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EFFECTS OF PREVIOUS CROP AND INOCULUM DEPTH ON SUSCEPTIBILITY OF THE EGYPTIAN COTTONS TO *Macrophomina phaseolina*

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ABSTRACT

Fifteen winter (previous) crops were evaluated as to their effects on pathogenicity of *M. phaseolina* on cotton cultivars Giza 75, Giza 80, Giza 83, and Giza 85 under greenhouse conditions. Onion and garlic were the best performing crops in controlling *M. phaseolina*. This superiority was attributed to the following reasons: first, they were the most effective crops in suppressing *M. phaseolina* during the postemergence stage as they decreased seedling mortality by 89.89 and 85.39%, respectively, regardless of the cotton cultivar. Second, onion was one of the two most effective crops in increasing the percentage of the surviving seedlings of Giza 80,while garlic and onion were the most effective crops in increasing survival of the other cultivars. Third, garlic significantly improved plant height of Giza 80 seedlings. Garlic and onion significantly increased plant height of Giza 85 seedlings. Fourth, garlic significantly improved dry weight of Giza 80 and Giza 83 seedlings by 57.66 and 99.04%, respectively. The effect of depth (5, 10, or 25 cm) of *M. phaseolina* inoculum, originating from artificially infested sorghum grains, on charcoal rot incidence of cultivar Giza 75, were studied in30-cm-diameter clay pots. Sallow placement of inoculum was accompanied by greater isolation frequency than placement of inoculum deep in soil; furthermore, *M .phaseolina* had no detrimental effects on cotton growth up to the time of flowering when inoculum was placed at a depth of 25 cm prior to planting.

INTRODUCTION

Macrophomina phaseolina (Tassi) Goid., the causal agent of charcoal rot on cotton, is a seed-borne and soil-borne pathogen with a wide distribution (Dhingra and Sinclair, 1978). *M. phaseolina* is also a plurivorous pathogen, attacking more than 500 host species (Sinclair, 1982). When *M. phaseolina* invades roots or stems of cotton, colonization of internal tissues proceeds rapidly and the plant dies. Examination of affected plants reveals a dry rot, with many tiny black sclerotia distributed throughout the wood and softer tissues (Watkins, 1981). A negative correlation ($r = -0.85$, $p < 0.01$) was found between the disease incidence and yield (Turini *et al.*, 2000).

M. phaseolina is of a widespread distribution in the Egyptian soil and it is easily and frequently isolated from cotton roots particularly during the late period of the growing season (Aly *et al*., 1996). Resistance to *M. phaseolina* is completely lacking in the commercial Egyptian cottons (Aly *et al*., 2006).

Susceptibility of cotton, as well as other crops, to *M. phaseolina* is markedly affected by previous crop and inoculum depth and such effects are well documented in the literature.

Previous crop

Pearson *et al.* (1987) examined the effect of a 2-year crop rotation on soil and tissue populations of *M. phaseolina*. The soil population of *M. phaseolina* was lower following a maize-after-maize cropping sequence (37.5 propagules/5 g soil) than it was following a soybean-after-soybean cropping sequence (68.3 propagules/5 g soil). Quantitatively, higher levels of fungus were obtained from soybean tissue (25.6 propagules/100 mg) than from maize tissue (2.4 propagules/100 mg).

Francl *et al.* (1988) grew the soybean cultivar Forrest in monoculture and in a 2-, 3- or 4-year rotation with maize, cotton, and grain sorghum during 1980-1984. This was to determine the population dynamics of *M. phaseolina* in response to cropping history and the quantitative effect of the fungus on soybean yields. Highest population densities of *M. phaseolina* were found after soybean were grown, but often not until early spring or at planting the following year, after crop residue had decomposed. Lower densities of *M. phaseolina* occurred as soybeans appeared less frequently in rotations. Cotton, in rotation with soybean, consistently reduced the population density of *M. phaseolina* more than sorghum- and maize-soybean rotations. Soybean yield could be related to densities of *M. phaseolina* in 1984, but not in 1983. Crop rotation may be an effective method of reducing charcoal rot in soybeans, even though other crops in the rotation are hosts of *M. phaseolina*.

Rothrock and Hargrove (1988) examined the influence of winter legume cover crops and of tillage on soil population of *M. phaseolina* in the subsequent summer sorghum crop. Cover crop treatments did not consistently influence soil population of *M. phaseolina*. The population was significantly greater under no tillage.

Singh *et al.* (1990) recorded an increase in the populations of sclerotia of *M. phaseolina* when rainy season sorghum was followed by either safflower or chickpea. Cropping systems with fallow in the rainy season, followed by sorghum or chickpea in the post-rainy season, stabilized the inoculum density of *M. phaseolina*.

Arafa (1994) found that susceptibility of soybean to *M. phaseolina* was appreciably affected by the preceding winter crop. Thus, barley or lentil decreased susceptibility, while clover or broad bean increased it. On the other hand, susceptibility was not affected by wheat.

Inoculum depth

Bruton and Reuveni (1985) found that sclerotia of *M. phaseolina* were concentrated in the top 3 cm of soil regardless of host crop. In general, sclerotia were almost absent in the 40-100 cm soil depth.

