

**BIOLOGICAL EFFECTS OF SOME PLANT EXTRACTS
AND ANTAGONISTIC FUNGI AGAINST SOME
SOIL BORN FUNGI**

BY

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ABSTRACT

This study aims to evaluate the efficiency of some plant extracts, i.e. *Solanum nigrum*, *Ammi visnaga*, *Piper nigrum* and *Schinus terbenthiofolius* as fungicides, in addition to study the antagonistic effect of some other fungi against pathogenic fungi i.e. *Trichoderma harzianum*, *Trichoderma viride* and *Gliocladium deliquescens*. Results showed that : Aqueous extract of *Schinus terbenthiofolius* was the most potent one against *R.solani*, followed by aqueous extract of *Solanum nigrum*, while aqueous extract of *Ammi visnaga* was the most effective against *S.rolfsii*, *M.phaseolina*, followed by *Solanum nigrum* aqueous extract. Also, aqueous extract of *Ammi visnaga* had the strongest activity against *P.debaryanum*, followed by *Schinus terbenthiofolius* aqueous extract. *Piper nigrum* solvent extract caused strong inhibition in the mycelial growth of *R.solani*, *S.rolfsii* and *M.phaseolina*, followed by solvent extract of *Ammi visnaga*. Solvent extract of *Piper nigrum* was the most effective compound against *P.debaryanum* followed by solvent extract of *Solanum nigrum*. Results of biological control revealed that *T. harzianum* gave the most antagonistic effect against *M.phaseolina* and *S.rolfsii*. *T. viride* gave the most antagonistic effect against *R. solani* and *P.debaryanum*. The joint toxic effect was studied between carboxin/thiram and other compounds. Synergistic effects were observed in some cases: The case of *R.solani* when combined with aqueous extracts of *Ammi visnaga*, *Piper nigrum* and *Schinus terbenthiofolius*, and solvent extract of *piper nigrum*. The most synergistic effect was in the case of carboxin/thiram+ solvent extract of *Piper nigrum*. In the case of *S.rolfsii*, when using the combination of carboxin/thiram with aqueous extracts of *Piper nigrum*, *Schinus terbenthiofolius* and *Solanum nigrum*; and solvent extract of *Piper nigrum*. The most synergistic

effect was in the case of carboxin/ thiram + aqueous extract of *Piper nigrum*. In the case of *M.phaseolina*, combinations of carboxin/thiram with both *Ammi visnaga* and *Piper nigrum* aqueous extracts gave synergistic effects. In the case of *P.debaryanum*, when carboxin / thiram was combined with solvent extracts of *Ammi visnaga*, *Piper nigrum* and *Solanum nigrum*. The most synergistic effect was in the case of carboxin / thiram + solvent extract of *Piper nigrum*. Carboxin/thiram at low concentration increased the efficacy of biological control against *R.solani*, *S.rolfsii* and *P.debaryanum*. The antagonistic effect was decreased in the case of *M.phaseolina*. *T. harzianum* gave the most antagonistic effect against *R. solani*, *S.rolfsii* and *P.debaryanum*. *T. viride* gave the most antagonistic effect against *M.phoseolina*.

INTRODUCTION

Soybean is affected by various diseases. Damping-off and root-rot diseases are the most important diseases that affect plant stand causing great losses in soybean crop, total nitrogen and protein content in soybean seeds. Constant use of synthetic pesticides for controlling diseases has resulted in several environmental problems such as long persistence period (Beye, 1978), pollutive effects (Dubey and Mall, 1972), phytotoxicity (Fawcett and Spencer, 1970), teratogenicity (Javoraska, 1978) and carcinogenicity (Epstein *et al.*, 1967). These factors emphasize the need for alternative methods to control disease (Wilson *et al.*, 1987). Plant extracts and plant oils are effective and alternative sources of fungi toxic chemicals showing considerable promise. Those compounds generally were inhibitory to growth and spore germination of the fungi and were potent at very low concentration (Omar *et al.*, 1993). Biological control of plant pathogens is becoming an important for plant disease management and several successful attempts have been made to control the pathogens by using *Trichoderma* spp. and / or *Gliocladium* spp. , which attack the mycelium which causes plant diseases. Different antagonistic fungi varied in their action. This variation may be due to the difference in the ability of each antagonist to grow, and to produce toxic substances. These also correlated with antagonistic ability of parasitizing the hypha of pathogens and the antibiosis potential (Elad *et al.*, 1982, 1983 and Upadhyay and Mukhopadhyay, 1986). The present study aims to investigate the efficacy of some solvent plant

extracts against four pathogenic and three antagonistic fungi (biocontrol agents) in vitro, (effect of antagonistic fungi on growth of pathogenic fungi), Joint toxic effects of carboxin /thiram with the tested compounds against the tested pathogenic fungi in vitro and the effect of carboxin/thiram fungicide on the efficacy of biological control

