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In Vitro and *in Vivo* Management of *Sclerotium rolfsii* the Cause of Sugar Beet Root Rot Disease

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Abstract: Management of sugar beet damping off and root rot diseases caused by *Sclerotium rolfsii* is urgently needed. Therefore, antifungal activities of 13 materials including 2 bio agents, 2 seaweeds, 3 chemical inducers and 6 fungicides were evaluated. Required inoculum potential of either fungal mass or sclerotia to reach more than 50% of disease incidence was firstly investigated. Fungal mass inoculation (40-500g/10kg of soil) provided 70-100% damping off and 100% root rot. Meanwhile, 6.7-36.7% of damping off and no or negligible root rot were obtained using 300-500 sclerotia /10kg of soil. On the other hand, *S. rolfsii* mycelial growth was completely suppressed *in vitro* by all tested materials. However, various antifungal activities of these materials were shown *in vivo* after seed soaking in the 1st (2020/2021) trail or seed soaking followed by soil drenching in the 2nd (2021/2022) trail. Tipo top (Tebuoconazole 25.9%: 1cm/L) fungicide was the most effective material in the 1st trail since the seedling survival was up to 80%, followed by potassium silicate (1cm/L) and Score (Difenoconazole 25%: 1cm/L) fungicide. Seed soaking followed by soil drenching with Tipo top in the 2nd trail were protected sugar beet from sowing to harvest and enhanced the root weight. Additionally, this study illustrated that both of sugar beet root weight and sucrose content were decreased as root rot severity increased. In conclusion, chemical fungicides are unfortunately still the fast and potent way for *S. rolfsii* management, especially with the limitation of resistant sugar beet cultivars.

Keywords: Root Rot, Beta Vulgaris, Sclerotia, Disease Control

1. Introduction

One of the major limiting factors of sugar beet (*Beta vulgaris* L.) production worldwide is root attacking fungal diseases [1-4]. Of these, sugar beet root rot caused by *S. rolfsii*, is the most serious and devastating fungal disease in the tropical and subtropical regions [5]. Sclerotia of this fungus that formed in the soil at the end of the disease life cycle and/or on dead plant materials are responsible for fungus survival and disease dissemination [6]. *S. rolfsii* also reported to be capable of attacking hundreds of cultivated and wild plant species belonging to wide range of plant families [7-10]. However, positive correlation has previously been documented between disease incidence in a sugar beet field and the density of the fungal structures in the soil [6]. Furthermore, due to the existence of wide range of hosts along with sclerotia, which may survive for many years in the soil, beet plants become vulnerable to continuous fungal attack in their cultivation areas [11, 12].

Nowadays, about 65% of the Egyptian sugar production is from sugar beet that cultivated in more than 0.5 million Feddan [13]. Unfortunately, continuous cropping and/or short rotation of this crop in the same cultivation areas instead of good rotations will stimulate infestation by soil borne pathogens affecting the final yield [14, 15]. Additionally, severe infection by *S. rolfsii* could be occurred in the favorable conditions resulting in yield reduction up to 50% of the root production [16].

Many control strategies have been reported against root rot on sugar beet caused by *S. rolfsii* including cultural practices [17], resistant varieties [9], crop rotation [14, 15], biological control [8, 16], and fungicidal control [18-20]. Despite these efforts, *S. rolfsii* is still a difficult pathogen to control with no

clear recommend strategy for its management. Therefore, searching for any approach controlling or at least minimizing the effect of this pathogen is urgently required. So, this study aimed to evaluate some materials to find out a satisfying management strategy of sugar beet Damping-off and root rot caused by *S. rolfsii*.

2. Materials and Methods

2.1. Sclerotium rolfsii Isolate

From sugar beet fields in Daqahliya governorate, Egypt, plants showed wilting symptoms along with white cottony mycelia and white or golden sclerotia on their roots were used for fungal isolation on PDA medium. Isolation and identification were as reported by Pethybridge *et al* [21]. The obtained isolates were tested for their aggressiveness in previous work and the aggressive one, S1, was selected and used in the current study [7].

