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CONTROL OF THE ROOT ROT DISEASES OF WHEAT USING FUNGICIDES ALONE AND IN COMBINATIONS WITH SELENIUM

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ABSTRACT

Five fungicides (tebuconazole, fludioxonil, iprodione, azoxystrobin and flutolanil) were evaluated separately or in mixtures with the antioxidant (selenium) against Fusarium graminearum and Rhizoctonia solani which causing root rot diseases of wheat. Depend upon EC_{50} values of the tested fungicides, the in vitro studies showed that the descending order of fungicidal activity against F.graminearum growth was as follows;fludioxonil> tebuconazole > iprodione > azoxystrobin > flutolanil, while it was flutolanil >fludioxonil> tebuconazole > iprodione > azoxystrobin against R. solani growth.The two fungi tolerated high concentration of selenium. Interestingly, the fungitoxic activities of the tested fungicides were greatly increased by adding selenium to the fungicide- amended medium. In greenhouse trials, the fungicides were applied at rates of 1.5 and 3.0 g /kg seeds, while selenium was applied at 10 and 20 µg / ml (as seed treatments) alone or in combinations with fungicides for controlling root rot diseases. The results showed that all fungicides treatments significantly reduced diseases incidence and increased emergence and plant stands. Generally the addition of the antioxidant (Se) at 20µg / ml to the higher rate (3.0 g/kg seeds) of each fungicide caused significant reduction of the pre- and post- emergence damping off and subsequently increased plant survivals. However, the addition of Se to the low rate of each fungicide gave satisfactory fungicidal action as well as minimizing the environmental pollution.

Keywords: Fungicides, Selenium, Wheat, Root rot, *Fusarium graminearum* and *Rhizoctonia solani*

INTRODUCTION

Wheat (*Triticum aestivum* L.), is one of the most important cereal crops in Egypt, either as a staple food grain for human or as a major source of straw fodder for animal feeding. This crop is subjected to be attacked by many diseases. Root rot diseases caused by soil-borne fungi are the most important wheat diseases. Common root rot of wheat is among the most important diseases attacking wheat in Egypt (**El-Kholy, 1999**). Several fungi were recorded as causal pathogens of root rot diseases such as *Fusarium graminearum*, *Rhizoctonia solani* and *Pyrenophora tritici – repentis* (**El-Kholy, 1999; and Asran & El-Eraky-Amal, 2011**). **Nourozian *et al*. (2006)** reported that both *F. graminearum* and *F. culmorum* fungi cause root rot, foot rot, crown rot, stem rot and head blight in wheat. **Smiley & Patterson (1996)** found that *F. graminearum* capable of killing wheat seedlings in the greenhouses. Also, *R. solani* is a ubiquitous soil- borne plant pathogenic fungus which causes significant yield losses in many agriculturally important crops (**Paulitz *et al*., 2006**). **Moubarak & Abdel-Monaim (2011)** found that *R. solani* cause damping off and root rot diseases on wheat crop in Egypt.

Fungicide seed treatment is a simple and inexpensive method for reducing seedling blight and is widely used in many crops. Several fungicides have been used by many investigators to control root rot diseases caused by *F. graminearum* and *R. solani*. For example, tebuconazole was evaluated by **El-Kholy (1999)**, **Hershman (2011)**, **Ivic *et al*. (2011)** and **Shomeet *et al*. (2018)**. The fungicidal action of fludioxonil was evaluated by **Hwang *et al*. (2006)**, **Xue *et al*. (2007)** and **Hamada *et al*. (2011)** who reported that this compound has a wide range of activity against fungal pathogens belonging to different fungal classes. Iprodione was evaluated by **El-Kholy (1999)**, **Hamada *et al*. (2011),** and **El-Ballat (2017)**. The fungicidal activity of azoxystrobin was studied by **Sundravadana *et al*. (2007)**, **Swett (2007)** and **Srivastava (2014)**. On the other hand, flutolanil was among fungicides evaluated against *F. oxysporum* f. sp. *lycopersici*, *F. solani* and *R. solani* in tomato under laboratory and greenhouse conditions (**El-Samadisy *et al*., 2008; Ali, 2008 and Haggag-Karima & El-Gamal-Nadia, 2012**).

Antioxidants including Selenium (Se) which save to human and environment had been used successfully to control some plant diseases such as faba bean chocolate spot (**Hassan *et al*., 2006**), root rot and leaf blight in lupine (**Abdel-Monaim, 2008**), damping-0ff in pepper (**Rajkumar *et al*., 2008**) and root rot and wilt in pepper (**Abdel-Monaim & Ismail, 2010**).

