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The Use of Antioxidants to Control Root Rot and Wilt Diseases of Pepper

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Abstract

Ten isolates of *Fusarium* spp were isolated from pepper plants collected from different locations in New Valley Governorate, Egypt. *Fusarium solani* isolate FP2 and *F. oxysporum* isolate FP4 were highly pathogenic isolates but the other isolates moderate or less pathogenic to pepper plants (cv. Anaheim-M). The four antioxidant compounds (coumaric acid, citric acid, propylgalate and salicylic acid each at 100 and 200 ppm) were evaluated for their *in vitro* and *in vivo* agonist to *Fusarium* pathogenic isolates caused root rot and wilt diseases in pepper plants. All tested antioxidant compounds reduced damping-off, root rot/wilt and area under root rot/wilt progress curve when used as seed soaking, seedling soaking, and soil drench especially at 200 ppm under greenhouse and field conditions compared with untreated plants. All chemicals increased fresh and dry weight of seedling grown in soil drenching or seed treatment with any antioxidants. At the same time, all tested chemicals significantly increase plant growth parameters i.e plant length, plant branching, and total yield per plant in case of seedling soaking or soil drench. In general, propylgalate at 200 ppm was more efficient in reducing infection with damping-off, root rot and wilt diseases as well as increasing the seedling fresh weight, dry weight, plant length, plant branching, number of pod plant^{-1} and pod yield plant^{-1}. On the other hand, all tested antioxidants had less or no effect on mycelial dry weight and mycelial leaner growth. On the contrary, all chemicals much reduced spore formation in both *Fusarium* species at 100 or 200 ppm and the inhibitory effect of antioxidants increased with increasing their concentrations.

Keywords: pepper, root diseases, antioxidants, soil borne fungi

Introduction

Sweet pepper (*Capsicum annuum* L.) belongs to the family solanaceae, which is an important group of vegetables cultivated extensively in Egypt and also widely cultivated in almost every country of the world. Pepper plants are liable to be attacked by several soil-borne pathogenic fungi which are responsible for considerable plant mortality and consequently high losses in the yield and quality in many parts of the world (Lu *et al.,* 1984; Abada, 1994; Mushtaq and Hashmi, 1997).

Fusarium root rot and wilt are the most important diseases caused by *Fusarium solani* and *F. oxysporum* in sweet pepper plants. *Fusarium* root rot/wilt first appears as slight vein clearing on the outer portion of the younger leaves, followed by epinasty (downward drooping) of the older leaves. At the seedling stage, plants infected may wilt and die soon after symptoms appear. In older plants, vein clearing and leaf epinasty are often followed by stunting, yellowing of the lower leaves, formation of adventitious roots, wilting of leaves and young stems, defoliation, marginal necrosis of remaining leaves, and finally death of the entire plant. Browning of the vascular tissue is a strong evidence of *Fusarium* wilt in sweet pepper (Agrios, 1988).

Because of hazards of pesticides in general, and fungicides in specific, on public health and environmental balance (Elad, 1992) a relatively recent direction of pest control management was introduced. Inducing or acquiring the systemic resistance in the host plant became a good target for minimizing disease incidence or severity with least cost and without environmental pollution.

Antioxidants which save to human and environment had been used successfully to control some plant diseases such as root and pod rot in peanut (Elwakil, 2003), *Fusarium* wilt in chickpea (Nighat- Sarwar *et al.*, 2005), faba bean chocolate spot (Hassan *et al.*, 2006), peanut root rot (Mahmoud *et al.*, 2006), *Fusarium* wilt in tomato (El-Khallal, 2007a; Mohamed *et al.*, 2007), root rot and leaf blight in lupine (Abdel-Monaim, 2008), damping- off in pepper (Rajkumar, 2008).

At the same time, Mostasa (2006) reported that application of antioxidants, e.g. ascorbic, salicylic, coumaric, benzoic acids and propylgalate as either seed soaking or soil drenching proved sufficient protection against cumin caused by *Fusarium oxysporum, F.* sp. *cumini* or *Acreromonium egyptiacum*.

The present study aims to evaluate antioxidant compounds i.e coumaric, salicylic, citric acids and propylgalate as chemical inducers on pepper plants against root rot and wilt diseases, effect on growth parameters and pod yield and their effect on dry weight, leaner growth and sporulation *in vitro,* of *Fusarium solani* and *F. oxysporum.*

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Materials and methods

Isolation and identification of the causal organisms

Samples of pepper plants showing root rot and wilt disease symptoms were collected from different locations of New Valley Governorate (Kharga, Dakhla, Farfrah and Baris Oozes). All samples were subjected to isolation trials for the organisms according to the method devised by Sahi and Khalid (2007). The purified isolated fungi were identified according to cultural and microscopically characters described by Gilman (1957), Barnett and Hunter (1972) and Nelson *et al.* (1983).

