

## CONTROL OF SESAME DAMPING-OFF DISEASE IN EL-BEHEIRA GOVERNORATE NEWLY RECLAIMED LAND.

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### ABSTRACT

*Sclerotium bataticola* (Taub.) But, *Fusarium oxysporum* (Schlecht), and *Rhizoctonia solani* (Kuhn) were associated with sesame damping-off in the newly reclaimed land in El-Beheira governorate. These fungi were isolated at frequencies of 38.4 % , 28 % and 20.4 % , respectively. *Pythium debaryanum*, *Mucor* sp, *Aspergillus niger* and *Rhizopus* sp, were also, isolated but at lower frequencies i.e. 6.4 % , 3.6 % , 2.0 % and 1.2 % , respectively. *Sclerotium bataticola* was the most pathogenic fungus on the sesame cultivars tested. The cultivar Shandawil was found to be the most tolerant among the cultivars tested. The percentages of both pre- and post-emergence damping-off incited on this cultivar were 16.5 % and 16.4 % compared to 29.5 % and 21.7 % for cv.Giza32. and the percentages of pre- and post-emergence damping-off were 32.3 % and 20.4 % for cv.Toshky. The biofungicide Plant-guard ( *Trichoderma* preparation at 6.0 ml/l ) inhibited effectively the *in vitro* growth of the most prevalent damping-off fungi of sesame. The use of Rizolex-T fungicide at 250 ppm as soil drench integrated with Plant-guard (as seed soaking) decreased the damping-off incidence to 5.41 % and 6.25 % , respectively in the two seasons (2003, 2004) compared to 27.11 % and 29.0 % for the non-treated control. Sesame yield was increased by 122.0 % and 103.0 % compared with non-treated infected sesame plants.

## INTRODUCTION

*Sesamum indicum L.* (Sesame), is important economic crop in various regions of the world. Great attention was paid to this crop in industry and food science technology as important and cheap source of oil and protein for human and animal consumption. Unfortunately in Egypt and several countries sesame is exposed to several soil borne fungi causing damping-off disease which affected both yield and quality of sesame.

Several methods were suggested for the control of damping-off of sesame. This included the use of resistant cultivars, (Chattopadhyay and Sastry, 2002) the chemical control (Abd-El-Hakem and Abou Salama, 1995; Shalaby, 1997 ; and Chattopadhyay and Sastry, 2002), as well as the biological control (Chung and Choi, 1990; Sanker and Jeyarajan, 1996; Wuike *et al.*, 1998). Each of these control measures was an important means in checking sesame damping-off. However, a combination of those measures was suggested for a sustainable and efficient control (Chattopadhyay and Sastry, 2002).

The present study, therefore, was conducted to isolate (i) the pathogens of sesame damping-off.(ii) the relative susceptibility of the widely grown sesame cultivars, and (iii) potential of Plant-guard biofungicide and certain fungicides for controlling sesame damping-off.

## MATERIALS AND METHODS

### **Isolation and identification of sesame damping-off fungi :-**

During 2003 and 2004 growing seasons, samples of sesame plants exhibiting damping-off symptoms were collected from different fields of El-Tahrir region ( a major area of sesame in Egypt). Samples were washed thoroughly with tap water. Small portions of the diseased samples were surface sterilized in 3 % colorox (NaCl) for 3 minutes, rinsed in sterilized distilled water and dried between sterilized filter papers. The portions were plated on PDA medium and incubated at 25 C°. The developed colonies were purified using hyphal tip or single spore techniques, (Tuitte,1969). Identification of

the fungi detected was conducted according to Booth, (1971), Barnett, (1972) and Ramirez, (1982).

### **Pathogenicity tests and varietal reactions:**

Inoculum was prepared by growing each of recovered fungi in 500 ml conical flasks containing 150 g autoclaved sorghum medium, each contains ( sorghum, sand and water in 1 : 20 : 4 ratio) Abd El-Rehim, (1984) and incubated at 25 ° C for 15 days.

Sterilized pots, 25 cm in diameter, were filled with 5 kg autoclaved sandy soil and infested with the inoculum of each fungus tested at the rate of 5 g / kg soil. Each pot was sown one week after fungal infestation with five surface sterilized sesame seeds. The sesame cultivars tested were Giza 32, Toshky and Shandawil. Check treatments were carried out in pots filled with sterilized sandy soil mixed with (non-inoculated) sorghum medium. Five replicates of pots were used for each treatment. Pre- and post-emergence damping-off were recorded 15 and 45 days after sowing, respectively ( Helal *et al.*, 1994).