It was suggested that the addition of certain antioxidants to the fungicides increased their fungitoxic activity. Se improves the activity of tetraconazole, penconazole, fenarimol, trifloxystrobin and flutolanil against *F. oxysporum* and *R. solani* (**El-Khawaga-Maii, 2006**), difenoconazole, azoxystrobin, trifloxystrobin and metalaxyl against *Alternaria solani* (**Sameer, 2013**). **Khalifa & Sameer (2014)** found that Se enhanced the fungitoxic activity of iprodione, myclobutanil, prochloraz, tetraconazole and trifloxystrobin against *Penicillium digitatum*. Also, **Shomeet *et al*., (2018)** found that Se enhanced the fungitoxic activity of tebuconazole against *Alternaria alternata*, *F. graminearum* and *Pyrenophora tritici – repentis*.

Based on these studies, laboratory and greenhouse experiments were conducted in the present investigation to evaluate the fungicidal activities of five fungicides belonging to different fungicidal groups used alone or in combinations with the antioxidant (Se) against *F. graminearum* and *R. solani* on wheat.

MATERIALS AND METHODS

Laboratory and greenhouse experiments were conducted at the Department of Plant Protection, Faculty of Agriculture, Al-Azhar University, Nasr City, Cairo, Egypt.

Experimental materials

The wheat (*Triticum aestivum* L.) seeds, (Gemiza 9 cv.) were obtained from Wheat Research Department, Agriculture Research Center, Ministry of Agriculture and Land Reclamation.

Fungicides: The following fungicides were used, tebuconazole (Raxil, 2.5 % F.S., fludioxonil (Celest, 10% F.S.), iprodione (Rovral, 50% W.P), azoxystrobin (Amistar, 25% S.C.) and flutolanil (Moncut, 25 % W.P.).

Antioxidant: Selenium was used as sodium selenite (Na_2SeO_3 98 % W/W).

Isolation and identification of the causal organisms:

Pure isolates of *Fusarium graminearum* and *Rhizoctonia solani*, obtained from wheat roots showing typical symptoms of root rot disease. Transverse sections of the wheat roots collected from the field were surface sterilized by 0.5 % sodium hypochlorite for 1 min., rinsed in sterilized distilled water and drained on paper towels. They were transferred on PDA medium in Petri dishes, each contained 3 pieces and replicated 3 times. The dishes were incubated at 27^{o} C for 5 days. The growing fungi were subcultured on PDA slants. Single spore technique was used for purification of the isolated fungi.

Identification of the isolated fungi was carried out in Department of Botany, Branch of Plant Pathology, Faculty of Agriculture, Al-Azhar University, Nasr City, Cairo, Egypt.

In vitro fungitoxicity test:

A study was conducted to estimate the fungicidal activity of the tested fungicides and antioxidant. The fungicides were diluted in sterile distilled water, then added to cooled PDA medium at concentrations of 0.01, 0.05, 0,1, 0.5, 1.0, 5.0, 10.0, 25.0, 50.0, and 100.0 µg a.i./ml for each, whereas antioxidant was evaluated at concentrations of 10.0, 25.0, 50.0, 75.0, 100.0, 150.0, 200.0, 250.0, 300.0, 350.0, 400.0, 450.0 and 500.0 µg / ml. In other trials, PDA media amended with different fungicide concentrations were further amended with 10 and 20 µg / ml antioxidant. The poisoned media were poured in plates (9 cm diameter), other plates containing compounds – free medium were used as control treatment. The medium was seeded by 0.4 cm diameter disk, removed from 7 day-old culture of the fungus, on solidified medium and incubated for 7 days at 25^{o} C. Each treatment was replicated four times. Growth on the fungicides, antioxidant and their mixtures amended medium was determined by measuring the colony diameter (cm). The percentage of growth inhibition was calculated relative to the control treatment. The concentration giving 50 % linear growth inhibition (EC_{50}) was determined by regression analysis of the log probit transformed data (**Finney, 1971**).

