f}	84.21^{abc}	7.01^{ef}	14.03^{bc}	45.61^{e}	84.21^{a}
Sodium benzoate	12.5	28.07^{abcd}	42.10^{ab}	87.71^{abcd}	21.05^{g}	21.05^{cdef}	42.10^{ab}	89.47^{abc}	35.08^{bcde}
	25	21.05^{bcde}	14.03^{cd}	79.53^{cd}	56.14^{cdef}	42.10^{abcd}	35.08^{abc}	94.73^{ab}	21.05^{de}
	50	0.00^{e}	28.07^{abc}	77.19^{d}	63.15^{bcdef}	28.07^{bcdef}	35.08^{abc}	79.53^{c}	12.05^{de}
	100	42.10^{ab}	0.00^{d}	77.19^{d}	56.14^{cdef}	7.01^{ef}	28.07^{abc}	67.83^{d}	63.15^{ab}
	200	49.12^{a}	21.05^{bcd}	88.88^{abcd}	35.08^{fg}	0.00^{f}	28.07^{abc}	66.66^{d}	63.15^{ab}
Ethylene diamine	12.5	7.01^{de}	21.05^{bcd}	75.43^{abcd}	63.15^{bcdef}	49.12^{abc}	28.07^{abc}	94.73^{ab}	21.05^{de}
	25	28.07^{abcd}	49.12^{a}	92.98^{abc}	21.05^{g}	35.08^{bcde}	14.03^{bc}	91.22^{abc}	56.14^{abc}
	50	35.08^{abcd}	28.07^{abc}	98.24^{a}	42.10^{efg}	28.07^{bcdef}	14.03^{bc}	91.22^{abc}	56.14^{abc}
	100	0.00^{e}	0.00^{d}	42.10^{g}	91.22^{ab}	21.05^{cdef}	35.08^{abc}	78.94^{c}	28.07^{cde}
	200	21.05^{bcde}	28.07^{abc}	93.56^{ab}	56.14^{cdef}	28.07^{bcdef}	42.10ab	87.71^{abc}	21.05^{de}
Mean		18.24	20.48	75.61	59.22	30.03	26.94	83.45	40.97
Control – (infested)		42.10	35.08	98.24	21.05	28.07	56.07	91.22	7.01
P value (sig)		0.000	0.000	0.000	0.000	0.000	0.012	0.000	0.000

(1) Within columns, means followed by a common letter do not differ significantly by Duncan's multiple range test ($P \le 0.05$).

(2) P value (sig) [≤ 0.05*, 0.01**and 0.001***] between: Isolates = 0.70
Concentrations = 0.25 Isolate × concentration = 0.625

Key: Pre = Pre-emergence damping-off. Post = Post-emergence damping-off.
D.I. = Disease index. S.P = Survival Plants.

Biological control agents great affected and minimized all disease parameters under stress of the four *M. phaseolina* isolates (3, 4, 10 & 13) under greenhouse conditions. Pre-, post-emergence damping-off as well as disease severity index (DI) were significant minimized by bioagents against stress of pathogen isolates in comparing to control treatments. Significant differences were noticed between all bioagents in DI under stress of isolate 3 & 4. Survival plants were increased significantly in case of isolate 4 on cucumber plants. All disease parameters were significantly affected by bioagents under stress of isolates 10 & 13 on cantaloupe plants. Generally, all *Trichoderma* spp. Isolates significantly affected the charcoal rot disease incidence in terms of the number of healthy survivals and infected plants. (Tables 8 and 9) These results were confirmed results obtained by many investigators i.e., Bandyoadhyaya dn Cardwel (2002), Adekunle *et al.* (2005) and Ndiaye (2007).

Table (8). Effect of some *Trichoderma* spp. isolates on charcoal rot disease of cucumber genotype Beit Alpha incited by two isolates of *Macrophomina phaseolina* under greenhouse conditions.

