

EFFECT OF SOME FUNGICIDES AND MULBERRY TOTAL SAPONIN EXTRACT ON FUNGAL AND BACTERIAL DISEASES

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Abstract

Copper sulfate, thiophanate-methyl, carboxin and mulberry total saponin extract were added to PDA medium at concentrations of 25, 50, 100, 200, 400 and 800µg/ml for copper sulfate against *Al. alternata*, of 0.5, 1, 5, 10, 25 and 50µg/ml for thiophanate-methyl against *B. cinerea*, of 0.5, 1, 5, 10, 25, 50 and 100µg/ml for carboxin against *R. solani*, *S. rolfisii* and *F. oxysporum* and of 250, 500, 1000 and 2000µg/ml for total saponin (TS) against the previous fungi as well as against *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas sp.*, *Proteus vulgaris*, *Streptomyces sp.* and *Erwinia amylovora*. The *in vitro* growth response (inhibition of linear growth), EC₅₀, EC₉₀ and slope of fungi and inhibition zone of bacteria were tested. Other experiments had been done in the greenhouse to test the effect of carboxin fungicide and mulberry TS extract on the change percentage of number healthy seedling, shoot & root length and dry weight for tomato seedling after 35days from sowing in sterilized and infected soil with *S. rolfisii* fungus. The results of laboratory experiments indicated that both of TS and carboxin fungicide have the same trend which they were more toxic against *S. rolfisii* than both *R. solani* and *F. oxysporum* at different concentrations. Also thiophanate-methyl and copper sulfate fungicides were more toxic than TS against *B. cinerea* and *Al. alternata*, respectively. Total saponin at different concentrations, had moderate activity against *Bacillus subtilis*, *Pseudomonas sp.* and *Proteus vulgaris*, weak activity against *Escherichia coli* and *Streptomyces sp.* and no effect against *Erwinia amylovora*. The greenhouse experiment results indicated that adding TS or carboxin to tomato seeds before sowing in both of sterilized and infected soil, enhanced the growth seedlings system included shoot and root length. TS was more effective than carboxin on the growth seedlings system. While carboxin fungicide expressed moderate effect on the growth seedlings system, it represented the best highest percentage in healthy seedlings.

INTRODUCTION

Fungicides act as a major factor in the agricultural production as they are used to protect plants from several fungal diseases which accordingly lead to a lot of fungi such as *Al. alternata* (black rot), *B. cinerea* (gray mold), *F. oxysporum* (fusarium wilt), *R. solani* (damping-off) and *S. rolfisii* (southern blight).

As long as the improper use of fungicides causes many problems for both human beings and environment, so using some of the natural components that can

fight against fungal and bacterial diseases is an essential matter that must be taken in consideration accordingly using saponin as an environmental non polluting fighter against both fungal and bacterial diseases.

Saponin is given to a group of natural glycosides occurring primarily but not exclusively in plants. It could be also produced by some marine animals, such as sea slug and starfish. Saponins are widely distributed in nature, being present in more than 1730 plant species belonging to 104 families. Of these species 627 were found to contain triterpenoid saponins and 127 to contain steroidal saponins i.e. triterpenoid saponins are most abundant in plant kingdom. The pentacyclic triterpenoid saponins are of rare occurrence in monocotyledons. They are more frequent in dicotyledons, being abundant in Caryophyllaceae, Sapindaceae, Polygalaceae, Sapotaceae and of common occurrence in Phytolaccaceae, Zygophyllaceae, Oleaceae, Psorbaceae, Sraliaceae, Linaceae, Rutaceae (Fenwick *et al.*, 1991).

The present investigation was undertaken with the aim to study the biological activities of total saponin isolated from root bark of mulberry as an antifungal and antibacterial agent.