Pot experiments:

Soil infestation:

To prepare inocula required for the test, the isolates of *F. graminearum* and *R. solani* were grown in 500 ml conical flask contained barley grains medium. The used soil (clay loamy), was air dried, sieved and packed in some polyethylene bags which were steamed in an autoclave until a temperature of 100^{o} C (1.3-1.4 pressure) was reached and then holding the temperature at $90\text{-}110^{o}$ C (1.1-1.4 pressure) for one hour (**Knudsen &Bin, 1990**). The inoculum of each fungus was incorporated into the autoclaved soil at the level of 5 % W/W and thoroughly mixed. Plastic pots (20 cm in diameter), which were previously sterilized by 5 % formalin solution, were filled with infested soil, except control without infestation (the same amount of the sterile barely grains without fungus). All pots were daily irrigated for 7 days to stimulate the fungal growth and to ensure its distribution within the soil before cultivation.

Fungicidal seed treatments:

This trial was carried out to investigate the efficiency of fungicides and Se alone or in combination (as seed treatments) for controlling the incidence of artificial infection with *Fusarium* and *Rhizoctonia* root rot on wheat growing in pots under greenhouse conditions. Seeds were treated with the tested fungicides (1.5 and 3.0 g product/kg seeds), Se (10 and 20 µg / ml) and their mixtures according to **Warhman *et al*. (1989)** with some modification. The appropriate amounts of the tested fungicides, Se and their mixtures were placed in plastic bags, seeds were added, then a few drops of 5 % animal glue solution were added and shaken by the hand until the seeds were uniformly covered by the chemical. Treated seeds were then dried at room temperature for 24 hrs. before planting.

The experiments were designed as follows:

1- Fungicide- free seeds in non-infested soil.
2- Fungicide- free seeds in soil infested with *F. graminearum* or *R. solani*.
3- Fungicide- treated seeds in soil infested with *F. graminearum* or *R. solani*.
4- Selenium-treated seeds in soil infested with *F. graminearum* or *R. solani*.
5- Fungicide + selenium-treated seeds in soil infested with *F. graminearum* or *R. solani*.

Each pot was planted with 15 wheat seeds and each treatment was replicated four times. Pre- and post- emergence damping –off % were recorded after 14 and 42 days from sowing, respectively.

RESULTS AND DISCUSSION

1- Sensitivity of the fungi to fungicides, selenium and their combinations:

The *in vitro* sensitivity of *Fusarium graminearum* and *Rhizoctonia solani* to five fungicides used alone or in combinations with Se were determined. The oncentration that inhibits 50 % of the fungal growth (EC_{50} values) of these treatments are listed in Table (1).

F. graminearum was highly sensitive to fludioxonil and tebuconazole (EC_{50} = 0.371 and 3.79 µg a.i. /ml, respectively), moderately sensitive to iprodione and azoxystrobin (EC_{50} = 8.83 and 17.38µg a.i. /ml, respectively). On the other hand, flutolanil was the least effective fungicide (EC_{50} = 96.86µg a.i. /ml).

The results also indicated that flutolanil, fludioxonil and tebuconazole were the most effective fungicides in inhibiting the mycelial growth of *R. solani* (EC_{50} values were 0.11, 0.29 and 0.541µg a.i. /ml, respectively). However, iprodione and azoxystrobin were the least effective ones (EC_{50} = 3.653 and 4.27 µg a.i. /ml, respectively).

Data listed in Table (1) showed high efficacy of tebuconazole against both *F. graminearum* and *R. solani* under laboratory condition. This result is in accordance with that obtained previously (**Hershman, 2011; Ivic *et al*., 2011 and Shomeet *et al*., 2018**). Also, fludioxonil fungicide exhibited a high effect against the growth of *F. graminearum* and *R. solani*. This result was confirmed by **Hwang *et al*. (2006), Fernandez *et al*. (2009) and Hamada *et al*. (2011)**. Fludioxonil provides excellent control seed borne *Fusarium* as well as several soil borne pathogens, with the exception of *Pythium* (**Hershman, 2011**). Iprodione fungicide exhibited a moderate effect against the growth of *F. graminearum* and high effect against *R. solani* (Table 1). This result was confirmed by **El-Ballat (2017)** who found that the EC_{50} values of iprodione to *F. graminearum* and *R. solani* were 23.0 and 1.8µg a.i. /ml, respectively. **Hamada *et al*. (2011)** showed that *R. cerealis* was sensitive to iprodione, difenoconazole and fludioxonil with mean EC_{50} values of 0.419, 0.062 and 0.033 µg a.i. /ml, respectively.