MATERIALS AND METHODS

1-Fungicide

Vitavax-200 (75% W.P) consists of (37.5% thiram + 37.5% Vitavax) :

- a) Vitavax (37.5% W.P) 5,6 dihydro - 2 methyl - N- phenyl - 1.4 oxathion -3-carboxamide. Trade Name : Vitavax. Common name carboxin
- b) Thiram (37.5 W.P) bis (dimethyl thio -carbonyl) disulphide
Common Name : TMTD

2- Tested plant extracts :

2-1 Plant materials :

Samples of plants shown in Table (1) were collected from the farm of faculty of Agriculture Kafr-El-sherikh, Tanta university. They were identified by specialists. Black pepper (*Piper nigrum*) was purchased from local market.

Table (1) : Plant materials used:

English Name	Scientific Name	Family	Part used	Solvent system or extractives
Pick tooth	<i>Ammi visnaga</i>	Umbelliferae	Leaves	Ethanol/acetone (1:1), Water.
Black night	<i>Solanum nigrum</i>	Solanaceae	Leaves	Ethanol/acetone (1:1), Water
Barazilian pepper	<i>Schinus terbenthio-folius</i>	Anacardiaceae	Leaves	Ethanol/acetone (1:1), Water.
Black Pepper	<i>Piper nigrum</i>	Piperaceae	Seeds	Ethanol/acetone (1:1), Water.

2-2 Preparation of Plant - extracts:

a) Solvent plant extracts:

The plant materials (mentioned in table 1) were air dried at room temperature and ground with blender into fine powder. Batches of 100

gm from powdered leaves of *Ammi visnaga*, *Solanum nigrum* and *Schinus terbenthiofolius* were macerated in 600 ml of ethanol and acetone at ratio of 1:1, 100gm of dried powdered seeds of *piper nigrum* was macerated in 300 ml of ethanol and acetone at ratio of 1:1. Maceration of both was done for a period of 5 days. During the maceration periods the samples were shaken for 5 hours using an electrical shaker. Extracts were filtered and the filtrates were sterilized by Seitz filter.

b) Aqueous plant extracts:

The same method of preparation of solvent plant extracts was used, but in water instead of solvents.

3- Tested fungi:

Four pathogenic fungi and three antagonistic fungi were used in these experiments namely:

a-Pathogenic fungi : *Rhizoctonia solani*, *Sclerotium rotfsii*, *Macrophomina phaseolina* and *Pythium debaryanum*.

b-Antagonistic fungi : *Trichoderma harzianum*, *Trichoderma viride* and *Gliocladium deliquescens*.

4- Biological tests

4-1 *In vitro* studies :

4-1.1 *Fungicidal activity of plant-extracts*

A laboratory study was performed to examine the sensitivity of the fungi to plant-extracts. Four concentrations of plant-extracts were used, i.e. 1.0, 10, 100 and 1000 ppm. The required concentrations were obtained by adding the appropriate amount of stock solution used to 100 ml portions of autoclaved PDA cooled to about 45°C. Four petri-dishes, 9 cm diameter, were used as replicates for each concentration. After solidification of the medium, each dish was inoculated centrally with a mycelial disk (0.5 cm in diameter) taken from the cultures of each fungus (7 days – old). Dishes were incubated at $28 \pm 1^\circ\text{C}$ for 4 days and colony diameters were measured till the untreated controls had just covered plate. Linear growth was measured in cm. by taking the average of two perpendicular diameter. The percentage of inhibition (I%) was calculated according to Topps and Wain equation (1957) as follows:

$$I \% = \frac{A-B}{A} \times 100$$

Where :

I % = Percent of inhibition.

A = Mean diameter growth in the control .

B = Mean diameter growth in a given treatment.

4-1.2 Antagonism between pathogenic fungi and antagonistic fungi (biocontrol agents) under laboratory conditions:

This experiment was carried out to study the relationship between the tested pathogenic fungi and antagonistic fungi. Petri-dishes (9cm in diameter) each contains 15 ml of PDA medium were used. Each petri-dish was divided into two equal halves, the first half was inoculated with a disk (0.5 cm in diameter) of the tested antagonistic fungi and the second half was plated with a similar disk of the pathogenic fungi. Plates inoculated only with the pathogenic fungi acted as control. Each treatment was replicated four times. All petri-dishes were incubated at 28±1°C and observed daily. After 4 days of incubation , when the pathogenic fungi almost covered the surface of the medium in control treatment, the percentage of inhibition (I%) was calculated as mentioned before.