### **Control of sesame damping-off :-**

#### **1- In vitro tests :-**

The biofungicide Plant-guard as well as four commercially available fungicides namely Kema-Zed, Rizolex-T, Sumisclex and Vitavax-200 (Table1) were tested in three concentrations i.e 3, 4 and 6 ml/l for Plant-guard. Six concentrations i.e. 3, 4, 6, 150, 250 and 300 ppm for the chemical fungicides were used. The different concentrations were added to the molted PDA just before pouring the plates. After solidification, 3 mm diameter disc of each fungus tested was placed in the center of each petri dish. Dishes were then incubated at  $25 \pm 1$  °C. Three replicates were used for each treatment and control too. Linear growth colony diameter was determined (in mm) when the growth reached its maximum in control treatment.

#### **2- Field Experiments :-**

A field experiment was carried out in complete randomized plots at El-Tahrir region in a heavily infested soil during two successive seasons, 2003 and 2004. The most tolerant sesame cultivar (Shandawil) revealed in the pathogenicity tests was used. Each plot was of 10.5 m<sup>2</sup> and comprised 4 rows, 60 cm. apart, and feeded with

sesame seeds in 20 cm distance was used. Each plot included 80 seeds. Sesame seeds were either dipped in a suspension of Plant-guard (soaking) or coated with sumisclex fungicide (seed dressing) before sowing. Soil of both treatments was also treated one month after sowing with Rizolex-T, Vitavax-200 and Kema-Zed at the rates (3, 4, 6, 150, 250, and 300 ppm) derived from the *in vitro* tests. Sowing date was in 1<sup>st</sup> May. All treatments were usually irrigated immediately after sowing, as well as 8 days intervals. Plants were fertilized twice by 100 kg / feddan potassium sulphate (48 %) after 20 and 40 days from sowing. Ammonium sulphate (20.6 % Nitrogen) was added at three intervals, (50 kg / f after sowing and before irrigation, 100 kg / f after 20 days of sowing, and 75 kg / f after 40 days of sowing).

Pre- and post-emergence damping-off were recorded 15, and 45 days after sowing (Helal *et al.*, 1994). Yield of sesame per plant was determined 120 days after sowing. (Anonymous, 2003).

**Table (1) : Active ingredients and chemical composition of the fungicides and bioagents tested .**

Fungicides and bioagents	Active ingredient	Chemical Composition
Plant guard	Spores of <i>Trichoderma harzianum</i>	Solution for seed treatment contained 30 million spores of <i>Trichoderma harzianum</i> / ml .
Kema-Zed	50 % WP	(Carbendazim) Methyl benzimidazol-2-YL carbamate.
Rizolex-T	50 %	(Tolclofos- methyl): Rizolex (20%) O.dimethyl-O-(2-6-dichloro-4-methylphenyl) phosphorothionate+ Thiram ( 30 % ) Bis ( dimethyl thiocarbamoyl ) disulphide.
Sumisclex	50 % WP	(Procymidone) : N-(3,5 dichlorophenyl 1,2 dimethylclopropanel dicarboximide.
Vitavax-200	75 % WP	(Carboxin): 5,6 dihydro- 2- methyl- 1,4 oxathiin -3- carabxenilide+ captan. Uniroyal Inc., Bethany, Connecticut, U.S.A.

**Statistical analysis :**

The data obtained were statistically analyzed according to Gomez and Gomez, (1984) at the Costat Computer Program. Means were compared using L.S.D test at 0.05 probability level.

## EXPERIMENTAL RESULTS

### 1- Fungi causing sesame damping-off disease :-

Data in Table (2) show that several fungal species were isolated from sesame plants showed damping-off symptoms. *Sclerotium bataticola* (Taub), *Fusarium oxysporum* (Schlecht), and *Rhizoctonia solani* (Kühn), were prevalent and recovered at frequencies of 38.4 %, 28.0 % and 20.4 %, respectively. *Pythium debaryanum* , *Mucor* sp., *Aspergillus niger* and *Rhizopus* sp. were also recovered but at lower frequencies of 6.4 %, 3.6 %, 2.0 % and 1.2 %, respectively.