Azoxystrobin fungicide exhibited a moderate effect against the growth of *F. graminearum* and it was high effective against *R. solani* growth (Table 1). This result was confirmed by **Sundravadana *et al*. (2007), Baturo-Ciesniewska *et al*. (2011) and Srivastava (2014)**. Azoxystrobin is registered for control of *Fusarium* on more than 100 ornamental crops (**Swett, 2007**).

Same table showed that flutolanil was the most effective fungicide against *R. solani* growth, but it was the least effective one against *F. graminearum*. Similar results were previously obtained by **Ali (2008)** who found that the EC_{50} values of flutolanil to *F. oxysporum* f. sp. *lycopersici* and *R. solani* were 67.61 and 0.29 µg a.i. /ml, respectively. **Yamaguchi *et al*. (1998)** found that flutolanil did not inhibit *F. oxysporum* growth up to 1000ppm.

Tebuconazole is member of triazole fungicides or demethylation inhibitors (DMIs) that inhibit the biosynthesis of ergosterol, which is an important component of the fungal cell membrane by inhibiting C_{14}- demethylase (**Munkvold, 2009**). Phenylpyrrole fungicidal ingredient fludioxonil, is a broad-spectrum fungicide which is non-systemic with a long residual activity. The mode of action of fludioxonil is the inhibition of transport-associated phosphorylation of glucose as well as preventing glycerol synthesis (**Hwang *et al*., 2006**). Iprodione is a dicarboximide fungicide which inhibits DNA and RNA synthesis in the germinating fungal spore as well as inhibiting

Table (1): EC_{50} values (µg a.i. /ml) of the tested fungicides and selenium and their mixtures against mycelial growth of two fungi grown on PDA medium.

Compounds	*Fusarium graminearum*					*Rhizoctonia solani*				
	EC_{50}* of the compound (µg / ml)	EC_{50}* of the fungicide + Se (µg / ml)				EC_{50}* of the compound (µg / ml)	EC_{50}* of the fungicide + Se (µg / ml)			
		10 µg / ml		20 µg / ml			10 µg / ml		20 µg / ml	
Selenium	306.250	EC_{50}*	S.E.**	EC_{50}*	S.E.**	287.160	EC_{50}*	S.E.**	EC_{50}*	S.E.**
Tebuconazole	3.790	1.540	2.46	0.753	5.03	0.541	0.178	3.04	0.110	4.92
Fludioxonil	0.371	0.160	2.32	0.130	2.85	0.290	0.090	3.22	0.050	5.80
Iprodione	8.830	3.130	2.82	1.890	4.67	3.653	1.070	3.41	0.568	6.43
Azoxystrobin	17.380	8.130	2.14	3.060	5.68	4.270	2.220	1.92	1.140	3.75
Flutolanil	96.860	16.600	5.83	10.780	8.99	0.110	0.030	3.67	0.020	5.50

*EC_{50} = Effective concentration as (µg a.i. /ml) that give 50% inhibition of the fungal growth.

**S.E. = Synergistic Effect = EC_{50} of the fungicide alone / EC_{50} of the mixture.

the enzyme NADH cytochrome C reductase, thereby preventing lipid and membrane synthesis and ultimately mycelium growth (**Lee *et al*., 1998**). Azoxystrobin is member of strobilurins fungicides that inhibit the mitochondrial respiration at complex III (**Swett, 2007**). Flutolanil is a benzanilide fungicide which inhibit respiration at complex II (**Leroux, 1996**). The chemical structures of the tested fungicides (their functional groups), their mode of actions and the nature of the target fungus may be contributory factors affecting their fungicidal activities.

Data of Table (1) showed that *F. graminearum* and *R. solani* tolerated high concentration of Se according to the EC_{50} values (306.25 and 287.16µg a.i. /ml, respectively). **Sameer (2013)** indicated that Se had no effect on *Alternaria solani*. Also, **Khalifa & Sameer (2014)** found that *Penicillium digitatum* tolerated high concentrations of Se according to EC_{50} values (295.22µg a.i. /ml). Se did not affect the growth of *Pyrenophora- tritici repentis* (**Shomeet *et al*., 2018**).