Trichoderma spp. isolate	*M. phaseolina* isolate							
	Isolate (3)				Isolate (4)			
	Disease parameters %			S.P %	Disease parameters %			S.P %
	Pre	Post	D.I		Pre	Post	D.I	
T. harzianum (1)	21.05^{a}	14.03^{a}	74.85^{abc}	70.17^{a}	14.03^{a}	28.07^{b}	86.54^{ab}	63.15^{ab}
T. harzianum (2)	7.01^{a}	21.05^{a}	56.17^{cd}	70.17^{a}	7.01^{a}	14.03^{b}	63.15^{bc}	84.21^{ab}
T. harzianum (3)	7.01^{a}	14.03^{a}	49.12^{de}	84.21^{a}	14.03^{a}	35.08^{b}	67.83^{bc}	56.14^{b}
T. harzianum (4)	0.00^{a}	28.07^{a}	63.15^{bcd}	77.19^{a}	7.01^{a}	28.07^{b}	63.15^{bc}	70.17^{ab}
T. harzianum (5)	14.03^{a}	7.01^{a}	35.08^{e}	84.21^{a}	0.00^{a}	7.01^{b}	42.10^{c}	98.24^{a}
T. harzianum (6)	14.03^{a}	14.03^{a}	59.64^{bcd}	77.19^{a}	7.01^{a}	14.03^{b}	63.15^{bc}	77.19^{ab}
T. hamatum (1)	14.03^{a}	21.05^{a}	70.17^{abc}	63.15^{a}	14.03^{a}	21.05^{b}	53.80^{bc}	63.15^{ab}
T. hamatum (2)	21.05^{a}	7.01^{a}	46.78^{de}	77.19^{a}	7.01^{a}	21.05^{b}	63.15^{bc}	77.19^{ab}
T. viride	7.01^{a}	28.07^{a}	88.88^{a}	70.17^{a}	7.01^{a}	35.08^{b}	98.24^{a}	63.15^{ab}
T. ressei	21.05^{a}	21.05^{a}	77.19^{ab}	63.15^{a}	21.05^{a}	70.17^{a}	88.88^{ab}	21.05^{c}
Mean	12.63	17.54	62.10	73.67	9.82	27.36	68.99	67.36
Control	23.52	47.05	92.15	23.52	31.37	54.90	94.11	7.84
P value (sig)	0.788	0.835	0.000	0.817	0.910	0.047	0.066	0.007

(1) Within columns, means followed by a common letter do not differ significantly by Duncan's multiple range test ($P \leq 0.05$).

(2) *P* value (sig) [≤ 0.05*, 0.01** and 0.001***] between:

Key: Pre = Pre- emergence damping-off — *M. phaseolina* isolates = 0.395

Post = Post- emergence damping-off. — *Trichoderma* spp. isolates = 0.170

D.I. = Disease index. *M. phaseolina* isolates × *Trichoderma* spp. isolates= 0.997

S.P = Survival Plants.

Table (9). Effect of some *Trichoderma* spp. isolates on charcoal rot disease of cantaloupe genotype Ananas (USA) incited by two isolates of *Macrophomina phaseolina* under greenhouse conditions.

Trichoderma spp. isolate	*M. phaseolina* isolate							
	Isolate (10)				Isolate (13)			
	Disease parameters %			S.P %	Disease parameters %			S.P %
	Pre	Post	D.I		Pre	Post	D.I	
T. harzianum (1)	30.00^{a}	15.00^{bc}	67.50^{ab}	55.00^{cd}	15.00^{a}	10.00^{ab}	50.00^{bc}	75.00^{a}
T. harzianum (2)	25.00^{ab}	10.00^{bc}	57.50^{bc}	65.00^{c}	15.00^{a}	0.00^{b}	35.00^{c}	85.00^{a}
T. harzianum (3)	0.00^{c}	10.00^{bc}	25.00^{cd}	90.00^{ab}	20.00^{a}	25.00^{ab}	75.33^{ab}	55.00^{abc}
T. harzianum (4)	25.00^{ab}	5.00^{c}	60.00^{bc}	70.00^{bc}	15.00^{a}	20.00^{ab}	52.50^{bc}	65.00^{abc}
T. harzianum (5)	15.00^{abc}	10.00^{bc}	35.00^{cd}	75.00^{abc}	25.00^{a}	20.00^{ab}	81.25^{ab}	55.00^{abc}
T. harzianum (6)	15.00^{abc}	10.00^{bc}	45.00^{bc}	75.00^{abc}	25.00^{a}	35.00^{a}	58.75^{abc}	40.00^{bc}
T. hamatum (1)	30.00^{a}	30.00^{ab}	90.00^{a}	40.00^{d}	35.00^{a}	30.00^{a}	90.00^{a}	35.00^{c}
T. hamatum (2)	20.00^{abc}	40.00^{a}	88.75^{a}	40.00^{d}	25.00^{a}	15.00^{ab}	70.00^{abc}	60.00^{abc}
T. viride	20.00^{abc}	20.00^{abc}	67.50^{ab}	60.00^{cd}	20.00^{a}	10.00^{ab}	71.66^{abc}	70.00^{ab}
T. ressei	5.00b^{c}	0.00^{c}	15.00^{d}	95.00^{a}	25.00^{a}	15.00^{ab}	66.66^{abc}	60.00^{abc}
Mean	18.50	15.00	65.11	66.11	22.00	18.00	55.99	60.00
Control	42.10	35.08	98.24	21.05	28.07	56.07	91.22	7.01
P value (sig)	0.026	0.023	0.063	0.000	0.925	0.210	0.00	0.021