Pathogenicity test

Ten isolates of *Fusarium* spp were selected to study their pathogenic ability to induce root rot and wilt on pepper seedling (40 d-old) in pots. Plastic pots (30 cm in diameter) containing sterilized sand loam soil infested individually with inoculums of each fungus, was grown on barley-sandy medium (140 g barley grains, 60 g sand and 60 ml water) for two weeks at 25±1°C. Five pots were used for each fungus. Check treatment (control) was prepared without the addition of the tested fungi. Healthy seedlings of pepper (cv. 'Anaheim-M') were sown at the rate of 5 seedlings/pot. The pots were kept under careful observation in the open field of plant pathology Dept., New Valley Agric. Eight weeks after sowing rot and wilt were examined. Re-isolation was carried out from some of the artificially diseased plants to fulfill Koch's postulations and the developed fungi were confirmed with the original isolates.

Disease assessment

60 days from planting, pepper plants were removed from the soil, washed thoroughly to remove soil debris, root rot/wilt assessment as follows:

0= neither root discoloration nor leaf yellowing, 1= 1-25% root discoloration or one leaf yellowed, 2= 26-50% root discoloration or more than one leaf yellowed, 3= 51-75% root discoloration or vascular discoloration plus one leaf wilted, 4= up to 76% root discoloration or more than one leaf wilted and 5= completely dead plants. For each replicate a disease severity index (DSI) similar to the one described by Liu *et al.,* (1995) was calculated as follows:

$DSI = \sum d/(d_{max} X\ n)X100$

Whereas: d is the disease rating of each plant, d max is the maximum disease rating and n is the total number of plants examined in each replicate.

Effect of antioxidant compounds on disease severity under greenhouse conditions

Coumaric acid (COA), citric (CA), salicylic acid (SA) and propylgalate (PG) were tested for controlling pepper root rot and wilt diseases caused by *Fusarium solani* and *F. oxysporum* at two concentrations (100 and 200 ppm) in pot experiments. These compounds applied by different methods as follows.

Seed socking

Healthy pepper seeds were soaked in the tested solutions for 12 hr. (600 seeds per 100 ml test solution), then 5 seeds were sowed in each pot (25 cm in diameters), five pots were used for each test as replicates. Percentage of damping-off and root rot/wilt severity were recorded 4 and 8 weeks after planting, respectively. At the end of the experiment, fresh and dry weight per plant was recorded.

Seedling socking

Healthy pepper seedlings (40 days old) were soaked in tested solution for 12 hr. (100 seedlings per 100 ml), then 5 seedlings were sowed in each pot, five pots used each test as replicates. Disease severity recorded each 10 interval days for 60 days. The mean of area under root rot/wilt progress curve (AUR/WPC) for each replicate was calculated by the formula suggested by Pandy *et al.* (1989) and calculated as follows:

$AUR/WPC = D\ [1/2\ (Y_1+Y_k) + (Y_2+Y_3+....+Y_{k-1})]$

Where D= Time interval; Y_1= First disease severity; Y_k= Last disease severity; Y_2, Y_3, ...Y_{k-1}= Intermediate disease severity.

Soil drenching

Non treated seedlings and/or seeds were sowed in pots (five per pot). Pots were irrigated with COA, CA, SA and PG solutions at two concentrations (100, 200 ppm) when they were planted. Irrigation with the test solution was applied at rate 50 ml test solution per pot. Five pots were used for each test as replicates. Damping-off, root rot/wilt and AUR/WPC were recorded as mentioned before.

All experiments grow in infested soil with *Fusarium solani* (isolate PF2) and/ or *F. oxysporum* (isolate PF4) at 3% of inocula prepared as before.

Effect of antioxidants on growth and sporulation of the tested fungi in vitro

Coumaric, citric, salicylic acids and propylgalate were used separately at 100 and 200 ppm for studying their effect on dry weight, leaner growth and sporulation of the tested fungi:

Mycelium disks (7 mm diameter) from the growing edge of 7-day-old cultures of each *Fusarium solani* and *F. oxysporum* were inoculated onto these plates containing 10 mm sterilized Czpeck's solid medium amended with each concentration of each antioxidant compounds in order to test the effect of the chemicals on linear growth. In addition, sporulation estimated after 7 days incubation under the same conditions using a heamocytometer with the help of a light microscope (Tzeng and DeVay, 1989). At the same time flask 250 ml containing 50 ml Czpeck's medium were prepared and amended with each concentration of each antioxidant compounds. Antioxidant free

medium was served as control. Three replicates were used for each treatment.