Data presented in Table (1) showed the effect of tested antioxidant (Se) on the fungitoxic action of the tested fungicides against *F. graminearum* and *R. solani*. The results clearly indicated that the EC_{50} values of the mixtures of fungicide-Se were lower than that of the values of the corresponding fungicide alone which indicated that the addition of Se enhanced the potency of the fungicides. This enhancement effect differed according to the used concentration of Se and the type of the examined fungicide. For example, the efficiency of tebuconazole was markedly increased by 2.46 and 5.03 fold for *F. graminearum* and 3.04 and 4.92 fold for *R. solani* when Se was added at 10 and 20 µg /ml, respectively. Generally, with increasing the concentration of Se from 10 to 20µg /ml its potentiation effect was increased. These findings are in agreement with other previous studies (**Ali, 2008; Sameer, 2013; Khalifa & Sameer, 2014 and Shomeet *et al*., 2018**).

2- Greenhouse pot experiments.

2.1- Effect of fungicides and Se and their combinations on the disease caused by *F. graminearum*.

It is clear from the data presented in Table (2) that all fungicides tested as seed treatment, significantly reduced disease incidence and increased emergence and plant stands compared to the control treatment (seeds planted in infested soil). Decrease of pre-emergence damping- off with the treated seeds may be attributed to the effect of fungicides on the pathogen attacking the seeds causing seed decay. Data indicated also that the tested fungicides were effective in reducing post-emergence damping – off when compared with untreated seeds. It was noticed that increasing the rate of the tested fungicides resulted in enhancing their efficiencies against the pathogenic fungus with increasing the growing plants. Fludioxonil and tebuconazole were more effective in controlling pre- and post-emergence damping- off caused by *F. graminearum*, followed by iprodione and azoxystrobin, while flutolanil was the least effective one. Regarding the plant survivals, the difference between fludioxonil and tebuconazole was not significant. Also, there was a significant difference between tebuconazole and iprodione. The statistical analysis showed that the difference between iprodione and azoxystrobin was significant at the two level of p. Table (2) showed also that flutolanil fungicide, which was the least effective one, significantly differed from all tested fungicides. The statistical analysis also showed that the difference between the efficiency of the two applied rates was highly significant in all treatments.

The results obtained are in agreement with those obtained by many investigators. **Mullenborn *et al*. (2008)** found that *F. graminearum* was sensitive to tebuconazole. Triazole fungicides have been reported to be the most effective substances in controlling *Fusarium* head blight (**Paul *et al*., 2008 and Scarpino *et al*., 2015**). **Hershman (2011)**

Table (2): Effects of fungicides alone and in combination with selenium against *F. graminearum* on wheat growing in pots under greenhouse conditions.

Treatments	Rate of application g / kg seeds	Selenium								
		0.00			10 µg / ml			20 µg / ml		
		Average % of			Average % of			Average % of		
		Pre-emergence damping off	Post-emergence damping off	Plant survivals	Pre-emergence damping off	Post - emergence damping off	Plant survivals	Pre-emergence damping off	Post - emergence damping off	Plant survivals
0.00*		3.33*	3.45*	93.33*						
*F. graminearum***		31.67**	46.34**	36.67**	30.00	42.86	40.00	30.00	45.24	38.33
Tebuconazole	1.5	13.33	17.30	71.67	8.33	10.91	81.67	5.00	3.51	91.67
	3.0	8.33	7.27	85.00	5.00	5.26	90.00	3.33	0.00	96.67
Fludioxonil	1.5	13.33	13.46	75.00	6.67	8.93	85.00	5.00	1.75	93.33
	3.0	8.33	5.45	86.67	3.33	1.72	95.00	3.33	0.00	96.67
Iprodione	1.5	20.00	22.92	61.67	15.00	17.65	70.00	13.33	11.54	76.67
	3.0	13.33	13.46	75.00	11.67	11.32	78.33	8.33	9.09	83.33
Azoxystrobin	1.5	23.33	28.26	55.00	20.00	20.83	63.33	16.67	16.00	70.00
	3.0	18.33	20.41	65.00	15.00	15.69	71.67	11.67	13.21	76.67
Flutolanil	1.5	28.33	34.88	46.67	23.33	30.43	53.33	18.33	26.53	60.00
	3.0	25.00	31.11	51.67	20.00	29.17	56 67	15.00	17.65	70.00

	Pre- emergence damping off		Post- emergence damping off		Plant survivals	
L.S.D. at	1 %	5 %	1 %	5 %	1 %	5 %
Treatments (T)	4.29	3.25	4.87	3.69	5.41	4.09
Rates (R)	1.86	1.40	2.33	1.76	2.61	1.98
T x R	4.63	3.51	5.57	4.22	7.96	6.03

* Untreated seeds in non-infested (sterilized) soil

** Untreated seeds in soil infested with *F. graminearum*

found that fludioxonil had a strong effect on *Fusarium* sp. Iprodione, which significantly proved the plant survivals and reduced damping-off (Table 2), was tested by **El-Ballat (2017)** who found that it was effective in reducing the *Fusarium* root rot incidence on wheat plants under greenhouse conditions and increase emergence and plant stands. Azoxystrobin was among compounds tested by **Gullino *et al*. (2002)** who found that azoxystrobin was highly effective against *Fusarium* wilt of carnation. This compound controlled the disease when applied at transplant as soil drenching at the rate of 1-2 g/m^2.