MATERIALS AND METHODS

Materials:

Total Saponin

Total saponin was prepared according to Ukpabi and Ukpabi, (2003). Root bark of mulberry was dried airy and ground to fine powder. The powder was soaked in petroleum ether (40-60°C) for 24 hours to remove fats. The defatted powder was extracted with 50% aqueous methanol till exhaustion. The methanolic extract was re-extracted with *n*-butanol several times. The butanolic extract was evaporated till dryness. The residue was dissolved in small amount of alcohol. Then, the total saponin precipitated by addition of large amount of acetone (1:5 v/v).

Bacterial strains

Antibacterial activity of total saponin was investigated against *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas sp.*, *Proteus vulgaris*, *Streptomyces sp.* and *Erwinia amylovora*. All tested bacterial strains were maintained on nutrient agar medium (Oxoid) and were subcultured every two weeks.

Fungal strains

Antifungal activity of total saponin and fungicides (shown in Table 1) was investigated against *Alternaria alternata*, *Botrytis cinerea*, *Fusarium oxysporum*, *Rhizoctonia solani* and *Sclerotium rolfsii*. All tested fungal strains were maintained on

Potato Dextrose Agar (PDA) medium (Oxiod CM 139) and were subcultured every two weeks.

Table 1. Trade, common and chemical names and active ingredient of tested fungicides.

Traditional name	Common name & Active ingredient	Chemical name	Formulation
Del cup 6%	Copper sulfate (23.5%)	Copper sulfate pentahydrate	Liquid
Topsin-M 70%	Thiophanate-methyl (70%)	Dimethyl (1,2phenylene) bis (iminocarbono-thioyl)bis carbamate	Waterable powder
Vitavax -T	Carboxin (75%)	5,6-dihydro-2-methyl - 1,4- oxathi-ine-3-carboxamide bis (dimethyl thio-carbamayl) disulfide	Waterable powder

Methods:

Laboratory experiments

Antibacterial activity

Antibacterial activity was determined by measuring the inhibition zone diameter (mm) using agar diffusion method according to Deans and Noble, (1995). Bacterial standard inoculums were prepared by adding 10ml of sterile water to slant culture (2-3days old) and shaken for 15min. The suspension of culture was collected and used as standard inoculums. 100ml of sterile medium were inoculated with 1ml standard inoculums of tested bacteria. Then, the inoculated medium was poured into sterilized Petri-dishes (9cm). When the culture become solid, wells of 9mm diameter were made by cork borer, then 50µl of total saponin were transferred to each well. Dishes were incubated at 37°C for 24 hrs.

Antifungal activity

Effect of different concentrations of tested fungicides and total saponin on the linear growth of some phytopathogenic fungi.

This experiment was conducted to evaluate the effectiveness of total saponin and tested fungicides at different concentrations on the mycelia growth of *Al. alternata*, *B. cinerea*, *F. oxysporum*, *R. solani* and *S. rolfsii*. Fifty milliliters of PDA medium were transferred to conical flask (150ml) and autoclaved at 121°C for 20min. Adequate drops of 25% of lactic acid were added to medium after sterilization and before pouring in Petri-dishes to prevent bacterial contamination. Dilutions of each

fungicide or total saponin were prepared as w/v or v/v by dissolving an appropriate amount of each of them in 10mls of sterile water. One milliliter containing each diluted fungicide and total saponin were added to the flasks containing PDA medium just before solidifying and shacked before pouring to get concentrations 25, 50, 100, 200, 400 and 800µg/ml of copper sulfate against *Al. alternata*, 0.5, 1, 5, 10, 25 and 50µg/ml of thiophanate-methyl against *B. cinerea*, 0.5, 1, 5, 10, 25, 50 and 100µg/ml of carboxin against *R. solani*, *S. rolfsii* and *F. oxysporum* and 250, 500, 1000 and 2000µg/ml of total saponin against the previous fungi. All concentrations were expressed as active ingredient. A zero concentration treatment was prepared for each fungus used as untreated check. The different concentrations of each fungicide and total saponin were then poured in Petri-dishes 9cm diameter. Dishes for each concentration were inoculated at the center with agar discs (5mm in diameter removed from the margins of 7days old culture) of the above mentioned pathogenic fungi. Five plates were used for each particular treatment as replicates. All plates were incubated at room temperature ($25\pm 2^{\circ}\text{C}$). The radial growth of each tested fungus was measured in mm when its mycelial growth covered the surface of the medium in untreated check by calculating the average of two perpendicular diameters of the fungal growth (Abd El-Ghany, 2001). The data of linear growth fungi as affected by the treatments were tabulated. Also, the estimated effective concentration of the materials tested to give 50 and 90% inhibition radial growth (EC_{50} and EC_{90}) for each fungus was determined. The percentage of inhibition was calculated.