2.2. Antifungal Bacteria

Pseudomonas fluorescens, strain FP805Pu KT88 1299.1 and *P. putida*, strain H9KX369582.1 were used in this study (Table 2) based on earlier performance for controlling Fusarium root rot of sugar beet [22]. These two bio-agents were grown on the broth of Kings B medium for two days at 27°C after that serial dilution methods were used for obtaining the concentration of 10^6 and 10^8cfu/ml.

2.3. Seaweeds

Two seaweeds (Table 2); *Ulva fasciate* Delile & *Enteromorpha flexuosa* Wulfen were collected from Sewiz canal, Port Said Governorate, washed using tap water, ear dried and grounded by kitchen blender and stored in refrigerator at 4°C until use [23].

2.4. Fungicides and Chemical Compounds

Number of 6 fungicides with two modes of action (DMI and QoI) and 3 silicate compounds were evaluated against *S. rolfsii* in this study. The concentrations and application rates were showed in Table 2.

2.5. S. rolfsii Inoculum

To evaluate the efficacy of the tested materials against *S. rolfsii*, determination of a suitable form of fungal reproduction unit and density as an inoculum producing at least 50% disease incidence is needed. In this study, each of fungal mass and/or sclerotia was used as a source of inoculum in different levels. PDA plates (9cm), were inoculated by disks of 5d-old *S. rolfsii* culture and incubated at 27°C either for 7 or 21 days for fungal mass and sclerotia production respectively [24].

2.5.1. Fungal Mass Inoculum

A quantity of 100g of sorghum seeds, soaked in water overnight, were mixed with 50g of clean sand (washed 3 times and ear dried previously), all deposited in 500 ml glass bottle, mixed and autoclaved for 1 hour. Afterward, sorghum seeds were inoculated with one piece (1x2cm) of 5-day old *S. rolfsii* culture grown on PDA medium and incubated at room temperature (25-27°C) for one week with daily shaking to homogenate the fungus growth [7]. According to soil infestation technique, colonized sorghum seeds were used as fungal mass inoculum in different levels of fungal density (10-500 g/10 kg loam soil) with three replicates of each level (Table 1). Fungus free pots served as control. Subsequently, all pots (35cm in diameter) were irrigated 7-days before planting. Under greenhouse conditions in 2019/2020, ten seeds of *cv.* Kawmera (fungicide-untreated) were planted separately in the infested soil, covered with a slight layer of soil and irrigated. Disease incidence was recorded until 120 days [7].

Table 1. *Effect of inoculum type (fungal mass and/or sclerotia) and rate on sugar beet damping off and root rot caused by S. rolfsii.*

Inoculation Rates	% Damping-off after 40 days[1]			% Root rot after 120 days[2]		
	Sclerotia No. /10kg	Fungal mass g/10kg	Mean	Sclerotia No. /10kg	Fungal mass g/10kg	Mean
0	00.00	00.00	00.00	00.00	00.00	00.00
10	3.33	23.33	13.33	00.00	87.04	43.52
20	3.33	30.00	16.67	00.00	95.83	47.92
30	00.00	20.00	10.00	6.67	96.30	51.49
40	00.00	70.00	35.00	3.33	66.67	35.00
50	00.00	60.00	30.00	3.33	100.00	51.67
60	00.00	70.00	35.00	00.00	100.00	50.00
70	13.33	70.00	41.67	00.00	66.67	33.34
80	6.67	80.00	43.33	3.70	100.00	51.85
90	6.67	80.00	43.33	00.00	100.00	50.00
100	3.33	66.67	35.00	00.00	66.67	33.34
200	00.00	83.33	41.67	00.00	88.89	44.45
300	13.33	100.00	56.67	00.00	00.00	00.00
400	6.67	100.00	53.33	7.04	00.00	3.52
500	36.67	100.00	68.33	00.00	00.00	00.00
Mean	6.22	63.56		1.61	64.54	

LSD (0.05) for the (the interaction) in damping-off = 22.138***, and in root rot = 31.488***

1) Data of damping off were calculated against 10 cultivated seeds.