Other studies have shown that some fungicide seed treatments were not effective against pathogenic *Fusarium* in *in vitro* tests, or that they varied in their toxicity and thus effectiveness against various *Fusarium* species. **Pirgozliev *et al*. (2002)** showed that applications of azoxystrobin reduced *Fusarium* head blight severity, while **Simpson *et al*. (2001)** noticed that treatment with azoxystrobin did not control *F. culmorum*. The effect of azoxystrobin on growth of *Fusarium* species was very low also *in vitro* test (**Mullenborn *et al*., 2008**). Flutolanil, which was the least effective in our results, was found to be one of the non active fungicides tested by **Yamaguchi *et al*. (1998)** against an isolate of *F. oxysporum* at a concentration of up to 1000 ppm.

Results in Table (2) also, showed that antioxidant (Se) alone did not control the incidence of pre- and post-emergence damping-off caused by *F. graminearum*. Interestingly, addition of Se to the fungicidal treatments greatly increased the efficiency of the fungicides against the incidence of the disease and increased the plant survivals. Moreover, the rate of the fungicide, required to give best control of the disease, was reduced when Se was added. It was found that tebuconazole, fludioxonil, iprodione, azoxystrobin and flutolanil at 3 g / kg seeds gave 85.0, 86.67, 75.0, 65.0 and 51.67 % plant survival,

Table (3): Effects of fungicides alone and in combination with selenium against *R. solani* on wheat growing in pots under greenhouse conditions.

Treatments	Rate of application g/kg seeds	Selenium								
		0.00			10 µg / ml			20 µg / ml		
		Average % of			Average % of			Average % of		
		Pre-emergence damping off	Post - emergence damping off	Plant survivals	Pre-emergence damping off	Post - emergence damping off	Plant survivals	Pre-emergence damping off	Post-emergence damping off	Plant survivals
0.00*		3.33*	3.45*	93.33*						
*R. solani***		38.33**	37.84**	38.33**	38.33	35.14	40.00	36.67	34.21	41.67
Tebuconazole	1.5	18.33	20.41	65.00	13.33	15.38	73.33	8.33	5.45	86.67
	3.0	11.67	9.43	80.00	6.67	5.36	88.33	5.00	3.51	91.67
Fludioxonil	1.5	15.00	13.73	73.33	10.00	9.26	81.67	6.67	5.36	88.33
	3.0	8.33	7.27	85.00	5.00	3.51	91.67	3.33	0.00	96.67
Iprodione	1.5	23.33	26.06	56.67	20.00	18.75	65.00	15.00	13.73	73.33
	3.0	15.00	13.73	73.33	11.67	11.32	78.33	10.00	5.55	85.00
Azoxystrobin	1.5	26.67	25.00	55.00	20.00	20.83	63.33	16.67	20.00	66.67
	3.0	16.67	14.00	71.67	13.33	9.62	78.33	10.00	7.40	83.33
Flutolanil	1.5	10.00	11.11	80.00	8.33	5.45	86.67	5.00	5.26	90.00
	3.0	6.67	5.36	88.33	1.67	0.00	98.33	0.00	0.00	100.0

	Pre- emergence damping off		Post- emergence damping off		Plant survivals	
L.S.D. at	1 %	5 %	1 %	5 %	1 %	5 %
Treatments (T)	3.16	2.37	3.86	2.89	4.26	3.19
Rates (R)	1.48	1.11	1.93	1.55	2.05	1.53
T x R	4.23	3.18	5.03	3.77	6.88	5.18

* Untreated seeds in non-infested(sterilized) soil

** Untreated seeds in soil infested with *F. graminearum*

respectively, when they were used alone. However, treatment with fungicide at 1.5 g / kg seeds plus 20 µg / ml of Se increased plant survivals to 91.67, 93.33, 76.67, 70.0 and 60.0 %, respectively. Regarding the plant survival, the effects of fungicides- Se mixtures significantly differed from that of fungicides applied separately. The statistical analysis also showed that the differences between the mixtures of fungicides with 10 and 20µg / ml of Se were significant. L.S.D. values indicated also that there was no significant difference between Se applied alone at 10 or 20 µg / ml and the control treatment. The statistical analysis also showed that the difference between the efficiency of the two applied rates was highly significant in all treatments.