**Table(2): Frequency of fungi isolated from sesame plants showing damping-off symptoms collected from El-Beheira governorate during the 2003- 2004 growing seasons.**

Isolated Fungi	Frequency %
<i>Sclerotium bataticola</i>	38.4
<i>Fusarium oxysporum</i>	28.0
<i>Rhizoctonia solani</i>	20.4
<i>Pythium debaryanum</i>	6.4
<i>Mucor</i> sp	3.6
<i>Aspergillus niger</i>	2.0
<i>Rhizopus</i> sp	1.2

### 2- Pathogenicity tests and varietal reactions :-

Pathogenicity tests conducted on the three sesame cultivars tested i.e. Giza 32, Toshky and Shandawil revealed that the fungi *S. bataticola*, *F. oxysporum*, and *R. solani* were pathogenic on the sesame cultivars tested. *S. bataticola* was the most pathogenic as incited pre-emergence ranged between 20.7 % and 48.1% and so post-emergence ranged between 16.4 % and 28.3 % on the tested sesame cultivars. *R. solani*, however, incited 20.2 % - 36.3 % pre-emergence damping-off and 12.4 % - 24.2 % post-emergence damping-off. However, *F. oxysporum* incited 8.5 % - 24.3 and 12.7 % - 24.6 % for pre-, and post-emergence damping-off, respectively.

Data in Table (3) indicated that sesame cultivar Shandawil was the most tolerant cultivar tested. Mean of pre- and post-emergence damping-off incited on cultivar Shandawil were as low as 16.5 % and 16.4 % respectively, compared to 29.5 % and 21.7 % on cv.Giza 32 and 32.3 % and 20.4 % on cv.Toshky.

**Table (3) : Pre- and post-emergence damping-off percentage incited on Giza 32, Shandawil and Toshky sesame cultivars sown in potted soil artificially infested with the fungi tested.**

Cultivars \ Fungi	Giza 32		Shandawil		Toshky		Mean	
	Pre-emer.	Post-emer.	Pre-emer.	Post-emer.	Pre-emer.	Post-emer.	Pre-emer.	Post-emer.
<i>Sclerotium bataticola</i>	40.2	24.3	20.7	16.4	48.1	28.3	36.3	23
<i>Fusarium oxysporum</i>	24.3	24.6	8.5	12.7	12.4	20.6	15.1	19.3
<i>Rhizoctonia solani</i>	24.1	16.4	20.2	20.2	36.3	12.4	26.9	16.3
Control (Non-inoculated)	4.0	4.0	0.0	0.0	0.0	0.4	1.3	2.7
Mean	29.5	21.7	16.5	16.4	32.3	20.4	26.1	19.5
% Total disease incidence	51.2		32.9		52.7		45.6	

Data are mean of 5 replicates.  
emergence

pre- emergence

post-

L.S.D.<sub>0.05</sub> among the fungi tested =

3.66

N.S.

L.S.D.<sub>0.05</sub> among the cultivars tested =

5.177

5.361

L.S.D.<sub>0.05</sub> among the interaction F × cvs. = N.S.

N.S.

### **The control experiments :-**

#### **1- In vitro tests .**

Results in Table (4) show the effect of the biofungicide Plant guard and different concentrations of four fungicides on the *in vitro* growth of *Sclerotium bataticola*, *Fusarium oxysporum*, and *Rhizoctonia solani*. Plant guard at 6 ml/l significantly inhibited linear growth (estimated as the colony diameter) of all tested fungi as completely inhibited growth. The Kema-Zed, was able also to inhibit the fungal growth by only 6 ppm. On the other hand, Sumisclax,

Rizolex-T and Vitavax-200 completely inhibited, *in vitro* growth of all fungi tested at 250 ppm and 300 ppm, respectively. Meantime, Plant guard and the fungicides tested differed in their inhibition effect on the different fungal species. Plant guard at 3 ml/l was able to inhibit *F.oxysporum* growth. However, *R. solani* growth inhibited at 6 ml/l. On the contrary, *R. solani* growth was inhibited at 4 ppm Rizolex-T while, *F. oxysporum* inhibited at 250 ppm.

Sumisclex and Vitavax-200 were able to inhibit *R. solani* growth at 6 ppm, while, 250 ppm were needed to each of the two fungicides to affect *F. oxysporum*. However, *S. bataticola* growth was only inhibited at 250 ppm and 300 ppm of the two fungicides, respectively (Table 4).