4-1.3 Combinations

4-1.3.1 Combinations of the tested compounds :

IC₂₅ of carboxin/thiram (Vitavax-200) as a traditional fungicide which exhibited the highest or the lowest effect on the pathogenic fungi or the antagonistic fungi, respectively, was mixed with IC₂₅ from every extract alone against the tested pathogenic fungi. The Joint toxic action of the different combinations was evaluated by the following equation (Mansour *et al* (1966)):

$$\text{Co-Toxicity Factor} = \frac{\text{Obs. I \%} - \text{EXP. I \%}}{\text{EXP. I \%}} \times 100$$

Where :

Obs. I % = Observed I%

EXP. I % = Expected I%

This factor was used to classify the results into 3 categories :

- A positive Factor of 20 or more meant synergism.
- A negative factor of -20 or more meant antagonism.
- An intermediate value of (-20 and +20) was considered as an additive effect.

4-1.3.2 Effect of carboxin/thiram as traditional fungicide on enhanced efficacy biocontrol agents.

IC₂₅ from the fungicide was added to PDA medium before solidification. Method as described previously for the study of antagonism between pathogenic fungi and antagonistic fungi was used in this experiment. Results were statistically analyzed according to **Finney (1952)**, . Analysis of variance. L.S.D (Least significant difference) according to (**Snendecor (1965)**). Correction for the control mortality was made by using **Abbott's formula (1925)**.

RESULTS AND DISCUSSION

1- Fungicidal activity of the tested aqueous plant extracts against pathogenic fungi:

Data in Table (2) showed that *Schinus terbenhiofolius* extract was the most potent against *R.solani* ,followed by *Solanum nigrum* and *Piper nigrum* while *Ammi visnaga* extract showed the lowest toxic effect against *R.solani* .In the case of *S.rolfsii*, results showed that *Ammi visnaga* extract was the most effective one, followed by *Solanum nigrum* and *Piper nigrum*. *Schinus terbenhiofolius* was the least toxic extract against *S. rolfsii*.*Ammi visnaga* was the most potent extract against *M.phaseolina*, followed by *Solanum nigrum* , while *Schinus terbenhiofolius* and *Piper nigrum* extract were the least effective. Results revealed that *Ammi visnaga* and *Schinus terbenhiofolius* extracts were the most toxic extracts against *P.debaryanum*, followed by *Solanum nigrum* and *piper nigrum* against the same fungus .Data presented in Table (2) showed that *Ammi visnaga* extract was the most toxic extract against the pathogenic fungi followed by *Schinus terbenhiofolius*, *Solanum nigrum* and *Piper nigrum*, except for *R.solani* which was the highly sensitive to *Schinus terbenhiofolius*. Several authers reported that aqueous plant extracts showed fungicidal effects against numerous

fungi i.e. *R.solani*, *M. phaseolina*, *S.rolfsii*, *P.debaryanum*, *F. oxysporum* and *Alternaria alternata* (Carcia and Lawas (1990), Warr *et al.*, (1992), Poswal *et al.*, (1993), Pandey and Dubey (1997) and Tasleem *et al.*, (1998)). El-Shoraky (1998), reported that aqueous extract of *Ammi visnaga* strongly inhibited the radial growth of *S.rolfsii*, *R.solani*, *F. solani* and *F. poae*.

2- Fungicidal activity of the tested solvent plant-extracts against Pathogenic fungi:

Data presented in Table (2) showed that *Piper nigrum* extract was the most toxic solvent extract against *R. solani*, followed by *Ammi visnaga* and *Solanum nigrum*, while *Schinus terbenthiofolius* was the least toxic extract against *R.solani*. *Piper nigrum* was the most potent extract against *S.rolfsii*, followed by *Ammi visnaga*, while *Schinus terbenthiofolius* and *Solanum nigrum* was the least toxic extract. In the case of *M.phaseolina*, results revealed that *Piper nigrum* was the most effective extract against the fungus. On the other hand, *Ammi visnaga* and *Solanum nigrum* were moderately toxic extracts against *M.phaseolina*. *Schinus terbenthiofolius* was the least toxic extract against *M. phaseolina*. Concerning the fungicidal activity of the tested solvent plant extracts on *P.debaryanum*, the data revealed *Schinus terbenthiofolius* extract showed the lowest toxic effect against the tested pathogenic fungi. This results were in accordance with the results of Carcia and lawas (1990), who found that the garlic and *Piper nigrum* extracts were effective against *R.solani* and *Sclerotium sp.*

3- Effect of. antagonistic fungi on growth of pathogenic fungi *in vitro*:

Data in Table (3) revealed that *T. viride* inhibited growth of *R. solani* more than *T.harzianum* and *G. deliquescens*. *T.harzianum*, *G. deliquescens* and *T.viride* reduced the radial growth of *S.rolfsii* compared with the control. *T. harzianum* was most effective on *M.phaseolina*, followed by *T.viride*, while *G.deliquescens* was the least effective one against the same fungus. *T.viride* was the most effective one against *P.debaryanum*, followed by *G. deliquescens*, while *T.harzianum* was the least effective one against *P.debaryanum*. Data revealed that different antagonistic fungi varied in their action against the tested pathogenic fungi. This variation may be due to the difference in the ability of each antagonist to grow, and to

produce toxic substance. These results agreed with that of Mohameud and Abo-Raya (1993a) They found that *T. harzianum* grew faster than *S.rolfsii*, *R.solani* and *F. oxysporum f. lycopersica* Howell and Stipanovic (1995) reported that production of antifungal antibiotics gliotoxin and gliovirin by the biocontrol fungus *Gliocladium virens* has been associated with its efficacy as a biocontrol agent of *R.solani* and *P.ultimum*. Shalaby et al., (1997) found that *T.viride* was the most antagonistic fungus against *R. solani* and *M. phaseolina*.

Table (2) :Effect of some plant- extracts (aqueous and solvent) on the linear growth of pathogenic fungi (*R.solani* , *S.rolfsii* , *M.phaseolina* and *P.debaryanum*) on PDA medium in vitro (Four days old)

Plant extract	Fungi	I C ₅₀	Confedence limits		Slope
			Lower	Higher	
Aqueous Plant extract <i>Ammi visnaga</i>	<i>R.solani</i>	80714.01	4215.81 -	7.057x10 ⁴	0.25
	<i>S.rolfsii</i>	1720.22	112.87 -	6.22x10 ⁸	0.15
	<i>M.phaseolina</i>	120.21	44.45 -	399.34	0.43
	<i>P.debaryanum</i>	5051.78	722.42-	4.7x10 ⁵	0.3
<i>Schinus terbenlthiofolius</i>	<i>R.solani</i>	141.8	61.92-	400.25	0.49
	<i>S.rolfsii</i>	10661.65	1069.29-	4.76x10 ⁶	0.26
	<i>M.phaseolina</i>	4.79x10 ⁵	11127.59-	1.14x10 ¹²	0.25
	<i>P.debaryanum</i>	11866.82	1154.76-	6.04x10 ⁶	0.26
<i>Solanum nigrum</i>	<i>R.solani</i>	637.16	215.86-	3608.74	0.43
	<i>S.rolfsii</i>	6643.3	709.43-	2.48x10 ⁶	0.25
	<i>M.phaseolina</i>	4.8x10 ⁸	88903.45-	1.0x10 ³⁸	0.14
	<i>P.debaryanum</i>	16291.3	2029.89-	8.74x10 ⁶	0.36
<i>Piper nigrum</i>	<i>R.solani</i>	295.9	99.46-	1521.5	0.38
	<i>S.rolfsii</i>	2119.18	311.97-	1.95x10 ⁵	0.25
	<i>M.phaseolina</i>	2310.94	687.09-	22159.28	0.48
	<i>P.debaryanum</i>	18768.27	1921.65-	3.43x10 ⁷	0.32
Solvent Plant extract <i>Ammi visnaga</i>	<i>R.solani</i>	98.45	50.14-	221.06	0.59
	<i>S.rolfsii</i>	25.32	14.68-	43.14	0.8
	<i>M.phaseolina</i>	98.45	50.15-	221.06	0.59
	<i>P.debaryanum</i>	842.99	299.79-	5083.36	0.48
<i>Schinus terbenlthiofolius</i>	<i>R.solani</i>	1309.84	362.04-	17394.71	0.4
	<i>S.rolfsii</i>	844.92	292.9-	6554.58	0.44
	<i>M.phaseolina</i>	1309.84	362.04-	17394.71	0.4
	<i>P.debaryanum</i>	6696.96	1108.16-	5.96x10 ⁵	0.36
<i>Solanum nigrum</i>	<i>R.solani</i>	32.6	16.2-	65.84	0.58
	<i>S.rolfsii</i>	9.55	6.02-	14.71	1.09
	<i>M.phaseolina</i>	32.6	16.2-	85.84	0.58
	<i>P.debaryanum</i>	70.05	32.02-	175.96	0.49
<i>Piper nigrum</i>	<i>R.solani</i>	154.35	87.14-	307.0	0.75
	<i>S.rolfsii</i>	111.08	344.32-	10231.26	0.43
	<i>M.phaseolina</i>	154.35	87.14-	307.0	0.75
	<i>P.debaryanum</i>	424.93	232.45	959.24	0.80

Table (3) : Effect of antagonistic fungi on the linear growth of *R. solani*, *S. rolfsii*, *M. phaseolina* and *P. debaryanum* on PDA medium *in vitro* (four days old)

Antagonistic fungi \ Pathogenic fungi	Inhibition % (I%)		
	<i>T. harzianum</i>	<i>T. viride</i>	<i>G. deliquescens</i>
<i>R. solani</i>	61.11	63.88	51.43
<i>S. rolfsii</i>	66.67	38.88	55.55
<i>M. phaseolina</i>	66.67	63.88	52.77
<i>P. debaryanum</i>	43.24	56.75	45.94

L.S.D for	Antagonistic fungi	Pathogenic fungi	Interaction
at 5%,	1.858	1.825	3.708
at 1%	2.555	2.765	5.109

4-Joint toxic effect of the tested compounds against the tested pathogenic fungi under laboratory conditions:

Carboxin / thiram fungicide was the most fungitoxic compound against pathogenic fungi and the least fungitoxic compound against antagonistic fungi. Carboxin / thiram was combined with the other tested compounds. Table (4) showed the obtained results from this test. .