2) Data of root rot were calculated against the remained two plants after thinning.

2.5.2. Fungal Sclerotia Inoculum

Formed sclerotia were dislodged from PDA fungal culture with the aid of painting brush [16, 24], counted and subsequently used with rates of 10-500 sclerotia/10 kg of soil. Sclerotia were mixed with the soil to provide inoculum uniformity and the pots were immediately irrigated. Cultivation, irrigation and disease assessment were as described above.

2.6. In Vitro Antifungal Assay

Listed materials (Table 2) except nano-sodium silicate and seaweeds were evaluated *in vitro* in 2019/2020. PDA were amended with tested materials under two doses just before pouring in Petri plates, subsequently the plates centrally inoculated with 5-mm agar plugs from the growing 5- day-old culture. The inoculated plates were then incubated at 27°C and the growth was measured after 5 days. Treated plates with sterile distilled water were served as control and four replicates were used for each treatment.

2.7. In Vivo Antifungal Assay

In this assay, *S. rolfsii* contaminated soil was done using 100g of fungal mass / 10Kg potted soil with three replicates in two trails. Ten seeds of *cv* Kawmera were individually cultivated / pot. First trail (2020/2021) was planted in November 11, after 2 h of seed soaking in the tested materials (Table 3) followed by air drying. Application rates were similar to that used in the *in vitro* experiment. Seeds soaked in distilled water prior to sowing in the *S. rolfsii* infested soil were served as control. Recommended cultural practices were carefully followed during the whole season. Pre and post emergence damping-off as well as root rot severity were recorded [21, 25].

Table 2. *Lest of materials used against S. rolfsii the cause of root rot disease of sugar beet roots.*

	Application rates (R)		**Fungicide (F)**		**Season**	
Materials	R1	R2	Active ingredient and concentration	Form	1st	2nd
1. Leader fungicide	1.0 Cm/L	2.0 Cm/L	Prochloraze 12.5%	EC	√	-
2. Score fungicide	0.5 Cm/L	1.0 Cm/L	Difenconazole 25%	EC	√	√
3. Secons fungicide	0.5 Cm/L	1.0 Cm/L	Difenconazole 15% + Propiconazole 15%	EC	√	-
4. Strong x fungicide	0.25 Cm/L	0.5 Cm/L	Pyraclostrobin 18.7% + Propiconazole 11.7%	EC	√	-
5. Tipo top fungicide	0.5 Cm/L	1.0 Cm/L	Tebuconazole 25.9%	WG	√	√
6. Opera fungicide	1.25 Cm/L	2.5 Cm/L	Pyraclostrobin 18.3% + Epoxiconazole 18.3%	SE	√	-
7. Nano Sodium silicate	0.2 g/L	-			-	√
8. Potassium silicate	1 Cm/L	2 Cm/L			√	√
9. Sodium Silicate	1 g/L	2 g/L			√	√
10. *P. fluorescens*	10^6 CFU	10^8 CFU			√	-
11. *P. putida*	10^6 CFU	10^8 CFU			√	√
12. *U. fasciate*	33 g/L	132 g/L			√	-
13. *E. flexuosa*	33 g/L	132 g/L			√	-

EC= Emulsifiable concentrate, WG= Water dispersible granule, SE= Suspoemulsion, 1st = first season, 2nd = second season

The second trail (2021/2022) was conducted in December 11, using the promising material (s) for *S. rolfsii* suppression with the most effective rate/dose according to the previous trail. Tested materials were used as seed soaking only or seed soaking followed by soil drenching. Three treatments of soil drenching according to the number of application times of tested materials were included. Drenching was done after 35; 35&90; 35, 90&135 days of planting respectively. Tipo top (TT), Sodium silicate (SS), Potassium silicate (PS), *P. fluorescens* (PF), Nano-sodium silicate (NSS) and Score (SC) were used. Additionally, TT fungicide was used for seed soaking followed by drenching with SS or PS. Three replicates per treatment were used and seeds soaked in distilled water, sowed in pots with fungal infested soil and drenched with water served as control. All cultural practices were done as recommended. Pre- and post- emergence damping-off (30 and 45 days of planting) as well as root rot severity and root weight (at the harvest time) were recorded.