The effect of antioxidant compounds on diseases severity, plant vigour and yield under field conditions

This experimental, factorial block design experiment was conducted at sowing date of 15 February of two successive growing seasons 2008-2009 in a field naturally infected with the causal organisms of root rot and wilt diseases of pepper located at the experimental farm of Kharga Agric. Station, New Valley Governorate. The main plots were antioxidant tested, sub plots were concentrations, sub sub plots were seedlings soaking and/or soil drenching. Seedlings were sacked at rate 100 transplanting per 100 ml for 12 hr. while in case of soil drenching, tested antioxidants were applied at rate 25 ml per hill when planting. The field plots (9 m^2) consisted of 3 rows of 3 m long and 1 m in between. One seedlings/hill was sown with 50 cm apart between hills. Untreated seedlings were grown in untreated soil used as control. Disease severity were recorded as above each 30 days for 4 months. At the end of the growing season, the plant height and the number of branches were calculated. At the same time, during the experiment, pods per plant were collated and calculated on number per plant and weight of pods per plant.

In all experiments the least significant difference (LSD) at 0.05 confidences was determined according to Gomez and Gomez (1984).

Results

Pathogeenicity tests indicated that these isolates significantly varied in the root rot/wilt severity detected in pepper plants cv. Anaheim-M (Tab. 1). The highest of disease severity of infected pepper was produced by PF4 (87%) followed by PF2 (82%), while isolates FP7 and FP9 were the weak ones. Identification procedures of the isolated fungi proved the presence of 3 *Fusarium solani* isolates (PF1 to PF3) and 7 isolates of *Fusarium oxysporum* (PF4 to PF10).

Tab. 1. Pathogenicity tests of fungal isolates obtained from naturally diseased paper roots growing in different location at New Valley Governorate, Egypt

Isolates	Location	Disease index	Disease severity
FP1	Kharga	3.60	72
FP2	Kharga	4.10	82
FP3	Baris	2.05	41
FP4	Dakhla	4.35	87
FP5	Dakhla	1.30	26
FP6	Baris	2.25	45
FP7	Farfrah	0.40	8
FP8	Farfrah	1.35	27
FP9	Kharga	0.80	16
FP10	Dakhla	1.45	29
Mean		2.17	43.3

The effect of antioxidants on pepper disease severity under greenhouse conditions:

In order to study, the role of the antioxidants on pathogenic process a determination procedure was carried out using one isolate each of *Fusarium solani* (isolate PF2) and *F. oxysporum* (isolate PF4). These compounds were applied with three methods i.e seed soaking, transplanting soaking and soil drenching.

The effect of seed soaking

Soaking pepper seeds in antioxidant solutions before planting resulted in the increase of resistance pepper seedlings and old plants against to infection with the tested fungi (Tab. 2). The resistance of pepper was enhanced by increasing antioxidant concentration. Propylgalate was the most effective antioxidant as they greatly retarded damping. Soaking of pepper seeds in the chemical solution (200 ppm) of propylgalate for 12 hr. before planting reduced damping-off caused by *F. solani* and *F. oxysporum*. The infection was from 45.6 and 38.4% in control to 6.3 and 6.4%. Root rot/wilt severity was reduced from 27.4 and 46.4 % in control to 4.4 and 9.0 %.

On the other hand, pepper seeds soaking in antioxidants increase fresh and dry weight compared to untreated plants under infection with *Fusarium solani* and/or *F. oxysporum*. Fresh weight and dry weight increase by increasing of antioxidant concentration. Seeds treated with propylgalate at 100 or 200 ppm resulted in higher fresh and dry weight compared to any of the other samples which underwent treatment. The weight per fresh plant was 4.900 and 4.215 g $plant^{-1}$ in case of seed treated with propylgalate at 200 ppm we can compare the values to 1.488 and 1.354 gm/plant in control when growing in infested soil with *F. solani* and *F. oxysporum* individually respectively. Dry weight was 1.275 and 1.205 gm/plant compared to 0.256 and 0.241gm/plant in control.

Soil drenching

Drenching soil with antioxidant solutions resulted in an increase pepper resistance against infection with the tested fungi (Tab. 3 and 4). Data in (Tab. 3) indicate that all antioxidants reduce damping-off, root rot/wilt caused by *Fusarium solani* and *F. oxysporum* and increase of fresh and dry weight per plant compared with control.

Propylgalate at 200 ppm was more effective than the other antioxidants to suppress pepper damping-off and root rot/wilt. The percentage of damping-off was 4.2 and 6.0 % in case of soil drenching with propylgalate compared to 45.6 and 38.4 % under control. As for this compound it reduces root rot/wilt severity from 27.4 and 46.4% to 3.6 and 8.2%.