4-1 Joint toxic effects against *R. solani*:

Data presented in Table (4) showed the joint action effects of compounds in pairs on the tested fungus. The values of Co-toxicity factor indicated that synergistic effects were obtained in the case of carboxin/thiram added to *Ammi visnaga*; *Piper nigrum* and *Schinus terbenhiofolius* as aqueous plant extracts and *Piper nigrum* as solvent plant extract. The most synergistic effect was obtained in the case of carboxin / thiram + the solvent extract of *Piper nigrum*. Additive effects were observed for the combinations of carboxin/thiram added to *Ammi visnaga*, *Schinus terbenhiofolius* and *Solanum nigrum* as solvent plant extracts. Meanwhile antagonistic effects were observed for the combinations of carboxin/thiram aqueous extract of *Solanum nigrum*.

4-2 Joint toxic effects against *S.rolfsii*:

Results in Table (4) showed the joint toxic effects of compounds in pairs on the tested fungus *S.rolfsii*. The values of Co-toxicity factors indicated that the combination of carboxin/thiram when mixed with *Piper nigrum*, *Schinus terbenthiofolius* and *Solanum nigrum* as aqueous plant extracts and solvent extract of *Piper nigrum* gave synergistic effects. Additive effects were obtained in the case of carboxin/thiram when mixed with solvent extract of *Schinus terbenthiofolius*. Carboxin / thiram when mixed with *Ammi vixnaga* (aqueous and solvent extracts) and *Solanum nigrum* (solvent extract) gave antagonistic effects.

Table (4):Percentage of inhibition (I%) and Co- Toxicity Factor (C.T.F) of carboxin/thiram combined with the tested compounds against pathogenic fungi (IC₂₅ for each).

Treatments	<i>R.solani</i>		<i>S.rolfsii</i>		<i>M.phoseolina</i>		<i>P.debarynum</i>	
	I%	C.T.F	I%	C.T.F	I%	C.T.F.	I%	C.T.F
Aqueous plant-extracts								
- <i>Ammi visaga</i>	61.88	+23.76	37.55	-24.90	83.70	+67.40	32.47	-35.60
- <i>Piper nigrum</i>	71.60	+43.20	79.81	+59.62	76.29	+52.58	50.02	+0.04
- <i>Schinus terebenthiofolius</i>	66.55	+33.1	65.23	+30.50	17.03	-65.94	21.31	-57.38
- <i>Solanum nigrum</i>	17.53	-64.94	76.05	+52.10	43.70	-12.60	30.35	-39.30
Solvent plant – extracts								
- <i>Ammi visnaga</i>	51.76	+3.52	16.43	-67.14	43.96	-12.08	66.50	+33.00
- <i>Piper nigrum</i>	84.05	+68.10	74.64	+49.28	44.44	-11.12	77.67	+55.34
- <i>Schinus terebenthiofolius</i>	47.48	-5.04	41.31	-17.38	27.40	-45.20	33.54	-32.92
- <i>Solanum nigrum</i>	41.26	-17.48	4.69	-90.62	13.70	-72.60	63.31	+26.62

4-3 Joint toxic effects against *M. phaseolina*

Data in Table (4) showed that mixtures of carboxin/thiram + aqueous extract of *Ammi visnaga* or aqueous extract of *Piper nigrum* gave synergistic effects. Antagonistic effects were observed in the case of carboxin/thiram added to *Schinus terbenthiofolius* (aqueous and solvent extracts) and solvent extract of *Solanum nigrum*. Carboxin/thiram when mixed with aqueous extract of *Solanum nigrum* and solvent extracts of *Ammi visnaga* and *piper nigrum* gave additive

effects. Zein *et al.*, (1990), Ahmed and Ali (1990) , Mohamed and Abo-Raya (1993b) and Abdel-Aziz *et al.*, (1996).