2.8. Impact of S. rolfsii Root Rot on Sucrose%

At harvest time a representative roots of various degree of rot severity were sampled, cleaned, weighted and prepared for sucrose estimation according to the method described by Association of official analytical chemists [26, 27] with the aid of Polarimeter (Propol, Digital Automatic Polarimeter, OR-Kernchen).

2.9. Statistical Analysis

All collected data were subjected to the analysis of variance (ANOVA) using WASP Package (Web Based Agricultural Statistics Software Package). The least significant difference (LSD) was used to identify differences and compare means. Significant and highly significant levels were shown as 2 (**) and 3 (***) stars respectively in the tables foot notes.

3. Results

3.1. Effective Inoculum Type and Rate

Presented data (Table 1) show the impact of different inoculum types and rates on sugar beet plants. ANOVA proved the significance of the interaction (inoculum rate x inoculum type), indicating that the effect of inoculum rate is

depending on the type of inoculum. For example, 400 and 500 Sclerotia / 10kg soil are significantly different, while 400 and 500g of fungal mass /10kg are significantly indifferent in causing damping-off. Meanwhile, 40 and 50g of fungal mass / 10kg are significantly different, while 40 and 50 Sclerotia / 10kg are significantly indifferent in causing root rot disease on sugar beet. However, all fungal mass treatments were effective, capable of attacking sugar beet causing 20-100% of both damping-off and root rot severity. In contrast, a highest degree of damping-off (36.67%) was obtained with 500 sclerotia / 10kg. Furthermore, soil that infested with less than 500 sclerotia / 10kg resulted in inconsiderable damping-off and root rot severity (Table 1).

3.2. In Vitro Antifungal Assay

Efficacy screening of used materials showed that all were effective and resulted in complete inhibition of *S. rolfsii* mycelial growth (Figure 1).

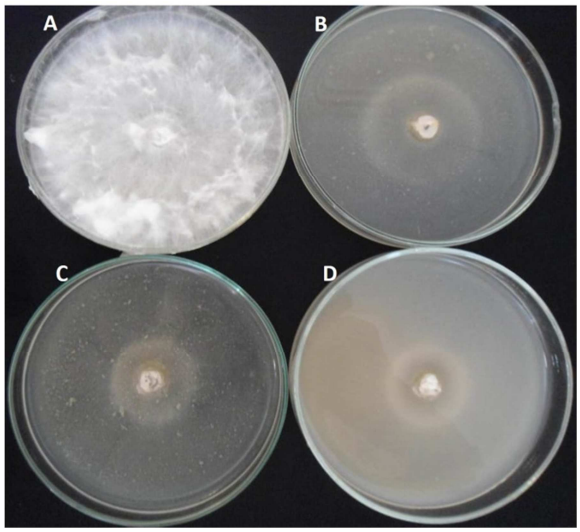

Figure 1. *Growth of S. rolfsii on PDA untreated control (A), compared with that treated with, P. fluorescens (B), Sodium silicate (C), and TT Fungicide (D).*

3.3. In Vivo Antifungal Assay

3.3.1. Season 2020/2021

Table 3 shows the efficacy of 2 different rates of the tested materials against *S. rolfsii*. Significance of the interaction (Materials x rate of application) according to ANOVA indicates that efficacy of any material is affected by its rate of application. For example, Tipo top and strong X fungicides are significantly different at rate1, while they are significantly indifferent at rate2 in causing pre-emergency damping-off. Meanwhile, no significant differences were found regarding the efficacy of the same two fungicides on the post-emergency damping-off or root rot severity at both rate 1 and 2. Tipo top was generally the most promising fungicide against damping-off providing more than 80 and 90% of survivability in pre and post-emergency damping-off followed by Score. Unfortunately, all tested materials have no extended effect till the period of root rot assay (Table 3).