Singh and Khara (1991) took samples of okra roots and soil from 4 different parts of the Indian Punjab. More fungi were isolated from the top 2 in of soil, except *M phaseolina*, which was more abundant at a depth of 2-4 in.

Singh and Kaiser (1994) reported that shallow burial of maize stalks infected with charcoal rot, to a depth of 10 cm, increased survival of *M. phaseolina* compared with material buried more deeply.

Muthukrishnan *et al.* (1995) reported that sclerotia of *M. phaseolina*, in stem and root pieces of urdbean (*Vigna mungo*), remained viable for 34 months up to 10 cm soil depth.

Kalra and Gandhi (1997) found that population levels of *M. phaseolina* in soil declined with depth, and were maximum (2.7x103 c.f.u./g soil) in the upper 5 cm.

However, due to the lack of studies, the effects of the previously mentioned factors on susceptibility of the Egyptian cottons *(Gossypium barbedense* L.) to *M. phaseolina* are unclear. Therefore, the objectives of this study were to evaluate the effects of previous crop and inoculum depth on susceptibility of the Egyptian cottons to *M. phaseolina*.

MATERIALS AND METHODS

Production of *M. phaseolina* inoculum used in soil infestation

Isolate of *M. phaseolina* used in the present study for soil infestation was obtained from the fungal collection of Cotton Disease Research Section, Plant Pathology Research Institute, Agric. Res. Cent., Giza, Egypt. This isolate was originally isolated from cotton roots.

Substrate for growth of the isolate was prepared in 500-ml glass bottles, each bottle contained 50 g of sorghum grains and 40 ml of tap water. Contents of each bottle were autoclaved for 30 minutes. Isolate inoculum, taken from one-week-old culture on PDA, was aseptically introduced into the bottle and allowed to colonize sorghum for three weeks. The mixture of sorghum and *M. phaseolina* was used for soil infestation.

Effects of previous crop on susceptibility of cotton to *M. phaseolina*

Autoclaved clay loam soil was artificially infested with *M. phaseolina* (50 g/kg of soil), and dispensed in 15-cm-diameter clay pots. In the last week of November 2001, 15 winter crops (Table 1) were planted in the infested pots. In the control treatments (infested and non-infested), pots were left without planting. There were five pots for each treatment. Pots were randomly distributed outdoors. In the end of May, winter crops were uprooted, and all pots were planted with cotton cultivars Giza 75, Giza 80, Giza 83, and Giza 85 (10 seeds per pot). Preemergence damping-off was recorded 20 days after sowing, while postemergence damping-off, survivals, plant height (cm), and dry weight (mg/plant) were recorded two months after sowing. The temperature regime during cotton-growing period ranged from 23 $\pm$ 2 to 38 $\pm$ 2.5°C.

Artificially infested field soil (40 g/kg of soil) was dispensed in 30-cm-diameter clay pots. The infested soil was also buried at a 5, 15, or 25 cm depth as a thin layer, and covered with field soil. In the control treatment, field soil was not artificially infested with *M. phaseolina*. In the middle of April, pots were planted with seeds of Giza 75 (100 seeds/pot), and pots were randomly distributed outdoors. Postemergence damping-off was recorded 15 days after planting, while postemergence damping-off and survivals were recorded 45 days after planting. After recording the data, the seedlings were thinned to ten seedlings per pot. The uprooted seedlings were used to determine plant height (cm) and dry weight (mg/plant). When the plants were 60 days old, they were thinned to five plants/pot, and dry weight (g/plant) of the uprooted plants was determined. Isolation frequency of *M. phaseolina* was also determined by plating 0.5-cm-long pieces from roots on PDA. The recommended production practices for cotton were followed during the growing season. At the end of the growing season, the following data were recorded: length (cm) of tap root, length of lesions on tap root, stem length (cm), the advancement of the lesions on stem after peeling bark off the stem, dry weight of the surviving plants (g/plant), no. of bolls/plant, seedcotton yield (g/plant), and the percentage of the dead plants.

Table 1: Plants used in studying effect of previous crop on pathogenicity of *M. phaseolina* on cotton.

Plant no.	Latin name	English name	Cultivar
C1	*Trifolium alexandrinum*	Egyptian clover	Miskawy
C2	*Trifolium alexandrinum*	Egyptian clover	Fahl
C3	*Vicia faba*	Broad bean	Giza40
C4	*Lycopersicum esculentum*	Tomato	Super Marmand
C5	*Lycopersicum esculentum*	Tomato	Castle Rock
C6	*Beta vulgaris*	Sugar beet	KWS 796
C7	*Cuminum cyminum*	Cumin	Balady
C8	*Allium sativum*	Garlic	Balady
C9	*Allium cepa*	Onion	Shandaweel
C10	*Zea mays*	Maize	SC 10
C11	*Trigonella foenum-graecum*	Fenugreek	Balady
C12	*Cicer arietinum*	Chickpea	Giza 2
C13	*Solanum tuberosum*	Potato	Nicola
C14	*Lens esculenta*	Lentil	Giza 9
C15	*Pisum sativum*	Pea	Little Marvell

Effect of inoculum depth on susceptibility of cotton to *M. phaseolina*

Statistical analysis of the data.