Tab. 2. Effect of pepper seed soaking in antioxidant solutions on damping- off and root rot/wilt caused by *Fusarium solani* isolate FP2 and *Fusarium oxysporum* isolate FP4 in pots

Treatments	Con. (ppm)	*Fusarium solani* FP2				*Fusarium oxysporum* FP4			
		% Damping-off	% Root rot/ Wilt	Fresh weight/ plant (gm)	Dry weight/ plant (gm)	% Damping-off	% Root rot/ Wilt	Fresh weight/ plant (gm)	Dry weight/ plant (gm)
Coumaric acid	100	15.0[a]	13.3	3.377	0.932	18.2	20.0	1.686	0.509
	200	11.2	8.8	3.581	1.061	11.1	13.5	2.525	0.892
	Mean	13.1	11.05	3.479	0.997	14.65	16.75	2.106	0.701
Citric acid	100	30.0	16.2	3.248	0.906	25.1	19.2	1.682	0.401
	200	18.0	14.1	3.542	0.937	20.0	16.0	2.498	0.733
	Mean	24.0	15.15	3.395	0.922	22.55	17.6	2.090	0.567
Propylgalate	100	10.0	6.0	4.615	1.113	8.3	13.2	2.666	0.625
	200	6.3	4.4	4.900	1.275	6.4	9.0	4.215	1.205
	Mean	8.15	5.2	4.7575	1.194	7.35	11.1	3.441	0.915
Salicylic acid	100	25.0	11.0	2.921	1.103	20.4	23.5	2.215	0.808
	200	15.3	9.5	3.144	1.198	13.1	14.0	2.619	0.969
	Mean	20.15	10.25	3.033	1.151	16.75	18.75	2.417	0.889
Control		17.73	27.4	1.488	0.256	38.4	46.4	1.354	0.241

LSD at 0.05 for:	Damping-off	Root rot/Wilt	Fresh weight/plant	Dry weight/plant
Treatments (A)=	3.522	0.436	0.291	0.080
Concentrations (B)=	**	**	**	**
Isolates (C)=	**	**	**	**
Interactions (AxBxC)=	5.636	ns	ns	0.140

[a]Values are means of 3 replicates

Tab. 3. Effect of soil drenching with antioxidant solutions on damping- off and root rot/wilt caused by *Fusarium solani* isolate FP2 and *F. oxysporum* isolate FP4 in pots

Treatments	Con. (ppm)	*Fusarium solani* isolate FP2				*Fusarium oxysporum* isolate FP4			
		% Damping-off	% Root rot/ Wilt	Fresh weight/ plant (gm)	Dry weight/ plant (gm)	% Damping-off	% Root rot/ Wilt	Fresh weight/ plant (gm)	Dry weight/ plant (gm)
Coumaric acid	100	12.4[a]	10.3	4.141	1.325	15.1	16.0	2.961	0.619
	200	8.4	7.2	4.861	1.610	9.3	10.2	2.910	0.993
	Mean	10.4	8.75	4.501	1.468	12.2	13.1	2.936	0.806
Citric acid	100	25.1	12.4	3.806	1.205	23.2	16.4	1.961	0.631
	200	15.4	10.4	3.963	1.310	16.2	12.1	2.631	0.803
	Mean	20.25	11.4	3.885	1.258	19.7	14.25	2.296	0.717
Propylgalate	100	8.0	5.1	4.810	1.351	6.1	10.4	3.061	0.811
	200	4.2	3.6	5.201	1.462	6.0	8.2	5.010	1.905
	Mean	6.1	4.35	5.006	1.407	6.05	9.3	4.036	1.358
Salicylic acid	100	26.4	13.5	3.410	1.061	14.6	20.1	2.41	0.905
	200	13.1	8.1	3.610	1.135	10.4	12.3	3.162	1.108
	Mean	19.75	10.8	3.510	1.098	12.5	16.2	2.786	1.007
Control		45.6	27.4	1.488	0.256	38.4	46.4	1.354	0.241

LSD at 0.05 for:	Damping-off	Root rot/Wilt	Fresh weight/plant	Dry weight/plant
Treatments (A) =	1.031	0.370	0.149	0.620
Concentrations (B) =	**	**	**	**
Isolates (C) =	**	**	**	**
Interactions (AxBxC) =	3.634	2.732	0.280	0.123

[a]Values are means of 3 replicates

Tab. 4. Effect of soil drenching with antioxidant solutions on area under root rot/wilt progress curve caused by *Fusarium solani* isolate FP2 and *Fusarium oxysporum* isolate FP4 in pots

Treatments	Con. (ppm)	Area under root rot/wilt progress carve caused by			
		Fusarium solani isolate FP2		*Fusarium oxysporum* isolate FP4	
		AURR/WPC	% Protection	AURR/WPC	% Protection
Coumaric acid	100	440.4[a]	54.13	483.3	54.09
	200	243.9	74.59	248.4	76.40
	Mean	342.15	64.36	365.85	65.25
Citric acid	100	459.5	52.14	612.0	41.86
	200	348.7	63.68	399.7	62.03
	Mean	404.1	57.91	505.85	51.95
Propylgalate	100	320.9	66.57	327.3	68.91
	200	192.0	80.00	180.0	82.901
	Mean	256.45	73.29	253.65	75.901
Salicylic acid	100	483.6	49.63	483.1	54.11
	200	356.0	62.93	284.0	73.02
	Mean	419.8	56.27	383.55	63.57
Control		960.0	-	1052.7	-

LSD at 0.05 for: Treatments (A) = 27.151; Concentrations (B) = **; Isolates (C) =**; Interactions (AxBxC) = 129.208; [a]Values are means of 3 replicates

The fresh and dry weight were increased from 1.488, 1.354 g FW plant^{-1} in control to 5.201, 5.01 g plant^{-1} and 0.256, 0.241g DW plant^{-1} in control to 1.462, 1.505 g DW plant^{-1} in PG treatment.

