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Inhibition of Carpogenic Germination of Sclerotia of *Sclerotinia sclerotiorum* (Lib.) De Bary by Cinnamic Acid Derivatives

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Cinnamic acid derivatives, *i.e. p*-coumaric and ferulic acids, significantly affected carpogenic germination of *Sclerotinia scelerotiorum* with different soil types. A complete inhibition of carpogenic germination was pronounced at 100 ppm *p*-coumaric acid especially with sandy soil (90% sand content). Ferulic acid did not affect carpogenic germination but it reduced number of stipes and number of apothecia per sclerotium. Both *p*-coumaric and ferulic acid caused malformation toward apothecial formation.

Both *p*-coumaric and ferulic acids had insignificant effect toward seed germination of cantaloupe, cucumber and watermelon even at the highest concentrations (200µg/g soil) tested. Plant infection by *S. sclerotiorum*, was greatly affected by sand contents, phenolic acids amendment and plant species. High sandy soil was the most favorable for disease development than low sand content soil. *Para*-coumaric acid was more effective to reduce disease incidence than ferulic acid. Cucumber showed high acquired resistance under soil amendment with either *p*-coumaric or ferulic acids against *S. sclerotiorum* infection than cantaloupe or watermelon.

Key words: Cantaloupe (*Cucumus melo* var. *cantaloupensis*), cucumber (*Cucumus sativus*), watermelon (*Citrillus vulgaris*), *p*-coumaric acid and ferulic acid

Diseases caused in economically important plants by *Sclerotinia sclerotiorum* (Lib.)de Bary and closely related species occur worldwide, cause considerable damage, typically have been unpredictable and difficult to control culturally or chemically (Lumsden, 1979). Apothecia formation is a critical stage in the life cycle of *Sclerotinia sclerotiorum* because of the importance of ascospore inoculum in the initial infection on host plants (Casale and Hart, 1986). Carpogenic germination and the number of stipes produced per sclerotium were affected by several external influences such as fungicides (Hawthorne and Jarvis, 1973), herbicides (Casale and Hart, 1986, and Huang and Blackshaw, 1995), calcium cyanamide (Wu, 1991), S-H mixture (Huang and Sun, 1991) and volatile substances from urea (Huang and Janzen, 1991). Singh *et al.* (1991) reported that physico-chemical properties and ion exchange capacity of soil samples influenced apothecial germination of *S. sclerotiorum*.

Higher plants regularly release organic compounds by volatilization from their surfaces and through leaf leachates and root exudates. Decomposition products are

often added to the soil and some of these have been reported as agents of plant-plant interactions, a phenomenon termed allelopathy (Rice, 1984 and Baleroni *et al.*, 2000). Phenolic acids are some allelochemicals commonly found in plants and soils and are products of lignin degradation (Einhellig, 1995). The biological activity of different allelochemicals comprises various magnitude orders (Lambers *et al.*, 1998). Ramakrishna *et al.* (1989) and Sathiyamoorthy (1990) reported that soybean root residues, aerial parts and seeds contain cinnamic acid derivatives such as ferulic and *p*-coumaric acids that may influence the germination and development of crop cultivated later.

To the best of our knowledge, no research has been reported on the effects of cinnamic acid derivatives such as *p*-coumaric and ferulic acids on either plant pathogens or on the host-pathogen interaction especially cucurbits-*S. sclerotiorum* interaction. Because these chemicals are exuded by several plant roots and other plant residues and mixed with soil we expected that such chemicals may play a vital role on the host/soil-borne interaction. Thus, the present work was planned to study the effect of soil types amendment with various concentrations of cinnamic acid derivatives, *i.e. p*-coumaric and ferulic acids on the carpogenic germination, stipes production and apothecial formation from sclerotia of *S. sclerotiorum*.

Materials and Methods

The pathogen:

Sclerotium sclerotiorum was isolated by the removal of sclerotia from the rotted stems of naturally infected cantaloupe plants cv. Shahd-Dokki (Fig. 1a) grown under low tunnels in Minia region in February 2000 (winter). The sclerotia were surface sterilized by immersion in 70% ethanol for 1 min followed by immersion in 0.5% NaOCl for 2 min then washed 3 times with sterile distilled water. Sclerotia were blotted dried in a laminar flow cabinet, then plated onto PDA medium amended with 250 ppm chloramphenicol and incubated in the dark at 20°C for 10 days. Cultures were examined daily for fungal growth. Pure cultures were maintained on PDA slants and stored at 4°C for use throughout this study. The pathogen was identified as *Sclerotinia sclerotiorum* using the key of Kohn (1979).