The experimental design of all studies was a randomized complete block with 5 replications. Analysis of variance (ANOVA) of the data was performed with the MSTAT-C Statistical Package. Duncan's multiple range test or least significant difference (LSD) were used compare treatment means. Percentage data were subjected to appropriate transformation before carrying out the ANOVA to produce approximately constant variance. Cluster analysis was performed with the software package SPSS 6.0.

RESULTS

Effect of previous crop on susceptibility of cotton to *M. phaseolina*

ANOVA (Table 2) showed that previous crop and cultivar were very highly significant sources of variation in preemergence damping-off, postemergence damping-off, survival, and plant height. In the case of dry weight, previous crop was a very highly significant source of variation, while cultivar was a nonsignificant source of variation. Previous crop x cultivar interaction was a significant or a very highly significant source of variation in all the tested parameters except postemergence damping-off.

Table 2: Analysis of variance of the effect of previous crop, cultivar, and their interaction on susceptibility of cotton to *M. phaseolina*.

Parameter and source of variation [a]	D.F.	M.S.	F. value	P > F
Preemergence damping-off				
Replication	4	367.211	3.2831	0.0119
Crop (P)	16	703.780	6.2923	0.0000
Cultivar (C)	3	4491.218	40.1547	0.0000
P x C	48	168.855	1.5097	0.0230
Error	268	111.848		
Postemergence damping-off				
Replication	4	14.734	4.7583	0.0010
Crop (P)	16	28.981	9.3592	0.0000
Cultivar (C)	3	21.022	6.7887	0.0002
P x C	48	3.685	1.1900	0.1975
Error	268	3.097		
Survival				
Replication	4	303.261	2.1989	0.0694
Crop (P)	16	1549.243	11.2332	0.0000
Cultivar (C)	3	4182.070	30.3231	0.0000
P x C	48	210.645	1.5273	0.0201
Error	268	137.917		
Plant height				
Replication	4	48.136	2.1576	0.0741
Crop (P)	16	55.813	2.5017	0.0014
Cultivar (C)	3	293.779	13.1677	0.0000
P x C	48	32.729	1.4670	0.0319
Error	268	22.311		
Dry weight				
Replication	4	1051.968	0.2559	
Crop (P)	16	31748.435	7.7218	0.0000
Cultivar (C)	3	4361.337	1.0608	0.3662
P x C	48	28559.566	6.9462	0.0000
Error	268	4111.544		

[a] Replication is random, while crop and cultivar are fixed.

Previous crop and cultivar showed almost the same relative contribution to variation in preemergence damping-off. Previous crop was the most important source of variation in postemergence damping-off and survival. Previous crop x cultivar interaction accounted for most of the variation in plant height and dry weight (Table 3).

M. phaseolina was nonpathogenic in preemergence stage on all the tested cultivars (Table 4); however when cotton was preceded by potato (crop no. 13), *M. phaseolina* caused significant increase in preemergence damping-off on Giza 80 and Giza 85. On Giza 80, a significant increase in preemergence damping-off was also found after pea (crop no. 15).

Susceptibility of cultivars to *M. phaseolina* during preemergence stage was not affected by any of the other crops.

All the tested crops were highly effective (P < 0.01) in reducing postemergence damping-off regardless of cotton cultivar; however, garlic (no. 8) and onion (no. 9) were the most effective crops in suppressing *M. phaseolina* during the postemergence stage as they reduced postemergence damping-off by 85.39 and 89.89%, respectively (Table 5).

Table 3: Relative contribution of previous crop, cotton cultivar, and their interaction to variation in preemergence damping-off, postemergence damping-off, survival, plant height, and dry weight of cotton seedlings in autoclaved soil infested with *M. phaseolina*.

Source of variation	Relative contribution [a] to variation in				
	Pre-emergence damping-off	Post-emergence damping-off	Survival	Plant height	Dry weight
Crop (P)	32.822	60.807	50.943	25.242	26.790
Cultivar (C)	39.273	8.270	25.784	24.912	0.690
P x C	23.625	23.195	20.780	44.405	72.298

[a] Calculated as percentage of sum squares of the explained (model) variation.

Table 4: Effect of previous crop, cotton cultivar, and their interaction on preemergence damping-off in autoclaved soil infested with *M. phaseolina*.