(1) Within columns, means followed by a common letter do not differ significantly by Duncan's multiple range test ($P \leq 0.05$).

(2) *P* value (sig) [≤ 0.05*, 0.01** and 0.001***] between:

Key: Pre = Pre-emergence damping-off — *M. phaseolina* isolates = 0.243

Post = Post- emergence damping-off. — *Trichoderma* spp. isolates = 0.143

D.I. = Disease index. — *M. phaseolina* isolates×*Trichoderma* spp. isolates= 0.043

S.P = Survival Plants.

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مرض العفن الفحمى فى القرعيات

محمد أحمد عوض ، محمد محمد عمار ، أمين بن عبد الله على بازنبور

قسم النبات الزراعى – كلية الزراعة – جامعة المنوفية – شبين الكوم – مصر

الملخص العربى

تُزرع نباتات القرعيات فى مساحات كبيرة وفى مواسم مُتعددة على مدار العام فى مصر فى الحقول المفتوحة أو تحت الزراعات المحمية ويُعتبر الخيار والكانتالوب من أهم محاصيل الخُضر فى مصر للاستهلاك المحلى والتصدير .

وتُصاب نباتات الخيار والكانتالوب بالعديد من الأمراض متباينة المُسببات والتى من بينها الأمراض الكامنة فى التربة والتى يأتى على رأسها مرض العفن الفحمى الذى يتسبب عن الفطر ماكروفومنيا فاسيولينا .

تم جمع عينات يظهر عليها أعراض المرض من أماكن زراعات القرعيات فى تسعة محافظات فى مصر وتم عزل المُسبب المرضى منها وتنقيته وتعريفه بالإضافة إلى عزل كائنات التضاد الحيوى من التربة والريزوسفير لتلك النباتات المصابة حيث تم عزل ثلاثة عشر عزلة من الفطر المُمرض وعشرة عزلات من كائنات التضاد الحيوى – وقد أظهرت الدراسة أن جميع عزلات الفطر كانت مُمرضة لنباتات الخيار والكانتالوب تحت ظروف الصوبة وتباينت القدرة لتلك العزلات حيث تم اختبار عزلتان قويتان على نباتات الخيار وعزلتان على نباتات الكانتالوب لعمل تجارب المقاومة .

تم استخدام طريقة تشميس التربة بواسطة تغطيتها بعد حقن التربة بعزلات المسبب المرضى وذلك باستخدام مُشمعات بلاستيك مختلفة الألوان (أسود – عديم اللون – أحمر – أخضر) وذلك لمدة شهر ثم الزراعة لنباتات الخيار والكانتالوب وقد أدت عملية التشميس إلى خفض معنوى لموت البادرات قبل وبعد ظهورها فوق سطح التربة وشدة الإصابة بالمرض مع زيادة معنوية فى عدد النباتات الحية من كلا النباتين المختبرين .

تم استخدام أربعة أملاح للكالسيوم لكلٍ منها تركيزين وقد أثبتت فعالية عالية فى تخفـيض القياسات المرضية ورفع أعداد النباتات الحية . كما استخدمت خمسة مركبات مضادات للأكسدة لكلٍ منها ٥ تركيزات حيث أدت تلك المعاملات إلى السيطرة على المرض وخفض معدلاته أيضاً خاصةً عند تطبيق تلك المعاملات فى تركيزاتها العالية .

هذا وكان استخدام كائنات التضاد الحيوى فعال بدرجة كبيرة فى التحكم فى المرض وحدوثه وشدته مع تباين واضح بين كائنات التضاد الحيوى المستخدمة فى عملية المقاومة تبعاً لعزلـة الفطر الممرض المستخدمة فى عملية العدوى .