2.2- **Effects of fungicides and Se and their combinations on the disease caused by *R. solani*.**

The effects of the tested fungicides (applied at 1.5 and 3 g /kg seeds) and Se (at 10 and 20 µg / ml) used alone and in combinations to control the incidence of pre- and post-emergence damping- off of wheat caused by *R. solani* are listed in Table (3). The incidence of pre- and post- emergence of untreated seeds were 38.33 and 37.84 %, respectively.

Results indicated that all fungicides tested as seed treatment were able to control damping-off caused by *R. solani*, with different degrees. Flutolanil was the most effective fungicide tested, followed by fludioxonil and tebuconazole, but iprodione and azoxystrobin were the least effective. The L.S.D. values for treatments in Table (3) revealed that flutolanil significantly raised the plant survival over all fungicidal treatments. The statistical analysis showed that the difference between fludioxonil and tebuconazole was significant. The two fungicides, tebuconazole and iprodione are significantly differed in their effect. The less effective fungicides, iprodione and azoxystrobin are not significantly differed from each other. In general, there was a significant difference between the activity of low and high rate of the tested fungicides. Such findings are in agreement with those previously reported. **Ali (2008)** reported that fungicides containing triazole, strobilurin and benzanilide fungicides are the most effective plant protection agents against *R. solani*. Tebuconazole provides protection from *R. solani* for a limited time following seeding (**Hershman, 2011**). Fludioxonil consistently reduced losses in seedling survival and establishment caused by *R. solani* and *F. avenaceum* (**Hwang *et al*., 2006**). Also, fludioxonil is a protectant fungicide used as a seed treatment on a range of crops for control of damping off and root rot caused by *Rhizoctonia* sp. (**Xue *et al*., 2007 and Hershman, 2011)**. The pre- and post-emergence damping – off and disease index of wheat plants caused by *F. graminearum* and *R. solani* were significantly reduced by the application of iprodione in a greenhouse study (**El-Ballat, 2017**). The strobilurin fungicides were introduced against a broad range of pathogens (**Bertelsen *et al*., 2001**). Azoxystrobin is an effective fungicide for the management of *Rhizoctonia* root and crown rot of sugarbeet caused by the fungus *R. solani* (**William *et al*., 2002**). **Sundravadana *et al*. (2007)** revealed that azoxystrobin at 1, 2 and 4 ppm completely inhibited mycelial growth of *R. solani*. Flutolanil, the most effective compound against damping off in our study, was effectively suppressed tomato damping off confirming the antirhizoctonial activety as carried out by **Kita *et al*. (2005)**.

Results in Table (3) also, showed that both concentrations of Se used alone did not control damping off on wheat plants. L.S.D. values indicated that there was no significant difference between Se applied alone at 10 or 20 µg / ml and the control treatment. Interestingly, fungicides- Se mixtures gave better control of the damping off incidence than the application of the fungicide alone. Moreover, the application rate of the fungicide, required to give best control of the disease, was reduced when Se was added. Results showed that the high rate of tebuconazole, fludioxonil, iprodione, azoxystrobin and flutolanil gave 80.0, 85.00, 73.33, 71.67 and 88.33 % plant survival, respectively, when they were used alone. However, 3 g / kg seeds plus 20 µg / ml of Se increased plant survivals to 91.67, 96.67, 85.0, 83.33 and 100.0 %, respectively. That means, the addition of the antioxidant (Se) at 20 µg / ml to the higher rate of each fungicide increased plant survivals about 1.13 – 1.16 fold. Regarding the plant survivals, the effect of fungicide- Se mixture significantly differed from that of fungicides applied separately. The statistical analysis also showed that the differences between the mixtures of fungicides with 10 µg / ml of Se and with 20 µg / ml of Se were significant, while the difference between the mixture of flutolanil at 3 g / kg seeds + 10 µg / ml of Se and the mixture of the same rate of this fungicide + 20 µg / ml of Se was no significant. The statistical analysis also showed that the difference between the efficiency of the two applied mixture rates was highly significant in all treatments.