## **2- Field Experiments :-**

Treatment of sesame seeds with Plant-guard coupled with the effect of Rizolex-T as soil drench which considered as the most effective treatment reduced sesame of damping-off diseases to 5.41 % and 6.25 %, compared to 27.11 % and 29.0 % for the non-treated control during the two growing seasons 2003 and 2004, respectively. This treatment followed by Plant-guard and Vitavax-200 soil drench as incited disease was 7.62 % and 8.31 %, respectively. Treatment with Kema-Zed and Plant guard exhibited the least effects as the incited disease was 8.75 % and 11.71 % for the two successive seasons, respectively. On the other hand, seed dressing with Sumisclex followed by soil drench with either Rizolex-T, Vitavax-200 or Kema-Zed decreased the sesame damping-off. It ranged between 9.21 % - 13.14 % and 10.52 % - 12.61 % for the two successive seasons, respectively (Table 5).

All treatments exhibited significant increase in yield when compared with the non treated control. The highest yield ( 6.04 – 5.73 g / plant) was obtained when seeds treated with Plant-guard and soil drench with Rizolex-T followed by seed treatment with Plant- guard coupled with Vitavax-200 soil drench as the yield of sesame was 4.71 , 3.86 g / plant in the two successive growing seasons 2003-2004, respectively,( Table 5)



**Table (5) : Effect of seed and soil treatments with Plant-guard and certain fungicides on damping-off incidence and yield of cv. Shandawil sesame plants in the field under natural infection.**

Treatments *	% disease incidence**		Yield ( g/ plant )	
	2003	2004	2003	2004
Plant guard + Rizolex T	5.41	6.25	6.04	5.73
Plant guard + Vitavax 200	7.62	8.31	4.71	3.86
Plant guard + Kema-Zed	8.75	11.71	4.69	3.63
Sumisclex + Rizolex T	9.21	10.52	4.36	3.99
Sumisclex + Vitavax 200	9.44	11.33	3.54	3.57
Sumisclex + Kema-Zed	13.14	12.61	4.14	4.05
Control (non-treated)	27.11	29.0	2.72	2.81
L.S.D at 0.05	1.63	1.97	1.28	1.11

## DISCUSSION

Several fungi were found to be responsible for the damping-off disease of sesame plants in the newly reclaimed soils in El-Beheira governorate, Egypt. *Sclerotium bataticola*, *Fusarium oxysporum* and *Rhizoctonia solani*, were prevalent and isolated at frequencies of 38.4 %, 28.0 %, and 20.4 %, respectively. *Pythium deparyanum*, *Mucor sp*, *Aspergillus niger* and *Rhizopus sp*, were also associated with damping-off of sesame seedlings, but were isolated at lower frequencies of 6.4, 3.6, 2.0, and 1.2 %, respectively. These results are in harmony with research works carried out in Egypt and other parts of the world (Al-Ahmad and Saidawi 1988; Shalaby and El-Korashy 1996; Wuike *et al.*, 1998; Karunanithi *et al.*, 1999 and Abdou *et al.*, 2001).

*Sclerotium bataticola*, *F. oxysporum*, and *R. solani* were the most pathogenic to sesame plants and caused damping-off disease. These results are somewhat agree with the findings of Eisa *et al.*, (1994) who concluded that *R. solani* caused damping-off and root-rot diseases of *Volca meriana* seedlings and with Nalim *et al.*, (1995) in case of groundnut.

Among the sesame cultivars tested Shandawil was the most tolerant one. The incited damping-off on the cv. Shandawil was as low

as 16.5 % and 16.4 % for the pre- and post-emergence damping-off, respectively. This compared to 29.5 % and 21.7 % on cv. Giza 32 and 32.3 % and 20.4 % on cv. Toshky. These findings are in agreement with Abd-El-Moneem *et al.*, (1997) and Gabr *et al.*, (1998).

The biofungicide Plant-guard at 6 ml/l was effective and completely inhibited growth of all sesame damping-off fungi tested. The control of damping-off was positively related to the concentration of biocontrol agent. This could be attributed to the increase of competitive reaction between biocontrol agent ( *T. harzianum* ) and soil-borne pathogens and also to the increase of antibiotic produced by biocontrol agent ( Abdel-Mageed, 1997 ).

Treatment of sesame seeds with biocontrol agent Plant guard before sowing was effective in controlling damping-off attributed to *S. bataticola*, *F. oxysporum*, and *R. solani* because the earlier application allowed the antagonist to spread out in the infested soil, establishes itself and increases its population to at least constant density sufficient to antagonize plant pathogens. This is in agreement with the findings of Essmat *et al.*, 1995 in case of Senna seeds.