Greenhouse experiment.

A. Preparation of pathogen fungus inoculum.

For preparing fungal inocula (*S. rolfsii*), Erlenmeyer flasks (500ml) containing cornmeal-sand medium (25g clean sand, 75g corn and enough water to cover the prepared mixture) were autoclaved for 30min. at 121°C , inoculated by 4 discs (5mm) of fungus taken from the previous culture and then incubated at room temperature ($25\pm 2^{\circ}\text{C}$) for two weeks.

B. Pots sterilization.

Plastic pots (15cm diameter) were sterilized by immersing in 10% formalin solution for 15min. and left for a week to get rid of formalin.

C. Clay sterilization.

Loam-soil were autoclaved an hour at 121°C three days frequency.

D. Seed treatments.

Tomato seed variety Castle rock was treated with total saponin or fungicide carboxin. The total saponin treated tomato seed were prepared by soaking tomato seed in EC_{50} of total saponin (118.19µg/ml) for 5mins, after that seeds were put on

the filter paper to dry. Fungicide carboxin treated tomato seeds were prepared by mixing seeds with carboxin at recommend concentration (3g/kg seeds) and glue suspension as adhesive material in closed glass container and vigorously shake-up 10mins. Seeds were put on filter paper to dry.

E. Experiment design.

An experiment was carried out under greenhouse conditions. Plastic pots were divided into two groups each of 30pots. The pots in the first group were filled with 750g sterilized uninfected loam-soil per pot. The pots in the second group were filled with the same amount of sterilized loam-soil after infected by mixing it with the prepared fungal inocula of *S. rolfsii* at 3% of soil weight (w/w) and watered regularly for ten days before sowing to insure the distribution and growth of inoculums. (Abd El-Ghany, 2001). Each group was divided into three subgroups each of ten pots as follows:

(I) Pots were sown with untreated tomato seeds (untreated check).

(II) Pots were sown with total saponin-treated tomato seeds.

(III) Pots were sown with carboxin-treated tomato seeds.

Each pot was sown with 5 tomato seeds treated or untreated. After 35 days of sowing, results were calculated as the percent healthy and unhealthy tomato seedlings and corrected with control group. Fresh and dry weight and shoot and root length of seedlings were determined.

RESULTS AND DISCUSSION

Laboratory experiments

Antibacterial activity

Effect of total saponin (TS) at different concentrations on growth of *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas sp.*, *Proteus vulgaris*, *Streptomyces sp.* and *Erwinia amylovora*.

Effect of total saponin (TS) at different concentrations i.e. 250, 500, 1000 and 2000µg/ml on the growth of *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas sp.*, *Proteus vulgaris*, *Streptomyces sp.* and *Erwinia amylovora*, as represented in Table (2). Data in Table (2) proved that, *Escherichia coli* and *Streptomyces sp.* were not affected by TS at concentrations 500 and 250µg/ml, respectively. At different concentrations of TS caused inhibition zone of growth for *Bacillus subtilis*, *Pseudomonas sp.* and *Proteus vulgaris*. The highest effect was observed at high concentration of TS (2000µg/ml) recording 16.0, 16.0 and 17.0mm for *Proteus vulgaris*, *Bacillus subtilis* and *Pseudomonas sp.* respectively. While *Erwinia amylovora* was not affected by TS at different concentrations.