Crop	Cultivar								Mean	
	Giza 80		Giza 83		Giza 85		Giza 75			
	%	Trans-formed	%	Trans-formed	%	Trans-formed	%	Trans-formed	%	Trans-formed
Clover (1)	40.00[a]	(38.358)	44.00	(40.842)	54.00	(47.308)	34.00	(35.392)	43.00	(40.475)
Clover (2)	14.00	(19.624)	30.00	(32.068)	46.00	(42.818)	30.00	(29.954)	30.00	(31.116)
Broad bean	20.00	(25.972)	22.00	(26.266)	48.00	(43.796)	16.00	(18.00)	26.50	(28.509)
Tomato (4)	34.00	(35.442)	24.00	(28.380)	48.00	(43.796)	38.00	(37.674)	36.00	(36.323)
Tomato (5)	26.00	(29.954)	32.00	(33.642)	64.00	(53.354)	44.00	(41.438)	41.50	(39.597)
Sugar beet	44.00	(40.892)	38.00	(37.380)	48.00	(43.972)	22.00	(27.176)	38.00	(37.355)
Cumin	48.00	(42.646)	16.00	(18.470)	46.00	(42.692)	20.00	(26.266)	32.50	(32.519)
Garlic	28.00	(31.754)	16.00	(23.312)	30.00	(29.220)	12.00	(20.064)	21.50	(26.089)
Onion	22.00	(27.470)	12.00	(20.064)	22.00	(27.596)	20.00	(26.266)	19.00	(25.349)
Maize	26.00	(27.000)	22.00	(27.596)	50.00	(45.050)	28.00	(31.628)	31.50	(32.819)
Fenugreek	22.00	(26.756)	20.00	(23.018)	52.00	(46.330)	18.00	(24.222)	28.00	(30.082)
Chickpea	16.00	(20.954)	26.00	(27.176)	46.00	(42.818)	22.00	(27.596)	27.50	(29.636)
Potato	60.00	(51.466)	42.00	(40.108)	76.00	(60.780)	26.00	(29.954)	32.19	(45.577)
Lentil	38.00	(37.800)	20.00	(23.908)	64.00	(54.420)	36.00	(36.646)	39.50	(38.194)
Peas	62.00	(52.670)	36.00	(36.420)	66.00	(54.380)	30.00	(32.958)	48.50	(44.107)
Control 1	28.00	(31.460)	24.00	(28.330)	38.00	(38.026)	22.00	(27.890)	28.00	(31.427)
Control 2	26.00	(29.660)	24.00	(29.220)	50.00	(45.000)	26.00	(30.550)	31.50	(33.608)
Mean	32.59	(33.522)	26.35	(29.188)	49.88	(44.786)	26.12	(29.628)	33.74	(34.281)

LSD (transformed data) for cultivar x crop interaction = 13.14 (P $\leq$ 0.05).

[a] Percentage data were transformed into arc sine angles before carrying out the analysis of variance.

Control 1 was infested and control 2 was noninfested.

Table 5: Effect of previous crop, cotton cultivar, and their interaction on postemergence damping-off in autoclaved soil infested with *M. phaseolina*.

Crop	Cultivar								Mean	
	Giza 80		Giza 83		Giza 85		Giza 75			
	%	Trans-formed	%	Trans-formed	%	Trans-formed	%	Trans-formed	%	Trans-formed
Clover (1)	28.00[a]	(5.040)	8.00	(2.442)	18.00	(3.832)	12.00	(2.906)	16.50	(3.555)
Clover (2)	36.00	(5.770)	14.00	(3.456)	4.00	(1.690)	12.00	(2.906)	16.50	(3.455)
Broad bean	16.00	(3.370)	14.00	(3.396)	10.00	(2.416)	2.00	(1.200)	10.50	(2.595)
Tomato (4)	14.00	(3.304)	18.00	(3.828)	20.00	(3.484)	12.00	(3.194)	16.00	(3.452)
Tomato (5)	36.00	(5.902)	12.00	(2.966)	4.00	(1.690)	24.00	(4.728)	19.00	(3.821)
Sugar beet	18.00	(4.208)	18.00	(3.360)	2.00	(1.200)	8.00	(2.214)	11.50	(2.745)
Cumin	14.00	(3.684)	10.00	(2.932)	22.00	(3.824)	16.00	(3.566)	15.50	(3.501)
Garlic	12.00	(3.194)	4.00	(1.690)	4.00	(1.690)	6.00	(1.952)	6.50	(2.131)
Onion	8.00	(2.442)	6.00	(1.952)	4.00	(1.690)	0.00	(0.710)	4.50	(1.698)
Maize	18.00	(3.426)	14.00	(3.308)	18.00	(3.714)	26.00	(4.350)	19.00	(3.699)
Fenugreek	16.00	(3.718)	6.00	(1.952)	12.00	(2.906)	18.00	(3.426)	13.00	(3.000)
Chickpea	22.00	(4.814)	8.00	(2.154)	8.00	(2.442)	16.00	(3.370)	13.50	(3.195)
Potato	22.00	(4.340)	8.00	(2.670)	10.00	(2.644)	18.00	(3.828)	14.50	(3.371)
Lentil	10.00	(2.704)	14.00	(3.396)	18.00	(4.088)	24.00	(4.576)	16.50	(3.691)
Peas	10.00	(2.704)	8.00	(2.670)	8.00	(2.704)	20.00	(4.122)	11.50	(3.050)
Control 1	46.00	(6.762)	46.00	(6.674)	40.00	(6.308)	46.00	(6.740)	44.50	(6.621)
Control 2	0.00	(0.710)	0.00	(0.710)	0.00	(0.710)	0.00	(0.710)	0.00	(0.710)
Mean	19.18	(3.888)	12.24	(2.915)	11.88	(2.767)	15.29	(3.206)	14.65	(3.194)

LSD (transformed data) for crop = 1.096 ($P \leq 0.05$) or 1.444 ($P \leq 0.01$); for cultivar = 0.532 ($P \leq 0.05$) or 0.700 ($P \leq 0.01$).