4-4 Joint toxic effects against *P.debaryanum*

The joint toxic effect of carboxin/thiram and tested compound mixtures were evaluated against *P.debaryanum*. Data in Table (4) showed the toxic effects of compounds pairs on the tested fungus. The values of co-toxicity factors indicated that synergistic effects were observed for the combinations of carboxin/thiram when mixed with solvent extracts of *Ammi visnaga*, *Piper nigrum* and *Solanum nigrum*. On the other hand additive effects were observed in the case of carboxin/ thiram added to aqueous extract of *Piper nigrum*. Antagonistic effects were observed for the combinations of carboxin / thiram with aqueous extracts of *Ammi visnaga*, *Schinus terbenthiofolius* and *Solanum nigrum* and *Schinus terbenthiofolius* (solvent extract). Many studies were carried out to evaluate the joint effect of different compounds against *pythium sp.* Ahmed and Ali (1990).

4-5: Effect of carboxin/thiram fungicide on the efficacy of biological control *in vitro*:

Data in Table (5) showed that carboxin/thiram increased the biological control efficacy against all the tested fungi, except, *M.phaseolina* in which a decrease in the biological control efficacy was observed as compared with data in Table (3). *T.harzianum* combined with carboxin/thiram, inhibited growth of *R.solani* more than *T.viride* and *G.deliquescens* in PDA medium. In the case of *S.rolfsii*, *T.harzianum* was effective, hence it gave more inhibition, followed by *G.deliquescens* and *T. viride*. Carboxin/thiram reduced the efficacy of biological control in the case of *M.phaseolina* , except for *G.deliquescens*; carboxin / thiram increased the efficacy of biological control of *M.phaseolina*. *T.harzianum*, *T.viride* and *G.deliquescens* reduced the radial growth of *M.phaseolina*, when the PDA medium was mixed with carboxin/thiram fungicide. In the case of *P.debaryanum* added to PDA medium which included carboxin/thiram an increase in efficacy of biological control was observed. *T.harzianum* was the most effective in increasing the biological control followed by *T.viride* and *G.deliquescens*. *T. harzianum* was the most effective biocontrol agent against the pathogenic fungi. These

results are in full agreement with that found by Khalifa (1987), Kaur and Mukhopadhy (1992) and Ali and Pathak (1997). They found that antagonistic fungi combined with fungicides effectively controlled phytopathogenic fungi caused different plant disease.

Table (5) : Effect of biocontrol agent in PDA medium combined with carboxin/thiram on pathogenic fungi *in vitro* (four days old).

Antagonistic fungi / Pathogenic fungi	Inhibition % (1%)		
	<i>T.harzianum</i>	<i>T.viride</i>	<i>G.deliquescens</i>
<i>Rhizoctonia Solani</i>	69.23	63.50	58.82
<i>Sclerotium rofsii</i>	74.28	48.57	68.57
<i>Macrophomia phaseolina</i>	58.82	60.00	57.50
<i>Pythium debaryanum</i>	60.00	57.14	51.45

REFRANCES

- Abdel – Aziz, M.A.; S.M.M. Mahmoud and A.A. Ismail (1996).** Impact of imidacloprid insecticide on efficacy of some fungicides in controlling damping-off and root – rot diseases of cotton seedlings. J. Agric. Res. Tanta Univ., 22 (2) : 243-255.
- Abbott, W.S. (1925).** A method for computation of the effectiveness of insecticide. J. Econ. Entomol; 18:265-267.
- Ahmed, J.M. and H.H.Ali (1990)** Chemical control of cotton damping-off in Ninevah province, Iraq. Arab. j. plant protection. 8(1) : 6-11.
- Ali, M.S. and A.K. Pathak (1997).** Hinosan (0.05%) : an ecofriendly fungicides for managing rice sheath blight. International Rice Res. Notes. 22(3) : 32-33.
- Beye, F. (1978).** Insecticides from the vegetable Kingdom Pl. Res. Dev., 7:13-31.
- Carcia, R.P. and M.V.P. Lawas (1990).** Note : Potential plant extracts for the control of Azolla fungal pathogens. Philippine Agric., 73(3-4): 343-348.
- Dubey, P.S. and L.P.Mall (1972)** . Herbicidal pollutant, Pollen damage by herbicide vapour s. Sc. Cult., 39:556-558