The obtained results concluded that, TS at different tested concentrations had moderate activity against *Bacillus subtilis*, *Pseudomonas sp.* and *Proteus vulgaris*. Also, it had weak activity against *Escherichia coli* and *Streptomyces sp.* and no effect against *Erwinia amylovora*. These results were similar to that reported by Okunji *et al.*, (1990), El-Hady *et al.*, (1994) and Boguslavskii *et al.*, (2001).

Table 2. Effect of total sponin (TS) on growth of *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas sp.*, *Proteus vulgaris*, *Streptomyces sp.* and *Erwinia amylovora*.

Concentration (µg/ml)	<i>Bacillus subtilis</i>	<i>Escherichia coli</i>	<i>Pseudomonas sp.</i>	<i>Proteus vulgaris</i>	<i>Streptomyces sp.</i>	<i>Erwinia amylovora</i>
	Inhibition Zone in (mm)					
2000	16.0	8.0	17.0	16.0	10.5	0.0
1000	15.0	5.0	15.0	14.0	7.0	0.0
500	12.5	0.0	15.0	12.0	5.0	0.0
250	10.5	0.0	10.0	10.0	0.0	0.0
0	0.0	0.0	0.0	0.0	0.0	0.0

Antifungal activity

Effect of total saponin (TS) and carboxin fungicide on the linear growth of *R. solani*, *S. rolfsii* and *F. oxysporum*

The effect of TS and carboxin fungicide at their tested concentrations on the linear growth of *R. solani*, *S. rolfsii* and *F. oxysporum*, is shown in Table (3). Results in Table (3) stated that, both TS and carboxin reduced the linear growth of three fungi and the highest concentration of both showed the highest inhibition of the fungal growth. The inhibition percentage of the linear growth of *R. solani*, *S. rolfsii* and *F. oxysporum* on PDA medium treated with 2000µg/ml of TS was recorded 69.2, 81.1, and 48.3, respectively, of that obtained in untreated medium. The results proved that *F. oxysporum* seems to be resistant or more tolerant to TS than *R. solani* and *S. rolfsii*. On the other hand, *S. rolfsii* was the most sensitive to TS at different tested concentrations comparing with *R. solani* and *F. oxysporum*. The EC₅₀ values of TS were 118.9, 1056.09 and 2160.26µg/ml against *S. rolfsii*, *R. solani* and *F. oxysporum*, respectively. These results were in agreement with those obtained by Lalitha and Venkataraman, (1991), Zehavi *et al.*, (1993) and levy *et al.*, (1999).

Table 3. Effect of total saponin (TS) and carboxin fungicide at different concentrations on the linear growth of *R. solani*, *S. rolfsii* and *F. oxysporum*

Con. (ppm)	<i>R. solani</i>								
	Total Saponin (TS)				Carboxin				
	Linear growth in mm	% inhibition	EC ₅₀ (ppm)	Slope value	Con. (ppm)	Linear growth in mm	% inhibition	EC ₅₀ (ppm)	Slope value
2000	27.70	69.2	1056.09	1.7	100	0.0	100	5.02	1.17
1000	48.00	46.7			50	15.00	83.3		
500	63.80	29.1			25	25.00	72.2		
250	76.80	14.7			10	30.00	66.7		
0	90.00	0.0			5	45.00	50.0		
-	-	-			1	80.00	11.1		
-	-	-			0.5	90.00	0.0		
-	-	-			0	90.00	0.0		
<i>S. rolfsii</i>									
2000	17.00	81.1	118.19	0.39	100	0.0	100	1.30	1.30
1000	30.50	66.1			50	0.0	100		
500	37.00	58.9			25	7.00	92.2		
250	41.60	53.8			10	12.00	86.7		
0	90.00	0.0			5	22.00	75.6		
-	-	-			1	48.00	47.8		
-	-	-			0.5	65.00	27.8		
-	-	-			0	90.00	0.0		
<i>F. oxysporum</i>									
2000	46.50	48.3	2160.26	1.86	100	10.00	88.9	10.01	1.18
1000	66.10	26.6			50	20.00	77.8		
500	81.20	9.8			25	35.00	61.1		
250	85.30	5.2			10	40.00	55.5		
0	90.00	0.0			5	54.00	40.0		
-	-	-			1	75.00	16.7		
-	-	-			0.5	90.00	0.0		
-	-	-			0	90.00	0.0		