[a] Percentage data were transformed into $\sqrt{x + 0.05}$ before carrying out the analysis of variance.

Control 1 was infested and control 2 was noninfested.

The data shown in Table 6 indicate that the differences in surviving seedlings between the previous crop and the infested control was not the same for each cotton cultivar-that is, cultivars responded differently to the previous crops. For examples, sugar beet (crop no. 6) was ineffective in increasing the surviving seedlings of Giza 80; however, it significantly increased the surviving seedlings of Giza 75 by 118. 75%. Lentil (crop no. 14) significantly increased the surviving seedlings of Giza 80 and Giza 83 by 136.36 and 113.33%, respectively; however, it was ineffective on Giza 85 and Giza 75. Broad bean (no. 3) and onion (no. 9) were the most effective crops in suppressing *M. phaseolina* on cultivar Giza 80, while garlic (no. 8) and onion (no. 9) were the most effective on the other cultivars.

Table 6: Effect of previous crop, cotton cultivar, and their interaction on survival in autoclaved soil infested with *M. phaseolina*.

Crop	Cultivar								Mean	
	Giza 80		Giza 83		Giza 85		Giza 75			
	%	Trans-formed	%	Trans-formed	%	Trans-formed	%	Trans-formed	%	Trans-formed
Clover (1)	40.00[a]	(39.004)	48.00	(43.796)	28.00	(31.754)	54.00	(47.358)	42.50	(40.478)
Clover (2)	50.00	(44.950)	56.00	(48.562)	30.00	(33.084)	58.00	(50.312)	48.50	(44.227)
Broad bean	64.00	(53.354)	66.00	(54.558)	42.00	(40.158)	74.00	(65.954)	61.50	(53.506)
Tomato (4)	52.00	(45.978)	58.00	(50.362)	32.00	(30.982)	50.00	(45.000)	48.00	(43.080)
Tomato (5)	38.00	(37.800)	56.00	(48.092)	32.00	(34.112)	32.00	(31.108)	39.50	(37.778)
Sugar beet	38.00	(37.380)	44.00	(38.954)	50.00	(44.824)	70.00	(56.916)	50.50	(44.518)
Cumin	38.00	(37.800)	74.00	(60.046)	32.00	(33.818)	64.00	(53.530)	52.00	(46.298)
Garlic	60.00	(50.820)	80.00	(64.028)	76.00	(60.780)	82.00	(65.358)	74.50	(60.246)
Onion	70.00	(57.042)	82.00	(65.358)	74.00	(59.996)	80.00	(63.734)	76.50	(61.532)
Maize	56.00	(49.108)	58.00	(53.530)	32.00	(33.642)	46.00	(39.462)	48.00	(43.935)
Fenugreek	62.00	(52.074)	74.00	(60.340)	36.00	(36.470)	64.00	(54.000)	59.00	(50.721)
Chickpea	60.00	(50.996)	66.00	(54.558)	46.00	(42.516)	62.00	(52.670)	58.50	(50.185)
Potato	18.00	(19.926)	50.00	(45.000)	14.00	(19.624)	56.00	(48.688)	34.50	(33.309)
Lentil	52.00	(46.104)	64.00	(57.512)	18.00	(19.154)	40.00	(38.534)	43.50	(40.326)
Peas	32.00	(33.768)	56.00	(48.562)	24.00	(28.926)	50.00	(45.176)	40.50	(39.108)
Control 1	22.00	(27.596)	30.00	(33.084)	22.00	(27.596)	32.00	(34.288)	26.50	(30.641)
Control 2	74.00	(60.340)	76.00	(60.780)	50.00	(45.000)	74.00	(59.450)	68.50	(56.392)
Mean	48.59	(43.767)	61.06	(52.184)	37.53	(36.614)	58.12	(50.090)	51.33	(45.664)

LSD (transformed data) for cultivar x crop interaction = 14.62 ($P \leq 0.05$).
[a] Percentage data were transformed into arc sine angles before carrying out the analysis of variance.
Control 1 was infested and control 2 was noninfested.

Plant height of any the tested cotton cultivars was not affected by *M. phaseolina* (Table 7). The comparisons between previous crops and the infested control within cultivars revealed that cumin (no. 7) and garlic (no. 8) significantly improved plant height of Giza 80 seedlings compared with the infested control. Garlic (no. 8) and onion (no. 9) significantly increased plant height of Giza 85 seedlings. Plant height of Giza 75 seedlings was significantly increased by fenugreek (no. 11) and lentil (no. 14). Plant height of Giza 83 was not affected by any previous crop compared with the infested control.