Pathogenicity tests:

The isolates obtained of *S. sclerotiorum* were subjected to pathogenicity tests. Disks of 7-day-old cultures grown on PDA were transferred individually to autoclaved barley grains (100 g barley grains mixed with 40 g sand and 60 ml distilled water in 300-ml Erlenmeyer flask) and incubated for 10 days at 20°C. Then the inoculated grains were mixed with steam sterilized soil at rate of 100gm infested grains per Kg soil and at rate of 2 kg. per pot (20-cm diameter). Control pots were filled with mixture of non-infested barley grains and autoclaved soil. All pots were irrigated and kept under the greenhouse condition. Seven days later, pots were cultivated by cantaloupe seeds cv. Shahd-Dokki which had been surface sterilized. Ten seeds were cultivated per pot, four pots were used per replicate and each treatment contained 3 replicates. At 10, 20 and 40 days after planting, percentages of pre- and post-emergence damping-off and dead plants were assayed, respectively.

Effect of p-coumaric and ferulic acids on the carpogenic germination in vitro:

Disks of 7-day-old cultures of *S. sclerotiorum* isolate SS3 (the highly pathogenic isolate) grown on PDA were transferred to autoclaved barley grains (100 g barley grains mixed with 40 g sand and 60 ml distilled water in 300-ml Erlenmeyer flask) and incubated at 20°C. After 30 days, the new sclerotia were formed. Sclerotia were sloughed from barley grains and transferred immediately (under aseptic conditions) to autoclaved soil in 16-cm Petri plates.

The soils used in this study were natural soils with the following physical properties: Soil A-sand 8%, silt 55%, and clay 37%and soil B-sand 90%, silt 6% and clay 4% (El-Ganieny *et al.*, 1997). Appropriate mixture of the two soils were blended to obtain soils with sand contents desired for the experiments, *i.e.* 8% (original soil A), 50% and 90% sand contents (original soil B) according to the International Pipette Method (Piper, 1950). Additionally, the role of *p*-coumaric and ferulic acids amendment to soil texture was investigated at 5 various rates, *i.e.* 0.0 (distilled water served as control), 10, 50, 100 and 200 µg/g soil. The test solutions of *p*-coumaric or ferulic acids were prepared singly (either *p*-coumaric or ferulic acids were dissolved in boiled distilled water for 10 min.) and added to soil for obtaining the final concentrations (each plate received 100 ml solution).

To each Petri plate, 20 sclerotia were set on the surface of soil and 5 plates were used as replicates for each treatment. After 35 days incubation at 15°C under darkness condition, the plates were placed under fluorescent light (2800 Lux, 14 hr photoperiod) at 15°C for 18 days (Casale and Hart, 1986). Thereafter, the sclerotia in each replicate were washed 3 times by distilled water then placed in a Petri plate (9-cm diameter) with 15 ml distilled water. The water was replaced with fresh distilled water after 24, 48, 96 hr, and at 3 days intervals till the end of experiment. All sclerotia were incubated under fluorescent light at 15°C. After 28 days, percentages of carpogenic germination, number of stipes / sclerotium and number of apothecia / sclerotium were assessed.

Effect of p-coumaric and ferulic acids on seed germination, disease incidence and carpogenic germination of Sclerotinia sclerotiorum in vivo:

Test solutions of *p*-coumaric and ferulic acids were added, as soil drenching, to steam sterilized soils (different textures as before) potted in 20-cm diameter sterilized pots, to obtain a final concentrations of 0.0 (control), 10, 50, 100 and 200 µg/gm soil. Pots were mixed with *S. sclerotiorum* infested barley grains, which was prepared as described above, at rate 5%. Non-infested barley grains were added to serve as control. All pots were irrigated and kept under the greenhouse condition. Seven days later, pots were cultivated by seeds of cantaloupe (*Cucumis melo* var. *cantaloupensis* cv. Shahd-Dokki), cucumber (*Cucumis sativus* L. cv. Medina) and watermelon (*Citrillus vulgaris* L. cv. Giza 1), which had been surface sterilized. Ten seeds were cultivated per pot, four pots were used per replicate and each treatment contained 3 replicates. After 10, 20 and 40 days of planting, percentages of pre- and post-emergence damping-off and dead plants were assayed, respectively. After 50 days of planting, number of apothecia/plant was assayed.