3.3.2. Season 2021/2022

Data in table 4 show the role of tested materials in preventing or minimizing damping-off disease of sugar beet caused by *S. rolfsii*. ANOVA showed that treatments are significant source of variation in pre-emergency damping-off caused by *S. rolfsii*. Obtained results also revealed that TT was the most promising treatment in pre-emergency damping-off, since disease incidence was ranged from 0 to 10% compared with 20% in the control (Table 4).

On the other hand, the interaction (Materials x application method) was a significant source of variation in post-emergency damping-off indicating that material efficacy depending on the method of application. Significant difference was found between the efficacy of SS and TT+SS treatments on the post-emergency damping-off when applied as seed soaking instead of soaking + drenching and vice versa. In general, most of the tested treatments were effective on the pre and post-emergency damping-off but TT and TT+PS were generally the most effective (Table 4).

Table 3. *In vivo efficacy of tested materials applied as seed soaking in 2 rates against S. rolfsii in 2020/2021 season.*

Materials (M)	Pre emergency damping-off %			Post emergency damping-off %			Root rot severity%		
	Rate1	Rate2	Mean	Rate1	Rate2	Mean	Rate1	Rate2	Mean
Leader	75.00	62.50	68.75	00.00	11.11	5.56	83.50	83.33	83.42
Tipo top	16.67	16.67	16.67	00.00	4.76	2.38	95.00	98.33	96.67
Strong x	54.17	29.17	41.67	16.67	5.56	11.11	90.83	98.33	94.58
Sodium Silicate	83.33	79.17	81.25	66.67	11.11	38.89	100.00	74.17	87.08
Potassium Silicate	100.00	100.00	100.00	Nil	Nil	Nil	Nil	Nil	Nil
P. fluorescens	91.67	83.33	87.50	00.00	33.33	16.67	Nil	100.00	100.00
P. putida	91.67	95.83	93.75	66.67	00.00	33.33	Nil	98.00	98.00
Score	29.17	33.33	31.25	00.00	4.76	2.38	93.33	92.50	92.92
U. fasciate	79.17	91.67	85.42	41.67	00.00	20.84	92.50	97.00	94.95
E. flexuosa	83.33	95.83	89.58	100.00	00.00	50.00	Nil	95.00	95.00
Sicons	70.83	70.83	70.83	33.33	50.00	41.67	98.00	98.00	98.00
Upera	37.50	41.67	39.59	26.19	28.33	27.26	95.00	97.33	96.17
Fung+W	91.67	91.67	91.67	66.67	66.67	66.67	100.00	100.00	100.00

LSD (0.05) for (the interaction) in pre-emergency damping-off = 22.98**, post-emergency damping-off = 40.16*** and root rot severity = 8.27***

Table 5 summarizes the impact of tested materials on the occurrence and severity of root rot disease of sugar beet

caused by *S. rolfsii*. ANOVA exhibited also the significance of the interaction (Materials x application method) as a source of variation in both of root rot severity and sugar beet root weight. Significance of the interaction indicates that material efficacy is depending on the application method. For example, insignificant difference was found between the efficacy of TT and TT+PS treatments on root rot severity when applied as seed soaking. In contrast they were significantly differed from each other in cases of soaking + drenching 1, 2 &3 times. TT was the most effective treatment providing full protection against the fungal attack when applied as a drenching for one time or more after the seed soaking (Table 5). Moreover, data of the impact of TT and TT+PS on the final root weight are presented in Table 6. Significant differences were shown between these treatments when applied as seed soaking followed by 1 or 2 times of soil drenching. Meanwhile, they were insignificantly differed from each other when applied as seed soaking only or seed soaking followed by 3 times of soil drenching (Table 6).