Dry weight of any of the tested cotton cultivars was not significantly affected by *M. phaseolina* (Table 7). The comparisons between previous crops and the infested control within cultivars showed that the effect of previous crop on dry weight varied from one cultivar to another. For example, tomato (no. 4) significantly increased dry weight of Giza 80 by 40.36%, while its effects on dry weights of the other cultivars were nonsignificant. Garlic (no. 8) significantly improved dry weight of Giza 80 and Giza 83 by 57.66 and 99.04%, respectively; however, dry weight of Giza 85 and Giza 75 were not affected by garlic. Maize (no. 10) significantly increased dry weight of Giza 85 seedlings by 56.10%, while dry weight of the other cultivars were not significantly affected by maize. Lentil (no. 14) significantly increased dry weight of Giza 83 seedlings by 125.94%, while it significantly decreased dry weight of Giza 85 seedlings by 68.71%.

Table 7: Effect of previous crop, cotton cultivar, and their interaction on plant height and dry weight in autoclaved soil infested with *M. phaseolina*.

Crop	Plant height (cm)					Dry weight (mg/plant)				
	Giza 80	Giza 83	Giza 85	Giza 75	Mean	Giza 80	Giza 83	Giza 85	Giza 75	Mean
1	28.638	20.474	22.034	27.692	24.710	395.000	337.200	387.200	319.000	359.600
2	25.566	24.894	21.516	25.812	24.447	344.600	314.200	286.000	279.800	306.150
3	23.518	22.678	22.034	25.638	23.467	272.200	257.000	389.000	328.600	311.700
4	27.534	24.062	19.092	25.248	23.984	355.400	303.000	362.200	330.800	337.850
5	29.106	23.954	26.860	27.382	26.826	391.400	377.400	313.000	324.200	351.500
6	27.396	20.078	20.274	27.996	23.936	392.800	239.600	289.800	379.000	325.300
7	31.300	24.488	22.936	26.738	26.365	400.600	401.600	318.400	379.400	374.850
8	31.360	23.286	28.892	26.314	27.463	399.200	454.200	318.800	267.000	359.800
9	26.638	25.462	29.972	28.142	27.553	357.400	450.200	353.400	391.000	388.000
10	26.576	23.380	27.066	22.660	24.920	290.600	273.600	448.000	348.800	340.250
11	28.060	23.282	24.526	30.228	26.524	339.400	345.400	629.200	351.000	416.250
12	29.562	26.852	25.094	29.594	27.775	382.800	425.600	319.200	475.000	400.650
13	21.384	24.076	20.640	25.346	22.861	331.400	287.800	263.600	309.200	298.000
14	25.094	25.842	14.634	30.244	23.953	307.400	515.600	89.800	397.400	327.450
15	23.198	26.072	21.398	23.716	23.596	325.400	288.400	273.800	395.200	320.700
Control 1	24.230	21.792	22.292	23.782	23.024	253.200	228.200	287.000	295.000	265.850
Control 2	27.094	24.104	26.150	25.946	25.823	291.800	288.600	289.000	334.600	301.000
Mean	26.838	23.810	23.259	26.616	25.131	342.918	340.447	387.200	319.000	347.391

LSD for cultivar x crop interaction = 5.882 ($P \leq 0.05$) **79.84 ($P \leq 0.05$)**
N.S. ($P \leq 0.01$) **105.20 ($P \leq 0.01$)**

Control 1 was infested and control 2 was noninfested.

1=Clover(1) 2=Clover(2) 3=Broadbean 4=Tomato(4) 5=Tomato(5) 6=Sugarbeet 7=Cumin 8=Garlic 9= Onion 10= Maize 11=Fenugreek 12=Chickpea 13= Potato 14= Lentil 15=Peas

The previous crops were highly correlated ($P < 0.01$) in their effects on susceptibility of cotton to *M. phaseolina* (Table 8).

Grouping of the previous crops based on their effects on susceptibility of cotton to *M. phaseolina* (Fig. 1) was unrelated to their taxonomic position. For example, Egyptian clover (C1), tomato (C4), fenugreek (C11), tomato (C5), and cumin (C7) were members of a clearly defined cluster, although they were belonging to leguminosae (C1 and C11), solanaceae (C4 and C5), and umbelliferae (C7). Broad bean (C3) and maize (C10) showed high level of similarity in their effects; however, broad bean is a dicotyledonous belonging to leguminosae, while maize is a monocotyledonous belonging to gramineae. A noteworthy peculiarity in the phenogram shown in Fig. 3 was the individuality of lentil (C14), which was remotely related to the other previous crops.