Statistical analysis:

Data were transformed from percentages to arcsines when if needed and then subjected to statistical analysis to determine the standard deviation (SD) or the least significant differences (LSD) between treatments at a 0.05 confidence (Gomez and Gomez, 1984).

Results and Discussion

Pathogenicity test:

Four fungal isolates (designated from SS1 to SS4) were isolated from the naturally infected cantaloupe plants showing basal stem rot symptoms (Table 1). Pathogenicity test indicated that all isolates were pathogenic to cantaloupe.

Isolate SS3 of *S. sclerotiorum* was the most virulent one causing the highest disease incidence (88%) followed by isolates SS1 and SS2 that gave 70% disease incidence. Isolate SS4 gave the least disease incidence (56%).

Effect of p-coumaric and ferulic acids on the carpogenic germination in vitro:

Cinnamic acid derivatives interfere to some degree with much vital plant processes, including water use, transpiration, shoot and root growth (Blum and Rebbeck, 1989). These allelochemicals affect the membrane properties into which they may be incorporated, modify its fluidity and affect certain enzymes (Baleroni *et al.*, 2000).

The present results showed that *p*-coumaric and ferulic acids had various effects toward carpogenic germination of sclerotia of *S. sclerotiorum* (Table 2). Ferulic acid had no effect where sclerotia were carpogenically germinated with 100% even at the highest concentration (200 µg/gm soil) tested. As for *p*-coumaric acid, significant reductions in the carpogenic sclerotial germination were obtained at concentrations commencing 50 µg of *p*-coumaric in soil mixed at levels of 50% and 90% sand content. Such reductions are gradiently increased especially at 90% sand content.

Number of stipes per sclerotium of *S. sclerotiorum* was greatly affected in the presence of either *p*-coumaric or ferulic acids in soil (Table 2 and Fig. 1c). Generally, *p*-coumaric acid affected greater number of stipes in comparison with ferulic acid. Efficacy of the allelochemicals varied with different sand contents, *p*-coumaric was more effective with the highest sand content in soil but ferulic acid was more effective with the lowest sand content in soil.

A substantial reduction in the number of apothecia/sclerotium of *S. sclerotiorum* was obtained with either *p*-coumaric or ferulic acids amended soil (Table 2 and Fig.1c). Increasing such acids decreased number of apothecia, *p*-coumaric was more effective than ferulic acid. Increasing sand contents decreased number of apothecia as well. A complete inhibition to apothecial formation was expressed at 100µg *p*-coumaric/g soil with high sand content in soil (50 and 90% sand content in soil). However, a low sand content in soil (8% sand) gave complete inhibition to apothecia formation with 200 µg *p*-coumaric acid. Data indicate that phenolic acids tested

Table 1. Pathogenicity test of *S. sclerotiorum* isolates obtained from rotted stems of cantaloupe (*Cucumis melo* var. *cantaloupensis* cv. Shahd-Dokki)

Isolate	Pre-emergence damping off (%)	Post-emergence damping off (%)	Dead plants (%)	Total disease (%)
SS1	10	18	42	70
SS2	14	20	38	72
SS3	8	28	52	88
SS4	10	16	30	56
LSD at 0.05	NS	2.8	4.1	5.4

Table 2. Effect *in vitro* of sand content in soil and amendment with *p*-coumaric (*p*-CA) and ferulic (FA) acids on the percentage of carpogenic germination, No. of stipes and No. of apothecia of *Sclerotinia sclerotiorum* sclerotia