S. rolfsii was sensitive to fungicide carboxin at all concentrations under study. The linear growth of *S. rolfsii* was completely inhibited at concentration 50µg/ml. The inhibition percentage was 83.3 and 77.8 at the same concentration for *R. solani* and

F. oxysporum, respectively. The linear growth of *R. solani* was also completely inhibited at concentration 100µg/ml. On the other hand, *F. oxysporum* seems to be more tolerant to this fungicide than *R. solani* and *S. rolfsii* as it was still growing even on medium treated with 100µg/ml carboxin. The EC₅₀ of carboxin values were 5.02, 1.30 and 10.01µg/ml against *R. solani*, *S. rolfsii* and *F. oxysporum*, respectively.

Data in Table (3) indicated that the fungicide carboxin was more toxic *in vitro* against *S. rolfsii* than *R. solani* and *F. oxysporum*. These results were in agreement with those obtained by Youssef, (1990), Henriquez and Montealegre, (1992) and Abd El-Ghany, (2001).

Results in Table (3) concluded that both of TS and carboxin fungicide were of the same trend as they were more toxic *in vitro* against *S. rolfsii* than *R. solani* and *F. oxysporum* at different concentrations.

Effect of total saponin (TS) and thiophanate-methyl fungicide on the linear growth of *B. cinerea* .

The effect of TS (at concentration of 250, 500, 1000 and 2000µg/ml) and thiophanate-methyl fungicide (at 0.5, 1, 5, 10, 25 and 50µg/ml), on the linear growth of *B. cinerea* is shown in Table (4).

Data in Table (4) indicated that mycelia growth of *B. cinerea* was differentially inhibited on treated medium with TS at different concentrations (250-2000µg/ml). Such effect was more pronounced with high concentration (2000µg/ml) and the inhibition percentage was 61.4 of that obtained in untreated medium. The EC₅₀ and EC₉₀ values of TS against *B. cinerea* recorded 938.1 and 2931.6µg/ml, respectively. These results were in agreement with those observed by Singh *et al.*, (1992) and Quidde *et al.*, (1998). They suggested that saponins could be used for disease control in the field. The toxic action of saponins to fungi is associated with the ability of these compounds to complex with membrane sterols and causes pore formation.

The inhibitory effect of thiophanate-methyl fungicide at different concentrations (0.5:50µg/ml) on *B. cinerea* mycelia growth, is shown also in Table (4). Completely inhibition was achieved at concentration 50µg/ml, while, no effect was observed at concentration 0.5µg/ml. The EC₅₀ and EC₉₀ values recoded 0.89 and 25.0µg/ml, respectively.

Data in Table (4) proved that, thiophanate-methyl fungicide was more toxic *in vitro* than TS against *B. cinerea*.

Table 4. Effect of total saponin (TS) and thiophanate-methyl at different concentrations on the linear growth of *B. cinerea*

Concentration (ppm)	Linear growth in mm	% inhibition	EC ₅₀ (ppm)	EC ₉₀ (ppm)	Slope value
Total Saponin (TS)					
2000	34.70	61.4			
1000	42.00	53.3			
500	55.80	38.0	938.1	2931.6	1.39
250	67.80	14.7			
0	90.00	0.0			
Thiophanate-methyl					
50	0.00	100			
25	9.00	90			
10	15.00	83.4	0.89	25.0	1.49
5	28.00	68.9			
1	40.00	55.6			
0.5	90.00	0.0			
0	90.00	0.0			

Effect of total saponin (TS) and copper sulfate fungicide on the linear growth of *Al. alternata*.