On the other hand, soil drenching with antioxidants protect of pepper seedling against infection with tested fungi (Tab. 4). The effect of antioxidants was enhanced by increasing antioxidant concentration. Using Propylgalate solution at 200 ppm proved to reduce the area under root rot/wilt progress curve (AURR/PC) caused by *F. solani* and *F. oxysporum* (80, 82.91% protection), followed by coumaric acid at 200 ppm (74.95 and 76.40% protection). Soil drenching with citric acid at 100 ppm was less effective (52.14 and 41.86% protection).

Seedling soaking

Pepper seedling (40 day-old) soaking in antioxidant solutions reduced AURR/WPC caused by *F. solani* and *F. oxysporum* compared to untreated seedling (Tab. 5). Increasing antioxidant concentration enhanced pepper resistance. The lowest area under root rot/wilt progress carve was provided by PG at 200 ppm (212.1 and 207.3 PG treatment at 200 ppm in case of *F. solani* and *F. oxysporum*, rrespectively compared with 960 and 1052.7 in control.) 4.215 g plant^{-1} in case of seed treated with propylgalate at 200 ppm compared with 1.488 and 1.354 g plant^{-1} in control when growing in infested soil with *F. solani* and *F. oxysporum* individually respectively. Also, dry weight was 1.275 and 1.205 g plant^{-1} compared with 0.256 and 0.241 g plant^{-1} in control.Generally, seed seedling and/or soil drenching with propylgalate increased of the resistance to infection with tested and gave highly fresh and dry weight of pepper seedlings.

The effect of tested antioxidants on the fungal growth:

The effect of the tested antioxidants on linear growth, dry weight and spore formation of the isolated fungi was carried out. Data in (Tab. 6) show that all the tested antioxidants have little effect on linear growth and dry weight of both fungal isolates. On the other hand, all tested antioxidants greatly inhibit the spore formation at the two tested concentrations (100 and 200 ppm). The effect of chemicals increase by increasing the concentration. Propylgalate was the highest inhibitory effect on spore formation for both tested fungal isolates (15.60 and 21.7 x 10^5 compared with 83.0 and 68.4 x 10^5 in control).

Effect of antioxidants on pepper disease severity under field conditions:

Under field conditions, the tested antioxidants were applied with seedling soaking and/or soil drenching.

Soil drenching

Significant variants between antioxidants were tested for controlling pepper root rot/wilt diseases in both seasons under field conditions (Tab. 7). Every antioxidants used was significantly reduced on the area under root rot / wilt progress curve. Propylgalate was the highest efficient in reducing AURR/WPC followed by salicylic acid especially at higher tested concentration (200 ppm). The protection of pepper plant were 75.79 and 76.19% in both seasons, respectively when soil was drenched with propylgalate at 200 ppm. On the other hand, soil drenching with citric acid recorded the lowest protection against root rot/wilt (35.60 and 33.56 % protection at 100 ppm in both seasons, respectively).

Tab. 5. Effect of paper seedling soaking in antioxidant solutions on area under root rot/wilt progress curve caused by *Fusarium solani* isolate FP2 and *Fusarium oxysporum* isolate FP4 in pots

Treatments	Con. (ppm)	Area under root rot/wilt progress carve caused by			
		Fusarium solani FP2		*Fusarium oxysporum* FP4	
		AURR/WPC	% Protection	AURR/WPC	% Protection
Coumaric acid	100	624.0[a]	35.00	627.2	40.42
	200	368.1	61.66	392.8	62.69
	Mean	496.05	48.33	510.0	51.55
Citric acid	100	740.0	22.92	744.0	29.32
	200	608.1	36.66	488.0	53.64
	Mean	674.05	29.79	616.0	41.48
Propylgalate	100	392.6	59.10	567.6	46.08
	200	212.1	77.91	207.3	80.31
	Mean	302.35	68.52	387.45	63.19
Salicylic acid	100	728.2	24.15	711.0	32.46
	200	404.6	57.85	327.3	68.91
	Mean	566.4	41.00	519.15	50.68
Control		960.0	-	1052.7	-

LSD at 0.05 for: Treatments (A)= 29.250; Concentrations (B)= **; Isolates (C)=**; Interactions (AxBxC)=120.451[a]Values are means of 3 replicates