Chemicals and conc. (µg/g soil)	Carpogenic germination (%) and sand contents (%)			Mean	NO. of stipes/ sclerotium and sand contents (%)			Mean	No. of apothecia / sclerotium and sand contents (%)			Mean
	8	50	90		8	50	90		8	50	90	
Distilled water	100	100	100	100	23	22.2	17.2	20.8	9.6	9.0	6.0	8.2
p-CA,												
10	100	100	100	100	17.4	16.8	14.6	16.3	8.1	8.1	8.1	8.1
50	100	90	50	80	14.4	12.8	6.4	11.2	6.0	6.6	3.3	5.3
100	95	20	5	40	7.5	2.8	0.2	3.5	3.8	0.0	0.0	1.3
200	35	10	5	16.7	4.6	1.2	0.1	2.0	0.0	0.0	0.0	0.0
Mean	82.5	55	40		11	8.4	5.8	8.3	4.5	3.7	2.9	
FA,												
10	100	100	100	100	26	25.2	25.4	25.5	6.3	6.9	6.2	6.5
50	100	100	100	100	17.3	18	24	19.8	5.8	6.9	5.8	6.2
100	100	100	100	100	13.4	17	20	16.8	4.2	4.2	4.6	4.3
200	100	100	100	100	12.8	16.4	18.2	15.8	1.8	1.8	1.2	1.6
Mean	100	100	100		17.4	19.2	21.9		4.5	5.0	4.5	
G. Mean	94.2	85	80		17.1	16.6	15		6.2	5.9	4.5	

LSD at 0.05 for:

Chemicals	(A)=	6.4	3.8	1.3
Conc.	(B)=	4.5	2.4	0.6
Sand content	(C)=	3.8	3.2	1.8
A X B X C	=	NS	NS	NS

variously affected carpogenic germination and soil texture plays an important role in this respect. A strong dependence between inoculum density and soil texture (clay/sand content) on the infection of sorghum by *Peronosclerospora sorghi* (Pratt and Janke, 1978 and Schuh *et al.*, 1987) and on the infection of onion by *Urocystis cepula* (El-Ganieny *et* al., 1997). Although ferulic acid has no effect toward sclerotial germination it has strong inhibitory effect to reduce number of stipes and inhibit apothecial formation. Moreover both *p*-coumaric and ferulic acids exhibited malformation to apothecia formed as compared to control (Fig. 1b and 1c).

Effect of p-coumaric and ferulic acids on seed germination of cantaloupe, cucumber and watermelon:

Both *p*-coumaric and ferulic acids did not affect seed germination of cantaloupe, cucumber and watermelon (Table 3) even at the highest concentrations (200µg/g soil) tested. Seed germination was the best under high sand content in soil. Cucumber seeds gave the highest percentage of germination (92%) followed by watermelon (80%) and cantaloupe (78%). The present data are consistent with the findings of Yaklich *et al.* (1979) on soybean and with El-Ganieny *et al.* (1997) on onion plants, who demonstrated that seedling emergence and stand were better on a sandy soil than on the heavy soil. Blum and Rebbeck (1989) found that ferulic acid at 0.25 mM stimulated secondary root initiation and increased the ratio root-shoot in cucumber.

Table 3. Seed germination (%) cantaloupe, cucumber and watermelon as influenced by amendment of soil at different contents % of sand with different concentrations of *p*-coumaric (*p*-CA) and ferulic (FA) acids

Chemicals And conc. (µg/g soil)	Plant species and sand contents (%)											
	Cantaloupe cv. Shahd-Dokki			Mean	Cucumber cv. Medina			Mean	Watermelon cv. Giza 1			Mean
	8	50	90		8	50	90		8	50	90	
Distilled water	78	78	78	78	92	92	92	92	80	80	80	80
p-CA,												
10	78	78	78	78	92	92	92	92	80	80	80	80
50	78	78	78	78	92	92	92	92	80	80	80	80
100	76	76	78	76.7	90	92	92	91.3	75	78	80	77.7
200	75	75	78	76	90	90	92	90.6	70	75	78	74.3
Mean	76.8	76.8	78		91	91.5	92		76.3	76.3	79.5	
FA,												
10	78	78	78	78	92	92	92	92	80	80	80	80
50	78	78	78	78	90	90	92	90.6	80	80	80	80
100	75	75	78	76	89	89	92	90	76	76	79	77
200	75	75	78	76	88	89	90	89.3	75	76	78	76.3
Mean	76.5	76.5	78		89.8	90	91.5		77.8	78	79.3	
G. Mean	77.1	77.1	78		90.9	91.2	91.8		78	78.8	79.6	

LSD at 0.05 for:				
Chemicals	(A) =	0.22	0.74	NS
Conc.	(B) =	0.34	NS	0.46
Sand content	(C) =	0.27	NS	0.35
A X B X C	=	0.84	NS	1.12

Effect of p-coumaric and ferulic acids on the disease incidence:

Data showed that the efficacy of phenolic acids toward *S. sclerotiorum* infection depending on the phenolic acid applied, soil texture and plant species tested (Table 4). Infection by *S. sclerotiorum*, was affected by sand contents, phenolic acids amendment and plant species. High sand content in soil was favorable for disease development than low sand content in soil. *Para*-coumaric acid at higher conc. was more effective to reduce disease incidence than ferulic acid. Cucumber showed lowest infection by *S. sclerotiorum* than cantaloupe and watermelon. Phenolic acids gave the best effect to reduce disease incidence with low sand content in soil and their efficacy decreased with increasing sand contents. The least disease incidence (16%) was expressed by amendment 200 µg *p*-coumaric acid/g soil of 8% sand content in soil planted with cucumber plants cv. Medina. Several researchers concerned the effects of such phenolic acids on the growth of various plants (Blum *et al.*, 1989; Lehman *et al.*, 1994; Baleroni *et al.*, 1997 and Baleroni *et al.*, 2000). But this study being the first report on plant-pathogen interaction particularly on the interaction between cucurbits and *S. sclerotiorum*.

Table 4. Disease incidence caused by *S. sclerotiorum* isolate SS3 to cantaloupe, cucumber and watermelon as influenced by amendment of soil at different contents % of sand with different concentrations of *p*-coumaric (*p*-CA) and ferulic (FA) acids

Chemicals And conc. (µg/g soil)	Plant species and sand contents (%)											
	Cantaloupe cv. Shahd-Dokki			Mean	Cucumber cv. Medina			Mean	Watermelon cv. Giza 1			Mean
	8	50	90		8	50	90		8	50	90	
Distilled water	80	80	85	81.7	85	85	88	86	80	80	80	80
p-CA,												
10	78	80	82	80	78	80	80	79.3	78	75	80	77.7
50	76	76	78	76.7	75	75	75	75	75	70	75	73.3
100	60	65	70	65	22	42	70	44.7	50	55	60	55
200	48	56	56	53.3	16	35	45	31.9	35	45	50	45
Mean	65.5	69.3	71.5		47.8	58	67.5		59.5	61.3	66.3	43.3
FA,												
10	80	80	82	80.7	76	78	78	77.3	80	85	85	83.3
50	78	78	78	78	68	61	58	62.3	60	65	65	63.3
100	72	74	74	73.3	61	56	56	57.7	46	45	50	47
200	68	70	70	69.3	40	52	56	49.3	38	38	35	37
Mean	74.5	75.5	76		61.3	61.8	62		56	58.3	58.8	
G. mean	73.3	74.9	77.5		64.7	68.3	72.5		65.2	66.5	68.4	

LSD at 0.05 for:				
Chemicals	(A) =	NS	2.10	2.87
Conc.	(B) =	3.68	3.32	4.50
Sand content	(C) =	2.85	2.58	3.5
A X B X C	=	9.0	7.98	NS