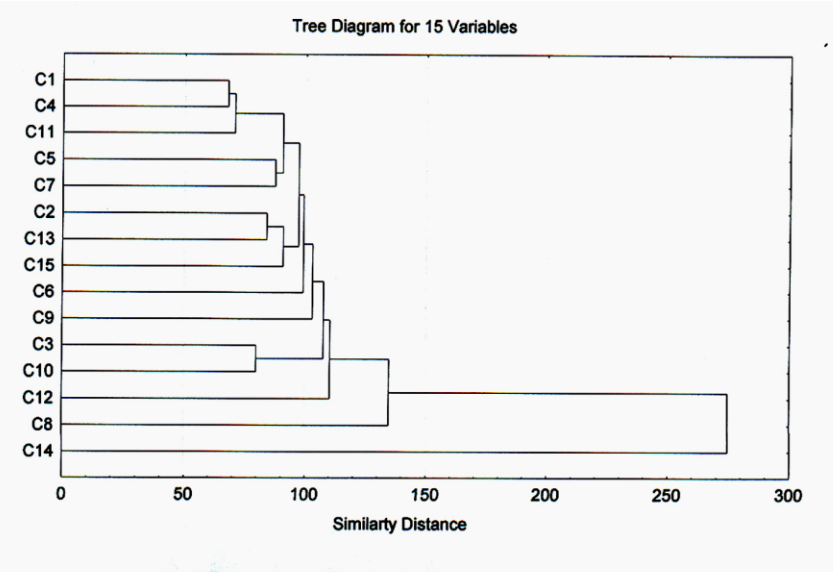

Fig. 1: Phenogram based on average linkage cluster analysis of effects of 15 previous crops on susceptibility of cotton to *M. phaseolina*. Identification of the previous crops are shown in Table 1.

Effect of inoculum depth on susceptibility of cotton to *M. phaseolina*

Plant height was not affected by the evenly distributed inoculum or the shallow inoculum, whereas it was significantly increased when the inoculum was placed at a depth of 25 or 15 cm (Table 9). Dry weight 1 was significantly reduced only when the inoculum was evenly distributed in soil, whereas dry weight 2 was significantly reduced when the inoculum was located at a depth of 15 or 5 cm. Isolation frequency was significantly increased when the inoculum was placed at a depth of 5 cm or when the inoculum was evenly distributed in soil. *M. phaseolina* significantly reduced no. of bolls when the inoculum was equally distributed in soil or placed at a depth of 15 cm. Dry weight 1 was significantly reduced only when the inoculum was evenly distributed in soil, while dry weight 2 was significantly reduced only when the inoculum was placed at the depths of 5 and 15 cm. Plant yield was significantly reduced by all treatments compared with the noninfested soil (control 1). Advancement of root lesions was significantly increased when the inoculum was evenly distributed or when it was placed at a depth of 5 cm. *M. phaseolina* did not significantly affect the other variables whether the inoculum was evenly distributed or placed at different depths.

Table 9: Effect of inoculum depth on susceptibility of cotton to *M. phaseolina*

Variable	Inoculum depth (cm)				
	25	**15**	**5**	**C 1 [a]**	**C2 [b]**
Preemergence damping-off (%)	43.00 A	44.50A	53.25A	47.00A	45.75A
Postemergence damping-off (%)	11.00A	13.25A	19.75A	24.00A	13.25A
Survival (%)	46.00A	42.50A	27.00A	29.00A	41.00A
Plant height (cm)	18.83A	17.40AB	16.60 BC	15.88BC	15.15C
Dry weight (1) (mg/plant)[c]	321.30 AB	337.50 AB	371.00 AB	282.80 B	404.00 A
Dry weight (2) (mg/plant) [d]	1.80 AB	0.91 B	1.23 B	1.81 AB	2.39 A
Isolation frequency [e] (%)	5.00 B	5.00 B	50.00 A	40.00 A	0.00 B
Stem length (cm)	94.20 A	68.89 A	111.60 A	90.05 A	96.85 A
Root length (cm)	28.35 A	21.80 A	20.80 A	21.75 A	25.00 A
Dead plants (%)	0.00 A	0.00 A	1.00 A	0.00 A	0.00 A
No. of bolls [f]	1.69 AB	1.31 B	1.69 AB	1.31 B	2.19 A
Dry weight (3) (g/plant)	15.94 AB	9.06 B	12.50 AB	12.81 AB	21.88 A
Plant yield (g)	1.28 B	1.17 B	1.21 B	0.96 B	2.19 A
Stem lesion (%) [g]	1.40 AB	0.00 B	2.63 A	2.05 AB	0.47 AB
Root lesion (%) [h]	0.00 B	1.23 B	32.98 A	36.16 A	0.00 B

Means within each row followed by the same letter(s) are not significantly different according to Duncan's multiple range test ($P < 0.05$).
[a]C1 (Control 1) = Inoculum was evenly distributed in soil.
[b] C2 (Control 2) = Noninfested soil.
[c] Dry weight 1 = Dry weight of 45-day-old plants.
[d] Dry weight 2 = Dry weight of 60-yad-old plants.
[e] Colonies of *M. phaseolina* was expressed as percentage of the total developing colnies.
[f] Dry weight 3 = Dry weight of plants at the end of growing season.
[g] [(Length of lesion on tap root (cm)/length of tap root (cm)] x 100.
[h] [(Length of lesion on stem (cm)/length of stem (cm)] x 100.