- Elad, Y.; I. Chet; P. Boyle and Y. Henis (1983).** Parasitism of trichoderma spp. On *Rhizoctonia solani* and *Sclerotium rolfsii*: Scanning electron microscopy and fluorescence microscopy. *Phytopathology*, 73, 85-88.
- Elad, Y.; I. Chet and Y. Henis (1982).** Degradation of plant pathogenic fungi by *Trichoderma harzianum*. *Can. J. Microbiol.*, 28: 719-725.
- El-Shoraky, Fathia, S.A. (1998)** Using extracts and oils of some plants in controlling plant diseases. Ph. D. Thesis, Fac. Agric. Tanta University.
- Epstein, S.S.; Andrae; H. Jaffec; S. Joshi; H. Folk and N. Natnel (1967).** Carcinogenicity of the herbicide melecic hydrazide. *Nature*, 215: 1388-1390.
- Fawcett, C.H. and D.M. Spencer (1970).** Plant chemotherapy with natural products. *A. Rev. Phytopatho.*, 8: 403-418.
- Finney, D.J. (1952).** Probit analysis. Cambridge Univ. Press 318p.
- Howell, C.R. and R.D. Stipanovic (1995).** Mechanisms in the biocontrol of *Rhizoctonia solani* induced cotton seedling disease by *Gliocladium virens*. *Antibiosis. Phytopathology*. 85 (4): 469-472.
- Javoraska, T. (1978).** Effect of combined herbicides on the occurrence of morphoses in the spikes of spring barley. *Agrochemia*, 18: 37-42.
- Kaur, N.P. and A.N. Mukhopadhyay (1992).** Integrated control of chickpea wilt complex by Trichoderma and chemical methods in India. *Tropical Pest Management*. 38(4) : 372-375. (c.f. CAB Abstracts 1993-1994).
- Khalifa, E.Z. (1987).** Further studies on some soil-borne fungi affecting soybean and their control. Ph.D Thesis, Fac. of Agric., Minufiya.
- Mansour, N.A.; M.E. El Defrawi; A. Topozada and M. Zeid (1966)** : Toxicological studies on the Egyptian cotton leaf worm *Spodoptera littoralis* (Boisd). VI. Potentiation and antagonism of organophosphorus and carbamate insecticides. *J. Econ. Entomol.* 49, 307-311.

- Mohamed, S.A. and Abo-Raya (1993 a).** Biological control of tomato damping-off. *Menofiya.J. Agric. Res.*, 18(4) : 2131-2140.
- Mohamed, S.A. and Abo-Raya (1993 b).** Integrated control of tomato damping-off. *Menofiya.J. Agric. Res.*, 18(4) : 2151-2162.
- Omar, S.A.M.; M.M.H. Rahhal; A.A.Hilal and A.Zayed (1993).** Influence of intercropping of broad bean with some medicinal plants on disease severity of chocolate spot and rust and seed yield. 4th conf. Agric Dev. Res. Ain-Shams Univ., Cairo, Feb. 13-18.
- Pandey, V.N and N.K. Dubey (1997).** Antifungal potentiality of some higher plants against *Pythium* species causing damping-off in tomato. *National Academy Science Letters*. 20(5-6) : 68-70.
- Poswal, M.A.T. ; G.Masunga; I.Javaid and B.C. Kwerepe (1993).** Potential of different toxic and medicinal plant extracts for the control of fungal plant pathogens in Botswana. *Mededelingen - van - de - Faculteit -Land-bowetenschappen, Universitait. Gent.*, 58 (36) : 1373-1381.(c.f. CAB Abstracts 1993-1994).
- Shalaby, M.S.; A.Z.Aly and A.E.A. Ismail (1997).** Chemical and biological control of some soil-borne fungi under intercropping conditions of maize and soybean. *Egypt, j.Agric. Res.*, 75(2) : 303-320.
- Snedecor, J.W. (1965).** *Statistical methods* . Ames, Iowa, U.S.A. Iowa state univ; press.
- Tasleem-uz-Zaman-khan;M.A.Nasir and S.A.A. Bokhari (1998).** Antifungal Properties of some plant extracts . *Pakistan. J. phytopathology*. 10(2) : 62-65.
- Topps, J.H. and R.L. Weim (1957).** Investigation on fungicides. III. The fungitoxicity of 3-and 5-alkyl salicylanilide and P-chloronilines. *Ann. Appl. Biol.* 45(3) : 506-511.
- Upadhyay, J.P. and A.N. Mukhopadhyay (1986).** Biological control of *Sclerotium rolfsii* by *Trichoderma harzianum* in sugar beet. *Trop pest Mgmt.*, 32 :215-220.

- Warr, S.J. ; K. Thompson and M.Kent (1992). Antifungal Activity in seed coat extracts of woodland plants . *Oecologia*. 92(2) : 296-298.
- Wilson, Ch.; J.D. Fanklin and B.E. Otto (1987). Fruit volatiles inhibitory to *Monilinia fruticola* and *Botrytis cinerea* Plant Dis., 71(4) : 316-319.
- Zein, A.A.; M.A.Ashry, A.E.El-Sherbeni and A.A. Ismail (1990). Fungicidal toxicity and joint fungitoxic action of some pesticides . *J. Agric. Res., Tanta Univ.*, 16(4) : 809-817

الملخص العربي

التأثير البيولوجي السام لبعض المستخلصات النباتية و الفطريات المضادة على بعض فطريات التربة