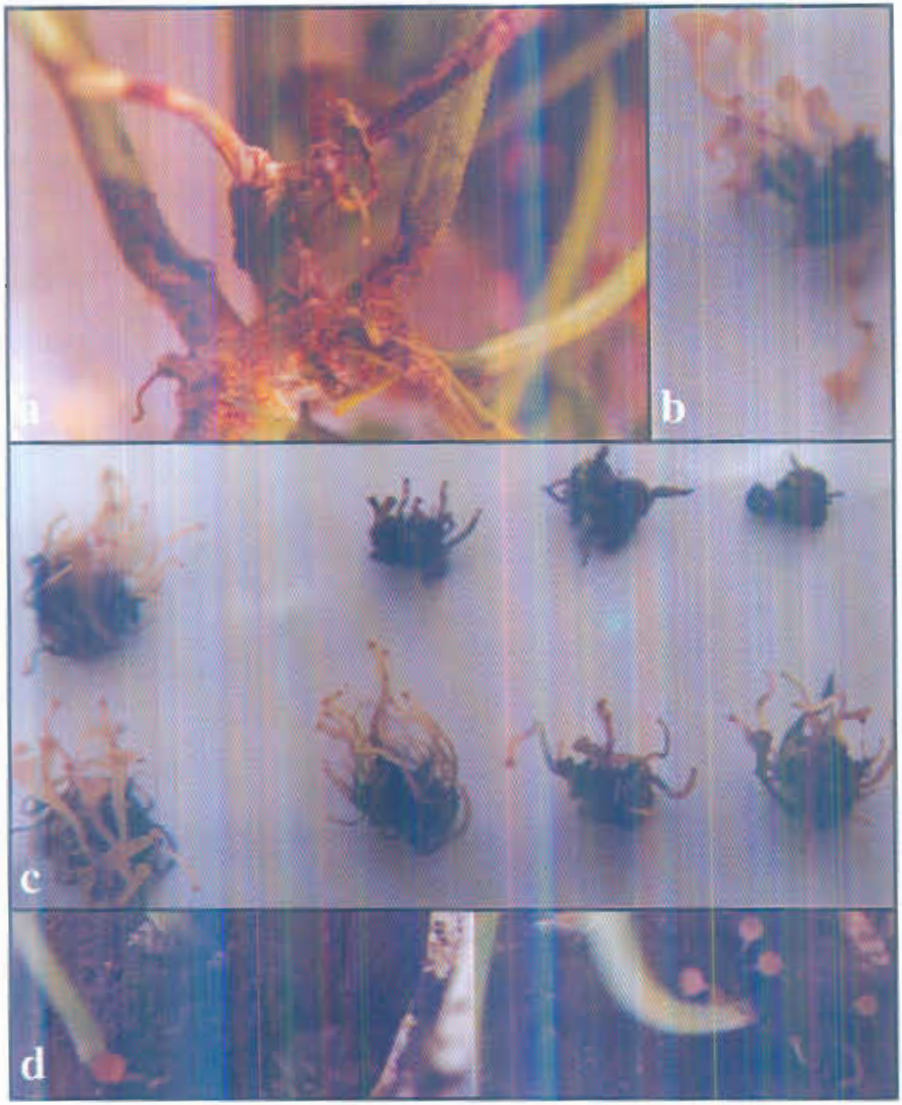

Fig. 1. a) Naturally infected cantaloupe plants (cv. Shahd-Dokki) showing basal stem rot caused by *S. sclerotiorum*.
b) Carpogenically germinated sclerotium of *S. sclerotiorum* (isolate SS3) in unamended (control) soil of 8% sand content *in vivo*.
c) Carpogenically germinated sclerotium of *S. sclerotiorum* (isolate SS3) in soil of 8% sand amended with 10, 50, 100 and 200µg (from left to right) *p*-coumaric acid (upper row) or ferulic acid (lower row) /g soil.
d) Carpogenically germinated sclerotium of *S. sclerotiorum* (isolate SS3) in presence of cantaloupe (left) and cucumber (right) plants grown in unamended soil of 8% sand content.

Effect of p-coumaric and ferulic acids on the number of apothecia/sclerotium in vivo:

Apothecial formation was variously affected by different sand content in soils, phenolic acid amendment and plant species (Table 5 and Fig. 1d). Under cantaloupe planting, sand contents have no effect toward apothecial formation while phenolic acids showed significant effect to inhibit apothecial formation. *p*-coumaric was more effective than ferulic acid, since complete inhibition to apothecial formation was pronounced at 100 and 200µg *p*-coumaric/g soil.

Table 5. Number of *S. sclerotiorum* apothecia/plant on cantaloupe, cucumber and watermelon as influenced by amendment of soil at different contents % of sand with different concentrations of *p*-coumaric (*p*-CA) and ferulic (FA) acids

Chemicals And conc. (µg/g soil)	Plant species and sand contents (%)											
	Cantaloupe cv. Shahd-Dokki			Mean	Cucumber cv. Medina			Mean	Watermelon cv. Giza 1			Mean
	8	50	90		8	50	90		8	50	90	
Distilled water	2.8	2.5	2.5	2.6	3.4	2.8	2.5	2.9	1.8	1.8	2.1	1.9
p-CA,												
10	2.5	2.4	2.4	2.4	2.5	2.1	1.1	1.9	1.6	1.8	1.9	1.8
50	1.8	1.4	1.4	1.5	2.5	2.1	0.0	1.5	0.2	0.1	0.0	0.1
100	0.0	0.0	0.0	0.0	1.1	0.6	0.0	0.6	0.0	0.0	0.0	0.0
200	0.0	0.0	0.0	0.0	0.8	0.1	0.0	0.3	0.0	0.0	0.0	0.0
Mean	1.1	1.0	1.0		1.5	1.2	0.3		0.5	0.5	0.5	
FA,												
10	2.6	2.4	2.4	2.5	2.6	1.5	0.0	1.4	1.8	2.0	2.0	1.9
50	2.1	2.4	2.4	2.3	2.1	1.2	0.4	1.2	0.0	0.0	0.0	0.0
100	0.6	0.2	0.0	0.3	0.8	0.8	0.6	0.7	0.0	0.0	0.0	0.0
200	0.0	0.0	0.0	0.0	0.3	0.6	0.6	0.5	0.0	0.0	0.0	0.0
Mean	1.3	1.3	1.2		1.5	1.0	0.4		0.5	0.5	0.5	
G. mean	1.7	1.6	1.6		2.1	1.7	1.1		0.9	0.9	1.0	