Table 4. *Efficacy of selected materials, as seed soaking or Seed soaking followed by soil drenching, on pre- and post-emergency damping-off caused by S. rolfsii (In vivo 2021/2022).*

Materials	Pre-emergency	Post-emergency		
	Soaking	Soaking	Soaking +Drenching1	Mean
TT	10.00	00.00	10.37	5.19
SS	40.00	40.74	32.21	36.48
PS	6.67	55.56	16.67	36.11
Pf	13.33	30.00	45.28	37.64
TT+SS	10.00	00.00	10.00	5.00
TT+PS	0.00	00.00	7.04	3.52
NSS	16.67	34.81	55.65	45.23
SC	13.33	23.21	10.74	16.98
Control	20.00	74.74	74.74	
Mean	14.44	28.78	29.19	

LSD (0.05) for pre-emergency = 20.08, and for (the interaction) post-emergency = 24.21**
Drenching1= soil drenching after 35 days of planting

Table 5. *Efficacy of selected materials on S. rolfsii root rot severity of sugar beet (In vivo 2021/2022 season).*

Materials	Soaking	Soaking +Drenching 1	Soaking +Drenching 2	Soaking +Drenching 3	Mean
TT	70.83	00.00	00.00	00.00	17.71
SS	79.17	83.33	96.67	79.17	84.59
PS	88.67	90.67	97.33	99.67	94.09
Pf	99.83	84.67	94.67	100.00	94.79
TT+SS	70.83	99.33	93.83	86.33	87.58
TT+PS	70.83	93.33	95.83	81.33	85.33
NSS	91.33	92.50	93.33	98.33	93.88
SC	100.00	90.00	53.83	62.50	76.58
Control	99.67	99.67	99.67	99.67	99.67
Mean	85.68	81.50	80.57	78.56	

LSD (0.05) for the interaction =18.17***
Drenching1, 2 & 3= soil drenching after 35, (35, 90) & (35, 90, 135) days of planting respectively.

Table 6. *Efficacy of selected materials against S. rolfsii on sugar beet root weight (gm), In vivo 2021/2022 season.*

Materials	Soaking	Soaking +Drenching 1	Soaking +Drenching 2	Soaking +Drenching 3	Mean
TT	19.88	144.47	119.58	99.74	95.92
SS	68.20	87.88	72.18	71.42	74.92
PS	96.47	65.89	86.77	89.58	84.68
Pf	64.66	144.61	122.36	44.76	94.09
TT+SS	19.88	37.79	91.76	62.81	53.06
TT+PS	19.88	57.23	35.43	60.26	43.20
NSS	69.95	79.75	90.96	82.62	80.82
SC	59.59	116.68	169.65	75.61	105.38
Control	24.35	24.35	24.35	24.35	
Mean	49.21	84.29	90.34	67.91	

LSD (0.05) for the interaction =69.18**
Drenching1, 2 & 3= soil drenching after 35, (35, 90) & (35, 90, 135) days of planting respectively.

Figure 2 shows the clear differences between healthy and *S. rolfsii* infected sugar beet plants as well as the deterioration of the root tissues. Furthermore the relationship between sucrose content and sugar beet root rot severities is shown in Figure 3. In the current study, sucrose content was generally decreased as root rot severity increased. Sucrose content was 15.40% at 10% of root rot severity and decreased to 14.0% when rot severity of roots progressed up to 50%. More than

50% and up to 100% root rot severity resulted in extremely decrease in sucrose content from 14% to 1.1% (Figure 3).

