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Pathogenicity of *Scleritinia sclerotiorum* to Beans (*Phaseolus vulgaris*, L.) Cultivars

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Sclerotinia rot caused by *Sclerotinia sclerotiorum* is a serious threat to green beans production in Egypt. The pathogenicity of this pathogen to 11 different cultivars was measured by survival plants % in the genotypes. Significant differences were observed between different cultivars (P≤0.005). Results indicate that, Amy and Giza cultivars were more susceptible to infection with *S. sclerotiorum* that produced 16% survival plants in both cultivars after 60 days. While, Duel cultivar was less sensitive to infection with the pathogen that giving 40% living plants at 60 days.

Key words: *Sclerotinia Sclerotiorum*, Pathogenicity, Beans.

Sclerotinia white rot caused by the ascomycete *Sclerotinia sclerotiorum*, is a serious hazard to green beans production with substantial yield losses from thisdisease recorded world-wide [1,2,3]. While, *S. sclerotiorum* is considered to show little host specificity[4], it is important to understand that the diversity of this pathogen to develop effective strategies to detect the identification and dissemination of host resistance. The pathogenicity and diversity studies of this fungus have been examined for different crops in the world [5,6,7,8]. Several past studies haveexplored the genetic diversity of *S. sclerotiorum*[3, 9,10,11,12]. Further, only limited studieshave been conducted so far, to understand the diversity and pathogenicity of *S. sclerotiorum* on beans or other hosts in Egypt [13]. These include the work of Sexton *et al.* [14] who established genotypicdiversity among *S. sclerotiorum* isolates collected from oilseed rape crops from Australia, utilizing microsatellite markers, and Ekins*et al.* [15], who compared aggressiveness of *S. sclerotiorum* isolates collected also from Australia on sunflower.Alterations in the morphology of *S. sclerotiorum* isolates have previously been noticed by Li *et al.* [16]and Garrabrandt *et al.* [17] where isolates producing tan sclerotia were identified. Very fewreports exist to date describing darkly-pigmented isolates of *S. sclerotiorum*, such as those from Canada and the south-western region of the USA [18,19]. Primarily, thedark color of the fungus colonies results from theconstruction of melanin, the main role of which inthis pathogen is to protect the sclerotia from adverse biological and environmental conditions[18,20]. An association of melanin with pathogenicity has also been reported in other pathogens. The objective of our study was evaluate of the pathogenicity of *S. sclerotiorum* from Ismailia governorate, Egypt to

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selected genotypes of bean,under greenhouse conditions.

MATERIALSAND METHODS

Sclerotinia sclerotiorum isolates

S. sclerotiorum was collected from Ismailia governorate, Egypt in 2008 was used inthis study. The initial cultures were then sub-cultured on to water agar and stored at 4°C. All isolates were subsequently sub-cultured to potato dextrose agar (PDA) as this medium allows the best expression of any pigmentation occurring in *S. sclerotiorum* colonies [18,20].

In this experiment, we studied the pathogenicity of *S. sclerotiorum* to 11 bean cultivars. The experiment was conducted under greenhouse condition. Pots (30×25×30) containing sterile soil (sand: loamy sand: compost, 1:2:1) were used, 5 seeds/pot and 5 replicates per treatment. Seedlings were grown until cotyledons were fully expanded. Five agar plug discs (each 5 mm^2diam) were cut from the actively growing margin of 3 day-old colonies of *S. sclerotiorum* on PDA at 20±2°C and transferred to 250 ml flasks containing 100 ml ofsterilized potato dextrose broth. Flasks were incubated at 20±2 °C for 7 days, colonies of *S. sclerotiorum* were harvested and washed twice with sterilized deionized water. The mycelial suspension was then filtered through four layers of cheesecloth and the concentration adjusted with the same liquid medium to 1×10^4cfu/ml using a haemocytometer. A total of 20 ml of mycelial suspension were applied to pots. The number of survival plants after 15, 30, 45 and 60 days were recorded.

Statistical analysis

Data collected from all experiments were statistically analyzed using the Statistic Analysis System package (SAS institute, Cary, NC, USA). Differences between treatments were studied using Fisher's least significant difference (LSD) test and Duncan's Multiple Range Lest[21]. All analysis were performed at P 5 % level.

RESULTS

Pathogenicity of *S. sclerotiorum* to different bean cultivars

After 15 days

Data in Table 1 and 2 reveal that no significant difference in degree of sensitivity for the tested cultivars to*S. sclerotiorum*. The genotype paulista was the lowest cultivar for sensitivity to *S. sclerotiorum* that giving 80% survival plants. This was followed by both saheland amy which produced the same result with 72% survival plants. While, giza-4 cultivar was the most sensitivity to infection with the fungus, which produced 32% survival plants.

After 30 day

There were non-significant differences between all cultivars for sensitivity of the white rot disease caused by *S. sclerotiorum*. The variety duel was the best cultivar for tolerant to infestation by *S. sclerotiorum* that giving 60.00% living plants

Table 1. ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Survival plant after 15 days	Between Groups	27.636	10	2.764	3.663	0.001
	Within groups	33.200	44	0.755		
	Total	60.63610	54			
Survival plant after 30 days	Between Groups	14.436	10	1.444	3.452	0.002
	Within groups	14.800	44	0.418		
	Total	32.836	54			
Survival plant after 45 days	Between Groups	14.109	10	1.411	3.979	0.001
	Within groups	15.600	44	0.355		
	Total	29.709	54			
Survival plant after 60 days	Between Groups	6.182	10	0.618	2.061	0.049
	Within groups	13.200	44	0.300		
	Total	19.382	54			

Table 2. Pathogenicity of S. sclerotiorumto different bean cultivars.