Seedling soaking

Data in (Tab. 8) show that all the tested antioxidants significantly reduced the area under disease progress curver compared to control treatment. The effect of antioxidant increased by increasing in concentration In this respect propylgalate was the highest efficient in reducing AURR/WPC compared to the other antioxidants. The AURR/WPC was recorded 315.4 and 345.2 in case of PG at 200 compared to 1450.60 and 1545.50 in control in both seasons, respectively. Seedling soaked in SA came next PG where it decreased of AURR/WPC from 1450.6 and 1545.50 in control to 399.2 and 355.2 in SA treatment in both seasons, respectively. Pepper seedling sacking in citric acid was recorded as the lowest protection to root rot/wilt diseases in both seasons, it was recorded by 835.0 and 615.4 at 100 and 200 ppm in season 2007-2008 compared

Tab. 6. Effect of antioxidant compounds on dry weight, linear growth and spore formation of *Fusarium solani* isolate FP2 and *Fusarium oxysporum* isolate FP4 grown on Czapek's medium

Treatments	Con. (ppm)	Dry weight (mg/ 50 ml medium)			Linear growth (mm)			Sporulation X 10^5/cm^2		
		FP2	FP4	Mean	FP2	FP4	Mean	FP2	FP4	Mean
Coumaric acid	100	397.7[a]	374.0	385.85	86.3	88.0	87.15	29.6	34.3	31.95
	200	381.0	363.3	372.15	83.6	88.3	85.95	18.6	29.5	24.05
	Mean	389.35	368.65	379.0	84.95	88.15	86.55	24.1	31.9	28.00
Citric acid	100	410.0	337.1	373.55	89.3	87.7	88.5	30.4	47.9	39.15
	200	388.0	328.4	358.2	85.7	87.8	86.75	29.8	35.6	32.70
	Mean	399.0	332.75	365.86	87.5	87.75	87.63	30.1	41.75	35.93
Propylgalate	100	386.3	374.3	380.3	82.8	81.6	82.2	18.5	22.0	20.25
	200	364.0	353.8	358.9	80.0	82.0	81.0	15.6	21.7	18.65
	Mean	375.15	364.05	369.6	81.4	81.8	81.6	17.05	21.85	19.45
Salicylic acid	100	404.7	353.6	379.15	88.2	86.7	87.45	37.8	33.6	35.70
	200	405.8	347.2	376.5	86.7	87.3	87.0	32.8	23.3	28.05
	Mean	405.25	350.4	755.65	87.45	87.0	87.23	35.3	28.45	31.86
Control		408.4	373.6	391.0	90.0	90.0	90.0	83.0	68.4	75.70

LSD at 0.05 for:	Dry weight	Linear growth	Sporulation
Treatments (A)=	5.431	3.069	1.768
Concentrations (B)=	**	**	**
Isolates (C)=	**	ns	**
Interactions (AxBxC)=	11.736	ns	5.076

[a]Values are means of 3 replicates

Tab. 7. Effect of pepper seedling soaking in antioxidant solutions on area under root rot/wilt progress curve during 2007-2008 and 2008-2009 growing seasons under field condition at New Valley, Egypt

Treatments	Con. (ppm)	Area under root rot/wilt progress carve at season			
		Season 2007-2008	% Protection	Season 2008-2009	% Protection
Coumaric acid	100	815.2[a]	43.80	951.4	38.44
	200	524.3	63.86	697.2	54.89
	Mean	669.75	53.83	824.3	46.66
Citric acid	100	934.2	35.60	1025.4	33.65
	200	747.0	48.50	829.4	46.33
	Mean	840.6	42.05	927.4	39.99
Propylgalate	100	437.9	69.81	481.5	68.85
	200	351.2	75.79	368.0	76.19
	Mean	394.55	72.80	424.75	72.52
Salicylic acid	100	641.4	55.78	628.2	59.35
	200	415.4	71.36	401.7	74.01
	Mean	528.4	63.57	514.95	66.68
Control		1450.60	-	1545.50	-

LSD at 0.05 for: Treatments (A)= 52.551; Concentrations (B)= **; Seasons (C)=**; Interactions (AxBxC)= 97.222; [a]Values are means of 3 replicates

Tab. 8. Effect of soil drenching with antioxidant solutions on area under root rot/wilt progress curve during 2007-2008 and 2008-2009 growing seasons under field conditions at New Valley, Egypt

Treatments	Con. (ppm)	Area under root rot/wilt progress carve at season			
		Season 2007-2008	% Protection	Season 2008-2009	% Protection
Coumaric acid	100	525.4	63.78	615.2	60.19
	200	368.2	74.62	525.7	65.99
	Mean	446.8	69.20	570.45	63.09
Citric acid	100	835.0	42.44	991.3	35.86
	200	615.4	57.58	608.4	60.63
	Mean	725.2	50.01	799.85	48.25
Propylgalate	100	395.8	72.71	425.8	72.45
	200	315.4	78.26	345.2	77.665
	Mean	355.6	75.49	385.5	75.06
Salicylic acid	100	525.7	63.76	508.4	67.10
	200	399.2	72.48	355.2	77.02
	Mean	462.45	68.12	431.8	72.06
Control		1450.60	-	1545.50	-

LSD at 0.05 for: Treatments (A)= 58.321; Concentrations (B)= **; Seasons (C)=**; Interactions (AxBxC)= 102.564; [a]Values are means of 3 replicates

with 1450.6 in control and 991.3 and 608.4 compared to 1545.5 in control.