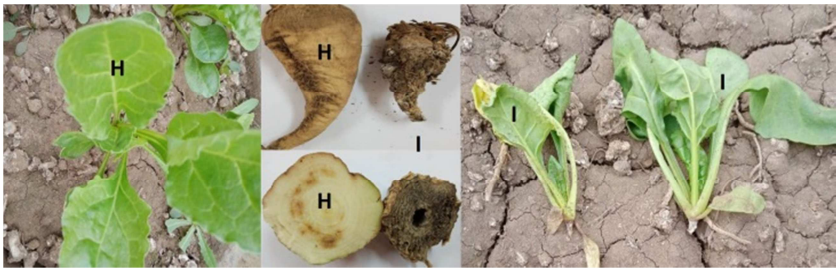

Figure 2. *Healthy (H) and S. rolfsii infected seedlings and rotted root (I) of sugar beet.*

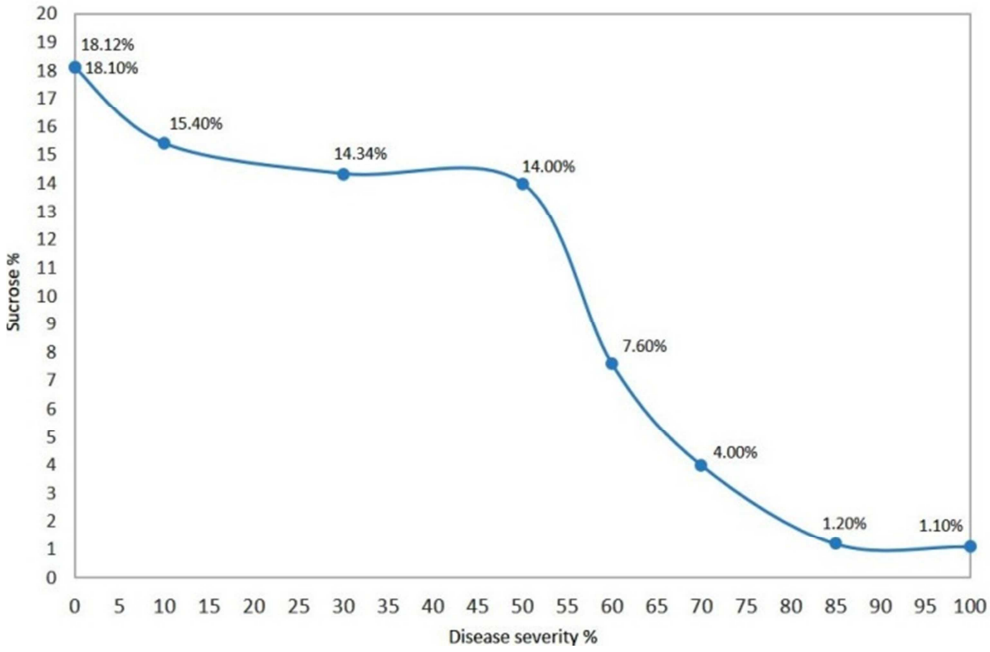

Figure 3. *Relationship between S. rolfsii root rot severity of sugar beet and sucrose content.*

4. Discussion

Sugar beet is one of the hosts that attacked by *S. rolfsii* resulting in damping off and rot of petioles, leaves and roots in tropical, subtropical and other warm temperate regions [4, 6, 7, 8, 16]. In addition to ecological factors affecting disease prevalence such as; soil temperatures, soil pH and relative humidity [11, 24, 28], density and longevity of fungal propagules also play an important role in disease development [29]. Thus current investigation showed that 70% of damping off and 66% of root rot were resulted from only 40g of fungal mass/10kg of soil compared with only 36.67% of damping off and no or negligible root rot were obtained even with 500 sclerotia/10kg of soil. So, the fungal mass herein was more efficient than sclerotial inoculation. However, the fungal pathogen produces a prolific mycelium on the host surface prior to its penetration into the host tissue within 2 to 10 days [30]. Moreover, sclerotia need energy and/or exogenous food base of nonliving matter for their germination, hyphal growth and successful penetration of host tissue [29; 31]. Thus, *S. rolfsii* mycelium mass is a potent way for soil inoculation, particularly for screening of sugar beet resistant varieties, ensuring quick host attack and immediate tissue decay with no skipping from infection [32].