Data in Table (5) proved that, the reduction of the linear growth of *Al. alternata* moderately increased with high concentrations of TS (1000 and 2000µg/ml). At 250, 500, 1000 and 2000µg/ml of TS, the inhibition percentages of the linear growth of this fungus recorded 15.6, 18.6, 31.3 and 50.0, respectively. The EC₅₀ and EC₉₀ values were 2000 and 4672.84µg/ml, respectively. These results were in agreement with those reported by Singh *et al.*, (1992). They demonstrated that saponin B from oats at 2.5mg/ml was highly effective against growth of *Alternaria sp.* They suggested that saponins could be used for disease control in the field.

On the other hand, the reduction of the linear growth of *Al. alternata* was pronounced with high concentrations of copper sulfate (200 and 400µg/ml). Completely inhibition of fungus growth was achieved at 800µg/ml. The EC₅₀ and EC₉₀ values were 182.81 and 620.12µg/ml, respectively. This result was in agreement with those recoded by Dewez *et al.*, (2005). They reported that copper sulfate is commonly used as fungicide in agriculture practice to protect fruits and vegetable crops.

Table 5. Effect of total saponin (TS) and copper sulfate fungicide at different concentrations on the linear growth of *Al. alternata*.

Concentration (ppm)	Linear growth in mm	% inhibition	EC ₅₀ (ppm)	EC ₉₀ (ppm)	Slope value
Total Saponin (TS)					
2000	45.00	50.0			
1000	61.80	31.3			
500	73.20	18.6	2000	4672.89	1.07
250	76.00	15.6			
0	90.00	0.0			
Copper sulfate					
800	0.00	100			
400	24.00	66.0			
200	40.00	55.6	182.81	620.12	2.33
100	65.00	27.8			
50	80.00	11.1			
25	90.00	0.0			
0	90.00	0.0			

Greenhouse experiment

Effect of total saponin (TS) and carboxin on number of healthy seedlings' shoot & root length and dry weight in tomato seedlings.

Effect of TS and carboxin on the change percentage (increase/decrease) of number of healthy seedlings' shoot & root length and dry weight for tomato seedlings after 35days from sowing in sterilized and infected soil is shown in Table (6).

Sowing tomato seeds treated with TS or carboxin in sterilized soil expressed increase in number of healthy seedlings compared to untreated seeds after 35days of sowing. Such increase was low significant ($p < 0.05$) in case of seeds treated with carboxin which reached 12.5% of that obtained in untreated seeds. On the other hand, sowing tomato seeds in soil infected with *S. rolfsii* induced highly decrease in number healthy seedlings comparing with those sown in sterilized soil which recorded 15.0 ± 1.3 and 40.0 ± 2.3 seedling, respectively, after 35days of sowing. Adding TS or carboxin to seeds before sowing in infected soil induced highly significant ($p < 0.001$) increase in number of healthy seedlings compared to untreated seeds. Such increase was more pronounced in case of carboxin which recorded 133.3% of that obtained in untreated seeds after 35days of sowing.

Data in Table (6) proved that, the increase of shoot and root length for tomato seedlings was observed after 35days of sowing seeds treated with TS or

carboxin in sterilized soil comparing with untreated. This effect was moderate significant ($p < 0.01$) and noticeable in case of treatment with TS and the change percentages of shoot and root length were 22.7 and 20.0, respectively, of that observed in untreated seeds. While, sowing tomato seeds in infected soil caused greatly decrease in shoot and root length (5.42 ± 0.3 and 1.2 ± 0.5 cm) comparing with those sown in sterilized soil (13.2 ± 1.3 and 2.5 ± 0.2 cm), respectively. Treating TS or carboxin to tomato seeds before sowing in infected soil induced significant increase in shoot and root length compared to untreated. Such increase was highly ($p < 0.01$) and moderate ($p < 0.01$) significant in case of TS and carboxin, respectively. The highest increase in shoot and root length were observed in case of sowing seeds treated with TS which reached 88.6 and 83.3%, respectively, of that detected in untreated. While, in case of carboxin, the percentage of change in shoot and root length were recorded 36.0 and 50.0, respectively, of that obtained in untreated, after 35 days of sowing in infected soil. In generally, shoot and root length of tomato seedlings in sterilized soil was still taller than those in infected soil.