The effect of antioxidants on pepper vigor and yield under field conditions:

Seedling soaking and/or soil drench with any of these antioxidants were submitted to various growth parameters i.e. plant height, branch number, pod number/plant, and pod yield/plant. The data in Tab. 9 show that seedlings soaking in antioxidants offer interesting results as they markedly enhanced the physiological activities of these plants especially at high concentrations (200 ppm). Propylgalate achieved the highest increase in all the mentioned parameters over the other entire three antioxidants ions durring both seasons. The average plant length of untreated seedling was 34.44 and 36.1 cm/plant in control, it recorded 65.44 and 61.40 cm/plant in PG treatment at 200 ppm in both seasons, respectively. The number of branches was 7.84 and 7.90 recorded in PG at 200 ppm compared to 5.07 and 5.00 branches/plant in control.

Soil drenching with antioxidants significantly increase all parameters compared to control (Tab. 10). Soil drenching with PG gave the highest results in all parameters while it recorded 69.27 and 68.90 cm/plant, 7.84 and 7.24 branch/plant, 30.60 and 33.40 pod/plant and 437.92 and 474.31gm/plant compared to 34.44 and 36.10 cm/plant, 5.07 and 5.0 branch/plant, 15.05 and 16.90 pod/plant, 248.52 and 308.74 gm/plant in control in both seasons, respectively. On the other hand, salicylic acid came next PG in both applied methods (seedling soaking or soil drench). However, citric acid came at the end compared with the other treatments.

Tab. 9. Effect of antioxidant solutions used as seedling soaking on pepper vigor plants (cv 'Anaheim-M') and pod yield/plant under field condition in season 2007-2008 and 2008-2009

Treatments	Con. (ppm)	Plant height (cm)		No. of branch/plant		No. of pod/plant		Pod yield (gm)/plant	
		2007-2008	20082009	2007-2008	2008-2009	2007-2008	2008-2009	2007-2008	2008-2009
Coumaric acid	100	35.67[a]	36.42	5.27	5.15	17.24	17.30	341.11	352.51
	200	38.48	38.83	5.54	5.50	21. 02	20.80	394.10	412.60
	Mean	37.08	37.63	5.41	5.33	19.14	19.05	367.64	382.56
Citric acid	100	43.59	40.2	6.87	6.74	21.16	23.20	386.96	369.20
	200	44.90	45.84	7.43	7.20	22.76	24.30	427.84	418.20
	Mean	44.25	43.02	7.15	6.97	21.96	23.75	407.40	393.70
Propylgalate	100	48.58	49.81	6.99	7.32	21.90	25.40	429.66	455.21
	200	65.44	61.40	7.84	7.90	29.74	32.60	465.50	486.25
	Mean	57.01	55.61	7.42	7.61	25.82	29.00	447.58	470.73
Salicylic acid	100	42.23	46.35	6.17	6.22	20.20	20.42	351.40	386.20
	200	56.67	53.20	6.77	6.80	28.52	28.62	463.96	439.17
	Mean	49.45	49.78	6.47	6.51	24.36	24.52	407.68	412.69
Control		34.44	36.1	5.07	5.00	15.05	16.90	248.52	308.74

LSD at 0.05 for	Plant height	No. of branch/plant	No. of pod/plant	Pod yield (gm)/plant
Treatments (A)=	2.574	0.232	0.235	16.937
Concentrations (B)=	**	**	**	**
Seasons (C)=	ns	ns	**	**
Interactions (AxBxC)=	ns	0.409	ns	34.208

[a]Values are means of 3 replicates

Tab. 10. Effect of antioxidant solutions used as soil drenching on pepper vigor plants (cv 'Anaheim-M') and yield/plant under field condition in season 2007-2008 and 2008-2009

Treatments	Con. (ppm)	Plant height (cm)		No. of branch/plant		No. of pod/plant		Pod yield (gm)/plant	
		2007-2008	2008-2009	2007-2008	2008-2009	2007-2008	2008-2009	2007-2008	2008-2009
Coumaric acid	100	38.55	36.20	5.27	5.20	17.10	17.40	345.31	362.40
	200	39.26	42.80	5.54	5.31	20.86	21.60	408.52	421.50
	Mean	38.91	39.50	5.41	5.26	18.98	19.50	376.95	391.95
Citric acid	100	46.71	41.54	6.87	6.80	18.62	20.10	330.61	364.10
	200	51.19	49.30	7.43	6.60	19.10	21.70	403.06	409.51
	Mean	48.95	45.42	7.15	6.70	18.86	20.90	366.87	386.81
Propylgalate	100	46.13	50.31	6.99	7.20	23.42	28.40	401.87	454.80
	200	69.27	68.90	7.84	7.24	30.60	33.40	437.92	474.31
	Mean	57.70	59.61	7.42	7.22	27.0	30.90	419.93	464.56
Salicylic acid	100	43.44	48.23	6.17	5.60	22.84	24.30	337.40	378.20
	200	59.38	57.40	6.77	6.48	27.96	26.80	436.80	429.82
	Mean	51.41	52.82	6.47	6.04	24.40	25.55	387.10	404.01
Control		34.44	36.1	5.07	5.00	15.05	16.90	248.52	308.74