Treatment	15 days			30 days			45 days			60 days		
	No.	Mo. %	Sur.%	No.	Mo. %	Sur.%	No.	Mo. %	Sur.%	No.	Mo. %	sur.%
Sahel	3.6bc	28.0	72.0	2.2bcde	28.0	44.0	2.2bc	56.0	44.0	1.4ab	16.0	28.0
Mael	2.8ab	44.0	56.0	2.0ab	16.0	40.0	1.6ab	68.0	32.0	1.4ab	4.0	28.0
Duel	3.4bc	32.0	68.0	3.0d	8.0	60.0	2.6c	48.0	52.0	2.0b	12.0	40.0
Samantha	2.4ab	52.0	48.0	1.4ab	20.0	28.0	1.2ab	76.0	24.0	1.0a	4.0	20.0
Belina	3.4bc	32.0	68.0	2.4cde	20.0	48.0	1.6ab	68.0	32.0	1.4ab	4.0	28.0
Paulista	3.0bc	40.0	60.0	2.6de	8.0	52.0	1.6ab	68.0	32.0	1.4ab	4.0	28.0
Branco	4.0d	20.0	80.0	2.4cde	32.0	48.0	1.6ab	68.0	32.0	1.2a	8.0	24.0
Amy	3.6bc	28.0	72.0	1.6ab	40.0	32.0	0.8a	84.0	16.0	0.8a	0.0	16.0
Julia	2.6ab	48.0	52.0	2.0ab	12.0	40.0	1.4ab	72.0	28.0	1.0a	8.0	20.0
SB4070	2.0ab	60.0	40.0	1.8ab	4.0	36.0	1.4ab	72.0	28.0	1.0a	8.0	20.0
Giza-4	1.6a	68.0	32.0	1.2a	8.0	24.0	0.8a	84.0	16.0	0.8a	0.0	16.0
LSD	071			0.53			0.49			0.45		

No. = Number of living plants; Mo. % = Mortality percentage; Sur. %= Survival plant%
Values within a column followed by the same letter are not significantly different according to Duncun's multiple range test (P£0.05).

when compared to other cultivars. These were followed by paulista and branco with 52 and 48%survival plants, respectively. Whereas, the most cultivar for sensitivity to the white rot disease was giza-4 and Samantha giving 24 and 28survival plants (Table 1 and 2).

After 45 day

There were non-significant differences between among all cultivars to sensitivity of the white rot disease caused by *S. sclerotiorum* (Table 1 and 2). The lowest cultivar for sensitivity was duel which produced 52% survival plants, when compared to other cultivars, followed by sahelthat giving 44% survival plants. Conversely, the cultivars amy and giza-4 were the most cultivars sensitivity to the fungus that giving 16% survival plants in both cultivars.

After 60 day

There were non-significant differences between all cultivars tested. The duel cultivar was the lowest cultivar for sensitivity to *S. sclerotiorum* which produced 40% survival plants, followed by sahel, mael, belina and paulistathat giving the same result (28% survival plants). While, the cultivars amy and giza-4 were the most sensitivity for the white rot disease caused by *S. sclerotiorum* giving 16% living plants in both cultivars (Table 1 and 2).

This finding is consistent with other reports for the pathogenicity of *S. sclerotiorum*to several crops[23,23]. Usually, the strong defense against the wild-type strain of *S. sclerotiorum* at the early stage of infection is not noticeable, whichmeans that the defense is most likely to be suppressed or postponedby this pathogen. If suppression is a means by which *S. sclerotiorum*is successful as a pathogen, then it is not surprising that *S. sclerotiorum* may secrete pathogenicity factors to aid in the suppression of host resistance. Previous studies on the pathogenicity of plant pathogenic fungigenerally focus on toxins (including proteinaceous effectors), proteinases and plant cell degrading enzymes such as pectinases andcellulase (EC 3.2.1.4, endo-1,4-beta-D-glucanase, beta-1,4-glucanase, beta-1,4-endoglucan hydrolase, celluase A, cellulosin AP, endoglucanase)[24,25]. Oxalic acid is considered a keypathogenicity factor for the killing of plant cells and tissues by *S. sclerotiorum*, and it is also involved in reducing host resistance and interjecting the host physiology rather than as

adirect killer[25,26,27,28]. However, this topic is also one of increasing complexity; several mutants of *S. sclerotiorum* produce considerable amounts of oxalic acid, but do not infect the plant, but virulence is weak[29]; in addition, the mutant cannot produce oxalic acid, but can still infect plant[30]. Recently, Williams *et al*[29] confirmed that reactive oxygen species was virtually absent in DAB stained leaf inoculated with the wild-type strain of *S. sclerotiorum*, while leaves inoculated with an oxalic acid deficient mutant A2 displayed strong DAB staining surrounding the infection point, and they believed that oxalic acid suppresses host defenses by manipulating the host redox environment at8 hpi, an early stage of infection.

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