Sowing tomato seeds treated with TS or carboxin in sterilized soil, resulted in significant increase in dry weight of seedlings compared to untreated tomato seeds after 35 days of sowing. Such increase was moderate significant ($p < 0.01$) and pronounced in case of seeds treated with TS comparing with untreated which reached 0.33 ± 0.001 and 0.22 ± 0.001 g/seedling, respectively. On the other hand, sowing tomato seeds in soil infected with *S. rolfsii* induced sharply decrease in dry weight of seedlings comparing with those sown in sterilized soil which equal to 0.083 ± 0.001 and 0.22 ± 0.001 g/seedling, respectively. While, adding TS or carboxin to seeds before sowing in infected soil, caused significant increase in dry weight of seedlings compared to untreated seeds after 35 days of sowing. This increase was highly significant ($p < 0.001$) and pronounced in case of sowing seeds treated with TS which recorded 80.7% of that obtained in untreated. Although, treating TS or carboxin to tomato seeds before sowing in infected soil improved in the dry weight of seedlings comparing with untreated, the dry weight still lower than those sown in sterilized soil.

Table 6. Effect of total saponin (I) and carboxin (II) on number healthy seedlings shoot & root length and dry weight in tomato seedlings.

Tretments	Sterilized soil		Infected soil with <i>S. rolfsii</i>	
	mean±SE	%	mean±SE	%
Number of the healthy seedlings				
Untreated	40.0±2.3	0.0	15.0±1.3	0.0
I-treated	42.0±4.1	5.0	30.0±8.1***	100.0
II-treated	45.0±3.2*	12.5	35.0±9.3***	133.3
Shoot length (cm)				
Untreated	13.2±1.3	0.0	5.42±0.3	0
I-treated	16.2±2.5**	22.7	10.22±1.2***	88.6
II-treated	15.4±1.8	9.8	7.37±1.0**	36.0
Root length (cm)				
Untreated	2.5±0.2	0.0	1.2±0.5	0
I-treated	3.0±0.1**	20.0	2.2±0.8***	83.3
II-treated	2.6±0.1	4.0	1.8±0.4**	50.0
Dry weight (g)				
Untreated	0.22±0.001	0.0	0.083±0.001	0
I-treated	0.33±0.001**	50.0	0.15±0.001***	80.7
II-treated	0.29±0.001*	31.8	0.12±0.001**	44.6

Results are expressed as mean ± SE for 5 replicates in each group.

% the percentage of change (increase/decrease).

*significant $p < 0.05$ ** $p < 0.01$ *** $p < 0.001$

From the above results it could be concluded that, adding TS or carboxin to tomato seeds before sowing in both of sterilized and infected soil, enhanced the growth seedlings system included shoot and root length. TS had more effect than carboxin on the growth seedlings system. While, carboxin fungicide expressed moderate effect on the growth seedlings system but it represented the best highest percentage in healthy seedlings. These results were similar to that recorded by **EI-Deeb** and Ibrahim, (1998). They demonstrated that, the seed dressing fungicide carboxin significantly decreased pre- and post-emergence damping-off and pod rot diseases and gave the highest percentages of healthy survival plants. Also, Hammouda *et al.*, (2001) might be interpret the role of fungicides and thyme extract in controlling soil borne pathogenic fungi that infect faba bean and their relationship with chitinase, peroxidase and phenolic compounds as antifungal substances in plants tissues and then subsequent increases in root & shoot lengths and dry weight of plants.