Moreover, the effect of Plant-guard, as conidial preparation of *Trichoderma spp*, in controlling the soil borne pathogens could be, due to the ability of conidia of *Trichoderma* to germinate, colonize the area, and constitute a defence barrier around the germinating seeds. These results are in agreement with those reported by Chung and Choi, (1990); Abd El-Hakem and Abou Salama, (1995); Sanker and Jeyarajan, (1996); Shalaby, (1997); Gabr *et al.*, (1998); Chattopadhyay and Sastry, (2002) and El-Ghannam Abeer (2003).

However, the chemical fungicides tested, i.e. Kema-Zed, Rizolex-T, Sumisclex, and Vitavax-200 were able to inhibit the fungi tested at 6 ppm, 250 ppm, 250 ppm and 300 ppm, respectively. This result may be attributed to the fact that some fungicides act within the fungal cell by inhibiting and interfering metabolism (Abada, 1995). In this respect, our results are in harmony with many investigators who registered similar trend, where *R. solani* growth was completely inhibited within a range of 200-400 ppm of Rizolex-T (Katania and Verma, 1990 and Katania *et al.*, 1991) .

In field study, use of Rizolex-T at 250 ppm supported the biofungicide Plant-guard effect and decreased incidence of damping-off to 5.41 % and 6.25 % in the two seasons (2003 and 2004), respectively. This may be due to the fact that Rizolex-T is systemic fungicide which interfer metabolism of pathogenic fungi, and the rate of pathogen growth inhibition positively correlated to the increasing of Rizolex-T concentrations. The effect was reflected in a significantly higher yield of sesame, i.e. 6.04 and 5.73 g / plant in the two successive growing seasons 2003 and 2004 compared to 2.72 and 2.81 g / plant for the non-treated control.

Consequently, use of the most tolerant cv. Shandawil of sesame, as seed soaking in the biofungicide Plant-guard at 6 ml/l before sowing, coupled with Rizolex-T soil drench, 25 days after sowing has provided a good management for damping-off diseases of sesame .

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## الملخص العربي

### مقاومة مرض سقوط البادرات لنباتات السمسم في الأرض المستصلحة الجديدة في محافظة البحيرة .

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تم عزل فطريات سكليروشيم باتاتيكولا ، فيوزاريوم أوكسيسبورم ورايزوكتونيا سولاني المصاحبة لمرض سقوط البادرات في نباتات السمسم من عينات تم جمعها من الأرض المستصلحة في محافظة البحيرة بنسبة 38.4 % ، 28 % ، 20.4 % على التوالي. بينما تم عزل فطريات بيتيم ديباريانم ، ميوكر ، اسيرجلس نيجر ، ورايزوبس بنسبة أقل حيث تواجدت بنسبة 6.4 % ، 3.6 % ، 2 % ، 1.2 % على التوالي. وقد أسفرت تجارب القدرة المرضية عن أن الفطر سكليروشيم باتاتيكولا كان الأكثر قدرة إمراضية على أصناف السمسم المختبرة وهي جيزة 32، شندويل ، توشكي حيث أحدث إصابة لسقوط البادرات قبل ظهورها فوق سطح التربة تراوحت بين (20.7% إلى 48.1%)، وما بعد الإنبات (16.4% - 28.3%) يليه الفطر رايزوكتونيا (20.2 % ، 36.3%)، وبعد الإنبات (12.4% ، 20.2%) ، ثم الفطر فيوزاريوم أوكسيسبورم قبل الإنبات (8.5 % ، 24.3%) ، وبعد الإنبات (12.7 % ، 24.6%).

كما أظهرت الدراسة أن الصنف " شندويل" كان الأكثر تحملا للإصابة بين الأصناف المختبرة حيث كانت نسبة الإصابة بسقوط البادرات قبل الظهور وبعد الظهور (16.5 % ، 16.4 % ) على التوالي مقارنة بصنف جيزة 32 (29.5% - 21.7%) ، صنف توشكي (32.3 % ، 20.4 % ) .

كما ثبت المركب الحيوي بلانت جارد بتركيز 6 مل / لتر نمو الفطريات سابقة الذكر ، كما أن استعمال رايزولكس- ت بتركيز 250 جزء في المليون كمعاملة غمر للتربة مع استعمال البلانت جارد كمعاملة نقع بذور قد أدى إلى خفض نسبة الإصابة بسقوط البادرات إلى 5.41 % ، 6.25 % ، في موسمي 2003 ، 2004 على التوالي وقد أدى ذلك إلى حدوث زيادة في المحصول 122 % ، 103 % مقارنة بنباتات السمسم المعدة غير المعاملة .