The risk of *S. rolfsii* comes from its ability to produce profuse mycelia, abundant sclerotia persist in soil for several years and attack a wide range of hosts [5, 7, 9, 16]. Besides, it has frequently been documented to be capable of attacking sugar beet from sowing to maturity [4, 7, 16]. So, current investigation aimed to find an effective way for protecting sugar beets from this fungus or at least minimizing its impact from sowing to harvest. *In vitro* screening of the tested materials exhibited that mycelial growth of *S. rolfsii* was completely inhibited. This finding was consistent with the documented results of fungicides (Tebuconazole, Difenoconazole, prpiconazole) [24, 33, 34], bio-agent (*P. fluorescens* and *P. putida*) [22, 35, 36] and seaweeds (*Ulva fasciata*) [16] as *in vitro* active agents against *S. rolfsii*. Moreover, sodium and potassium silicate compounds have previously been evaluated against some soil born plant pathogens of sugar beet [37, 38]. Despite the *in vitro* efficacy of all tested materials in present study, their activities were varied *in vivo* according to the application way. On the other hand, *P. fluorescens*, *P.*

putida, Ulva fasciata, sodium and potassium silicate were completely failed to protect sugar beet roots in the current study although they were reported as effective agents against *S. rolfsii* in different hosts [25, 37]. Tipo top fungicide (tebuoconazole) was generally the most effective treatment in 2020/2021 trial, providing up to 80% reduction in sugar beet seedlings mortality followed by potassium silicate and Score (Difenoconazole) fungicide. However, usage of these materials as seed soaking only, particularly with TT, was not enough to protect sugar beet roots more than 45 days since the root rot severity exceed 70% [38]. So, interval applications of soil drenching following the seed soaking were suggested in 2021/2022 trail to magnify the effect of promising materials. As a result, seed soaking only by TT (Tebuoconazole) and/or seed soaking followed by soil drenching either by TT or PS after 35 days from sowing were generally the most effective treatments in reducing sugar beet mortality in the pre and post emergency. Additionally, full protection against *S. rolfsii* root rot and increased weights of sugar beet roots were achieved by seed soaking followed by one or more times of soil drenching by TT. These findings could be attributed to the documented inhibitory effects of triazole class fungicides including active ingredients such as tebuoconazole, propiconazole and difenoconazole against *S. rolfsii* [34, 39, 40]. These fungicides are belonged to demethylation inhibiting group (DMI) which has ability to inhibit C14-demethlase enzyme. This enzyme plays a role in sterols (such as ergosterol) production which is essential for membrane structure and function of the fungus cell, thus its inhibition resulted in abnormal growth of the fungus and eventually death [38].

Deterioration of sugar beet roots due to *S. rolfsii* infection, subsequent yield loss and reduced sucrose content were shown in the present study. The fungal penetration occurs after production of pectinolytic and cellulolytic enzymes that degrade the outer cell layer of the host tissues resulting in a distorted appearance of the roots [41] that finally become unsuitable for sugar extraction or animals feeding. Unfortunately, completely resistant cultivars of sugar beet to *S. rolfsii* have not reported until now. So, chemical fungicides are still the fast and important way for management of sugar beet root rot caused by *S. rolfsii*.

5. Conclusion

We could be conclude that active fungal mass of *S. rolfsii* provided a potent inoculum affecting sugar beets from sowing to harvesting in this study followed by sclerotia. The reduction in sugar beet root yield due to varied root rot severity levels could be illustrated along with sucrose content. TT was the most effective treatment providing full protection against the fungal attack when applied as a drenching for one time or more after the seed soaking. Further studies should be conducted to determine the most effective rate and methods of application of such control material reaching the maximum benefit of sugar beet crop under Egyptian farming conditions particularly with nowadays climate change.

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