LSD at 0.05 for:				
Chemicals	(A) =	0.39	0.37	0.45
Conc.	(B) =	0.62	NS	NS
Sand content	(C) =	NS	0.46	NS
A X B X C	=	NS	NS	NS

Regarding cucumber planting, the highest number of apothecia per plant was exhibited with unamended soil (2.9 apothecia/plant) and high sand content in soil gave the lowest number of apothecia. A complete inhibition of apothecia was provided by 50 µg *p*-coumaric acid/g soil of 90% sand content in soil.

Otherwise, watermelon planting gave the lowest number of apothecia (1.9 apothecia per plant in the case of unamended soil). A complete inhibition for apothecia was shown by ferulic acid at 50 µg /g soil of different sand contents while *p*-coumaric acid at 50µg /g soil gave complete inhibition only with soil of 90% sand content.

In conclusion, cinnamic acid derivatives such *p*-coumaric and ferulic acids that considered as phenolic acids reacted as antioxidant compounds (Sert *et al.*, 1998). Thus the present study confirmed our previous studies (Galal and Abdou, 1996; Galal *et al.*, 1997; Galal *et al.*, 1999; Galal *et al.*, 2000 and Abdou *et al.*, 2001) on the effectiveness of antioxidant compounds to control different plant diseases.

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تثبيط الإنبات الثمرى للأجسام الحجرية للفطر سكليروتينيا سكليروتيورم باستخدام مشتقات حمض السينامك

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أظهرت الدراسة حدوث تأثير معنوي لمشتقات حمض السينامك (حمض الكيوماريك وحمض الفيريوليك) على الإنبات الثمرى للأجسام الحجرية للفطر سكليروتينيا سكليروتيورم المسبب لعفن الساق القاعدي في نباتات الكانتالوب صنف شهد الدقي. كان الحمض الفينولى كيوماريك اكثر فعالية عن حمض الفيريوليك في القدرة التثبيطية. تأثرت فعالية هذين الحمضين بمحتوى التربة من الرمل. التربة ذات المحتوى المنخفض من الرمل (٨% رمل) قللت من الفعل التثبيطي لهذه المركبات عند مقارنتها بالتربة الرملية (٩٠% رمل). على الرغم من ان حمض الفيريوليك لم يؤثر على الإنبات الثمرى للأجسام الحجرية الا أنة احدث انخفاض شديد فى تكوين الأجسام الثمرية. احدثا كلا الحمضين المختبرين تشوه في شكل الأجسام الثمرية المتكونة تحت ظروف المعمل.

أدت زراعة القرعيات في تربة محتوية على هذين الحمضين كلا على حده الى خفض نسبة الإصابة بالعفن الأسكليروتينى. زادت فعالية هذه المركبات فى المقاومة بزيادة التركيزات المستخدمة. تأثرت فعالية المركبات فى المقاومة بنوع التربة المستخدمة ونوع النبات المختبر. أوضحت النتائج إن حمض الكيوماريك اكثر فعالية فى المقاومة عن حمض الفيريوليك وان التربة الرملية هي الأكثر ملائمة لتطور المرض،كما ان نباتات البطيخ الأقل قابلية للإصابة. نباتات الخيار هي الأكثر اكتسابا للمقاومة ضد الإصابة بالفطر سكليروتينيا سكليروتيورم عند معاملة التربة بحمض الكيوماريك أو الفيريوليك. بينت الدراسة إن الإنبات الثمرى للأجسام الحجرية فى وجود العائل قد تأثرت بنوع النبات وتركيزات المواد المختبرة.