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تهدف هذه الدراسة إلى تقدير سمية بعض المستخلصات النباتية ، بالإضافة إلى دراسة تأثير بعض الفطريات المضادة ضد الفطريات الممرضة، وإشتملت الدراسة أيضا على تقييم التأثير المشترك لخلات هذه المعاملات بهدف الحصول على مخاليط لها خواص إبادية قوية على بعض الفطريات المسببة لمرض عفن الجذور وموت البادرات و أوضحت نتائج تقييم المستخلصات المائية على الفطريات الممرضة أن المستخلص المائي للفلفل عريض الاوراق كان أكثر تثبيطا للفطر ريزوكتونيا سولاني يليه في التأثير المستخلص المائي لعنب الديب ، بينما المستخلص المائي للخللة أكثر فعالية ضد الفطريات سكليروشيم رولفزياي ، ماكروفوميتا فاسولينا يليه المستخلص المائي للفلفل عريض الاوراق .المستخلصات المائية لعنب الديب ، الفلفل عريض الاوراق كان أكثر تثبيطا للفطر المضاد تريكودرما هارزيانم يليها المستخلص المائي للفلفل الاسود ، بينما المستخلص المائي للخللة أقل تأثيرا على نفس الفطر ، المستخلصات المائية لعنب الديب والفلفل الاسود كانت أكثر تأثيرا على الفطر المضاد تريكودرما فيردي ، بينما المستخلص المائي للفلفل الاسود كان أكثر فعالية ضد الفطر جيلوكلاديم ديليكويسنس يليه

المستخلص المائي لعنب الديب .المستخلص المذيبي للفلفل الاسود كان أكثر سمية لتثبيط النمو الميسليومي للفطريات ريزوكتونيا سولاني ، سكليروشيم رولفزياي ، ماكروفومينا فاسولينا يليه المستخلص المذيبي للخلة ، أيضا كان المستخلص المذيبي للفلفل الاسود أكثر تأثيرا على الفطر بيثيم ديباريانم يليه المستخلص المذيبي لعنب الديب .المستخلص المذيبي للفلفل الاسود كان أكثر فعالية ضد الفطريات المضادة تريكودرما هارزيايم ، تريكودرما فيردي يليه المستخلص المذيبي للخلة ، بينما المستخلص المذيبي لعنب الديب كان أكثر سمية للفطر المضاد جليوكلاديم ديليكويسنس ، يليه المستخلص المذيبي للفلفل عريض الاوراق. أوضحت نتائج المكافحة البيولوجية أن الفطر المضاد تريكودرما هارزيايم كان أكثر تضادا للفطرين ماكروفومينا فاسولينا، سكليروشيم رولفزياي، بينما الفطر تريكودرما فيردي كان أكثر تضادا للفطرين ريزوكتونيا سولاني ، بيثيم ديباريانم. وعند دراسة التأثير المشترك لمخاليط المبيد الفطري كاربوكسين / ثيرام مع المركبات الاخرى نجد أن هناك تأثيرا تشظييا في الحالات الآتية: في حالة الفطر ريزوكتونيا سولاني : عند خلط المبيد الفطري كاربوكسين ثيرام مع والمستخلصات المائية للخلة ، الفلفل الاسود ، الفلفل عريض الاوراق ، والمستخلص المذيبي للفلفل الاسود. وكان أكثر المخاليط تشظيا هو مخلوط كاربوكسين / ثيرام + المستخلص المذيبي للفلفل الاسود. في حالة الفطر سكليروشيم رولفزياي : عند خلط المبيد الفطري كاربوكسين / ثيرام مع المستخلصات المائية للفلفل الاسود ، الفلفل عريض الاوراق ، عنب الديب، والمستخلص المذيبي للفلفل الاسود . كان أكثر المخاليط تشظيا هو كاربوكسين / ثيرام + المستخلص المائي للفلفل الاسود .في حالة الفطر ماكروفومينا فاسولينا : عند خلط المبيد الفطري كاربوكسين / ثيرام مع المستخلصات المائية للفلفل الاسود. في حالة الفطر بيثيم ديباريانم : عند خلط المبيد الفطري كاربوكسين/ثيرام مع المستخلصات المذيبة للخلة ، الفلفل الاسود، عنب الديب، وكان أكثر المخاليط تشظيا هو كاربوكسين/ثيرام + المستخلص المذيبي للفلفل الاسود . عند خلط المبيد الفطري كاربوكسين / ثيرام بالبيئة زادت كفاءة المكافحة البيولوجية للفطريات ريزوكتونيا سولاني، سكليروشيم رولفزياي، بيثيم ديباريانم وقلت في حالة الفطر ماكروفومينا فاسولينا ، والفطر تريكودرما هارزيايم كان أكثر تضادا للفطريات ريزوكتونيا سولاني، سكليروشيم رولفزياي ، بيثيم ديباريانم بينما الفطر تريكودرما فيردي كان أكثر تضادا للفطر ماكروفومينا فاسولينا .