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تأثير بعض المبيدات الفطرية والصابونين الكلي علي بعض الأمراض الفطرية والبكتيرية

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الهدف الرئيسي من هذه الدراسة هو التعرف علي فعالية الصابونين الكلي المستخلص من قلف جذر شجرة التوت كمضاد للبكتيريا والفطريات الممرضة للنبات.

تم استخلاص قلف جذر شجرة التوت بواسطة ميثانول ٥٠% ثم الترشيح وتبخير الميثانولز استخلاص المتبقي بالبيوتانول ثم تركيزمستخلص البيوتانول واعادة ذوبان المتبقي في كمية صغيرة من الكحول وأضافته تدريجيا الي الاسيتون بنسبة (١ : ٥ حجم/حجم) للحصول علي راسب الصابونين الكلي. أجريت تجارب معملية لتقدير فعالية الصابونين الكلي والمبيدات الفطرية كربوكسين، ثيوفينات الميثيل و كبريتات النحاس بتركيزات مختلفة علي النمو الميسليومي للفطريات *Rhizoctonia solani*, *Sclerotium rolfsii*, *Fusarium oxysporum*, *Botrytis cinerea* and *Alternaria alternata* وكذلك استخدم الصابونين الكلي كمضاد للبكتيريا حيث اختبرت فعاليته ضد بكتيريا *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas sp.*, *Proteus vulgaris*, *Streptomyces sp.* and *Erwinia amylovora* باستخدام طريقة الأنتشار في الآجار. أجريت تجربة بالصوبة لتقييم الصابونين الكلي للتحكم في مرض عفن جذور نبات الطماطم والذي يسببه فطر *S. rolfsii* مقارنة بالمبيد الفطري كربوكسين في تربة معقمة واخري تم تلويثها بالفطر. أخذت قياسات طول الجذور والبادرات والوزن الجاف لهذه البادرات بعد ٣٥ يوم من الزراعة. وقد أظهرت النتائج أن الصابونين الكلي بتركيزاته المختلفة (٢٥٠، ٥٠٠، ١٠٠٠، ٢٠٠٠ ميكروجرام/ملي) له فعالية متوسطة ضد بكتيريا *Bacillus subtilis*, *Pseudomonas sp.* and *Proteus vulgaris*. كما أظهر فعالية ضعيفة ضد بكتيريا *Erwinia amylovora* and *Streptomyces sp.* لم تكن فعالة ضد بكتيريا *Erwinia amylovora* كما أوضحت النتائج أن كلا من الصابونين الكلي والمبيد الفطري كربوكسين لهما تأثير أكثر سمية ضد فطر *S. rolfsii* مقارنة بالفطرين *R. solani* and *F. oxysporum* عند التركيزات المختلفة. وأوضحت النتائج أيضا أن المبيد الفطري ثيوفينات الميثيل أكثر سمية من الصابونين الكلي ضد فطر *B. cinerea* حيث تحقق التثبيط الكلي للنمو الميسليومي لهذا الفطر عند تركيز ٥٠ ميكروجرام/ملي. وبالمثل كان المبيد الفطري كبريتات النحاس أكثر سمية من الصابونين الكلي ضد فطر *Al. alternata* حيث تحقق التثبيط الكلي للنمو الميسليومي للفطر عند تركيز ٨٠٠ ميكروجرام/ملي. وأظهرت نتائج التجربة التي أجريت بالصوبة أن زراعة بذور الطماطم المعاملة بالصابونين الكلي أو المبيد الفطري كربوكسين في تربة معقمة أحدثت زيادة معنوية في الوزن الجاف (وزن الجذر والمجموع الخضري) لبادرات الطماطم بعد ٣٥ يوم من الزراعة وكذلك أظهرت النتائج أن معاملة بذور الطماطم بكل من الصابونين الكلي أوالمبيد الفطر كربوكسين قبل زراعتها في تربة معدية بالفطر أحدثت تحسن ملحوظ في القياسات مقارنة بالبذور الغير معاملة.