LSD at 0.05 for:	Plant height	No. of branch/plant	No. of pod/plant	Pod yield (gm)/plant
Treatments (A)=	2.574	0.232	0.235	16.937
Concentrations (B)=	**	**	**	**
Seasons (C)=	**	ns	*	**
Interactions (AxBxC)=	ns	0.449	ns	34.208

[a]Values are means of 3 replicates

Discussion

Pepper plants are subject to infection with many diseases (Lamb *et al.,* 1999; Utkhede and Mathur, 2005 and Demirci and Dolar, 2006), among which the soil-borne diseases are the most important. Many fungi i.e *Fusarium*, M*acrophomina, Rhizoctonia, Pythium, Verticilium* and *Sclerotinia* .causing damping-off, root rot and wilt diseases are commonly encountered in the greenhouse, nurseries and fields (Lu *et al.,* 1984; Fletcher, 1994; Soner Soylu *et al.,* 2005 and Goicoechea, 2006).

Results of the present study showed that fungi isolated from naturally rotted roots and wilt foliage of pepper plants collected from four locations in New Valley Governorate, Egypt (Gharga, Farafrah, Dakhla and Baris), represented 2 species, belonging to *Fusarium genus* (*F. solani* and *F. oxysporum*). *Fusarium oxysporum* isolate PF4 was highly pathogenic towards pepper plant caused 87% disease severity followed by *F. solani* isolate FP2 which caused 82% disease severity. Data are in accordance with those reported by Abada (1994), Mushtaq and Hashmi (1997), Sahi and Khalid (2007).

Results indicated that all tested antioxidants reduced damping-off, root rot/wilt and the area under root rot/wilt progress curve caused by artificial infection with the tested fungi, when applied any application methods (seed soaking, seedling soaking and/or soil drenching).

Generally, the pest control was the tested solutions which was applied as soil drenching with high concentration (200 ppm) for the tested antioxidants. Propylgalate was better than the other antioxidants to reduced damping-off, root rot/wilt and the area under root rot/wilt progress curve caused by any tested fungi. Similar results were reported by Galal and Abdou (1996) who found that application of salicylic or ascorbic acid as a soil drenching was better than foliar application to control fusarial diseases of cowpea. Also, Mostafa (2006) reported that soaking cumin seeds or soil drenching with antioxidant solutions (salicylic, ascorbic, coumaric, benzoic acids, and propylgalate) before planting resulted in resistant cumin seedlings against infection with the *Fusarium oxysporum cumini* and *Acremonium egyptiacum*. Abdel-Monaim (2008) showed that soaking lupine seeds in antioxidant solutions (reduced the damping-off and root rot diseases caused by *Fusarium solani* and *Macrophomina phaseolina*. *In vitro* all antioxidants show less effect on dry weight. Propylgalate had the highest effect on spore formation especially highly concentration (200 ppm).

The antioxidants mode of action was reported in many host-pathogen interactions i.e. many oxidative enzymes such as peroxidase, catalase, ascorbate oxidase and polyphenol oxidase were detected as a result of infection with many pathogens (Clark *et al.,* 2002) or as a result of treatments with various antioxidants (Takahama and Oniki, 1994, El-Khallal, 2007b and Abdel-Monaim, 2008). Moreover, Lyon and McGill (1989) reported that the phenolic acids benzoic, ferulic, coumaric and protocatechoic acid inhibit *in-vitro* activity of polygalacturonase and polygalacturonic acid lyase from *Erwinia carotovora***.** Chen *et al.,* (1993) reported that SA binds to, or inhibits catalase.

Also in the present study shows that all tested antioxidants reduced the area of the root rot/wilt under field conditions and increased the plant vigor and pod yield per plant in both seasons. The increase in pepper yield may be due to the role of antioxidants in stimulation of physiological processes which reflect the improving vegetative growth that followed by active translocation of the photoassimilation. In this respect, antioxidants might be right being regulating plant growth by increasing enzyme activity as α-amylase and nitrate reductase, which accelerate the sugar translocation from the leaves to developing fruit (Sharma *et al.,* 1986). In addition, application of SA inhibits ethylene production leading to an increase in fruit number and consequently increases fruit yield per plant (Leslie and Romani, 1986). Abdel-Monaim (2008) found that lupine seed soaking in antioxidant solutions increases of chlorophyll and carotenoids content in leaves and this reflects the health condition of the plant.

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