DISCUSSION

Members of the Lilaceae produce antimicrobial compounds. Garlic oil inhibited growth and sclerotia production in *Rhizoctonia solani* (Sing and Sing, 1980) and growth and spore production of 10 other fungi (Murthy and Amonkar, 1974). Agrawal (1978) reported the *in vitro* inhibitory effects of onion root and bulb extracts on the growth of several rhizosphere fungi. Parkinson and Clarke (1964) showed that the microflora levels in rhizosphere of onion and garlic were significantly lower than those of other plants. Onion bulb extract or root exudates inhibited both sclerotial germination and mycelial growth of *Sclerotium rolfsii* (Zeidan *et al.*, 1986). In the present work, the effects of 15 winter (previous) crops on susceptibility of cotton seedlings (cultivars Giza 75, Giza 80, Giza 83, and Giza 85) to *M. phaseolina* were studied in autoclaved soil. The findings demonstrated that onion and garlic were the best performing crops in controlling *M. phaseolina*. This superiority was attributed to the following reasons: first, they were the most effective crops in suppressing *M. phaseolina* during the postemergence stage as they reduced seedling mortality by 89.89 and 85.39%, respectively. Second, onion was one of the two most effective crops in increasing the percentage of surviving seedlings of Giza 80, while garlic and onion were the most effective crops in increasing survival of the other cultivars. Third, garlic significantly improved plant height of Giza 80 seedlings. Garlic and onion significantly increased plant height of Giza 85 seedlings. Fourth, garlic significantly improved dry weight of Giza 80 and Giza 83 seedlings by 57.66 and 99.04%, respectively. Therefore, in normal cropping sequence of cotton it would be desirable to introduce onion and garlic, which would be useful and inexpensive means of reducing susceptibility of cotton to *M. phaseolina*.

No information is available on the effect of *M. phaseolina* inoculum, placed at various depths, has on cotton. In the present study, shallow placement of inoculum was accompanied by greater isolation frequency than placement of inoculum deep in soil; furthermore, *M. phaseolina* had not detrimental effects on cotton growth and development up to the time of flowering when inoculum was placed at a depth of 25 cm prior to planting. According to these findings charcoal rot of cotton can be effectively controlled by plowing under the infested residues of previous crops to depths greater than 23 cm. However, this conclusion needs to be verified under field conditions. The findings of this study are in agreement with those of Singh and Kaiser (1994) who reported that shallow burial of maize stalks infected with charcoal rot, to a depth of 10 cm, increased survival of *M. phaseolina* compared with material buried more deeply.

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تأثير المحصول السابق وعمق اللقاح على قابلية الأقطان المصرية للإصابة بفطر ماكروفومينا فاسيولينا

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قيم ١٥ محصول شتوى من الشائع زراعتها قبل القطن ، وذلك من حيث تأثيرها على قدرة فطر ماكروفومينا فاسيولينا على إصابة أصناف القطن جيزة ٧٥ و جيزة ٨٠ وجيزة ٨٣ وجيزة ٨٥ ، تحت ظروف الصوبة . أظهرت الدراسة أن البصل والثوم كانا أفضل المحاصيل أداءً من حيث القدرة على مقاومة الإصابة بالفطر. يعزى هذا التفوق إلى الأسباب التالية: أولاً ، كان البصل والثوم أكثر المحاصيل قدرة على مقاومة الإصابة بالفطر بعد ظهور البادرات فوق سطح التربة ، مما أدى إلى إنخفاض النسبة المئوية للبادرات الميتة خلال هذه المرحلة بمقدار ٨٩,٨٩% و ٨٥.٣٩% على التوالى ، وذلك بغض النظر عن الصنف المستخدم. ثانياً ، كان البصل واحداً من أكفأ محصولين من حيث القدرة على زيادة النسبة المئوية للبادرات الباقية على قيد الحياة لجيزة ٨٠ ، فى حين كان الثوم هو الأكفأ من حيث القدرة على زيادة النسبة المئوية للبادرات الباقية على قيد الحياة لباقى الأصناف. ثالثاً ، أحدث الثوم زيادة معنوية فى أطوال بادرات جيزة ٨٠ ، كما أن الثوم والبصل أحدثا زيادة معنوية فى أطوال بادرات جيزة ٨٥. رابعاً ، أحدث الثوم زيادة معنوية فى الوزن الجاف لبادرات جيزة ٨٠ وجيزة ٨٣ بمقدار ٥٧,٦٦% و٩٩,٠٤% على التوالى. درس تأثير عمق اللقاح الفطرى (أعماق ٥ و ١٥ و ٢٥ سم) على قابلية صنف جيزة ٧٥ للإصابة بفطر ماكروفومينا فاسيولينا. أجريت الدراسة تحت ظروف الصوبة فى أصص فخارية قطر ٣٠ سم ، وكان مصدر اللقاح الفطرى هو حبوب ذرة رفيعة معدية بالفطر. أحدث اللقاح السطحى زيادة معنوية فى تكرار عزل الفطر مقارنة باللقاح العميق ، كما أن التأثيرات الضارة للفطر على نمو النبات إنعدمت عند وضع اللقاح على عمق ٢٥ سم قبل الزراعة ، هذا وقد إستمر إنعدام التأثيرات الضارة حتى وقت الإزهار.