Results in Tables (2 and 3) showed the antioxidant (Se) alone did not control the incidence of pre- and post-emergence damping off caused by *F. graminearum* or *R. solani*. Similar results were obtained by **Sameer (2013)** who found that Se slightly reduced the incidence of early blight on tomato plants. **Khalifa & Sameer (2014)** found that the usage of Se alone at 500, 1000 and 1500 µg / ml did not consistently control green mold disease on orange fruits comparing with the control treatment.

Results in Tables (2 and 3) also showed that the addition of antioxidant (Se) to the tested fungicides greatly increased their efficiencies against the root rot disease and reduced their rates required to give the best control rates. These effects were also reported by (**Simonetii *et al*., 2003; Ali, 2008; Sameer, 2013; Khalifa & Sameer, 2014 and Shomeet *et al*., 2018)**.

The mechanism of the potentiating effect of the antioxidant is not clearly explained. However, **Khan *et al*. (2001)** reported that antioxidant may make the fungal membrane more leaky and allowing more fungicide to enter into the fungal cells. **Simonetii *et al*. (2003)** stated that antioxidants appear to promote fungicide activity by increasing cell membrane permeability leading to the leakage of cellular enzymes. Also, **Baider & Cohen (2003)** reported that antioxidants may enhance host resistance to fungal infections. Another explanation is the antioxidants may play a part in reducing the oxidation of the fungicides in the fungal cells, which may reduce the fungitoxic action, thus increase the fungitoxicity **(Ali, 2008)**. Generally, this work suggests that the tested antioxidant could be used with low rates of the fungicides to increase their fungicidal action as well as to minimize environmental pollution.

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الملخص العربي

مكافحة أمراض عفن جذور القمح باستخدام مبيدات الفطريات منفردة و مخلوطة مع السيلينيوم

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تم تقييم خمسة مبيدات فطرية (تيبوكونازول وفلودايوكسونيل و ابروديون وازوكسيستروبين وفلوتولانيل) منفردة أو في مخاليط مع مانع الأكسدة (سيلينيوم) ضد فطريات فيوزاريوم جرامينيرم و ريزوكتونيا سولاني المسببين لامراض اعفان الجذور في القمح. بناء على قيم EC_{50} لمبيدات الفطريات أظهرت الدراسات المعملية ان كفاءة المبيدات على نمو فطر فيوزاريوم جرامينيرم كانت تتناقص على النحو الاتي: فلودايوكسونيل > تيبوكونازول > ابروديون > ازوكسيستروبين > فلوتولانيل،بينما كانت فلوتولانيل > فلودايوكسونيل > تيبوكونازول > ابروديون > ازوكسيستروبين ضد نمو فطر ريزوكتونيا سولاني. أظهر الفطران تحملا للتركيز العالي من السلينيوم. علاوة على ذلك فان كفاءة مبيدات الفطريات المختبرة ازدادت باضافة السلينيوم في البيئة المعاملة. بالنسبة لتجارب الصوبة فانه تم تطبيق مبيدات الفطريات بمعدلين ١,٥ ، ٣ جم / كجم بذرة اما السيلينيوم فطبق بتركيزين ١٠ ، ٢٠ ميكروجرام / مل كمعاملات بذور منفردين او في مخاليط لمكافحة اعفان الجذور. أوضحت النتائج ان كل معاملات مبيدات الفطريات سببت نقص ملحوظ في حدوث المرض مع زيادة في انبثاق واقامة النباتات. عموما كانت اكثر المبيدات كفاءة في مكافحة عفن الجذور المتسبب عن فطر فيوزاريوم جرامينيرم فلودايوكسونيل وتيبوكونازول يليهما مبيدي ابروديون وازوكسيستروبين بينما كان فلوتولانيل هو الاقل كفاءة مقارنة بالكنترول. كان أكثر المبيدات كفاءة في مكافحة المرض المتسبب عن فطرريزوكتونيا سولاني هو فلوتولانيل يليه فلودايوكسونيل وتيبوكونازول أما الاقل كفاءة فكانا مبيدي ابروديون و ازوكسيستروبين. لم ينجح السيلينيوم المستخدم منفردا في مكافحة اعفان الجذور المتسببة عن الفطرين. مخاليط مبيدات الفطريات مع السيلينيوم كانت أكثر كفاءة من المبيدات المستخدمة منفردة في مكافحة امراض أعفان الجذورالمتسببة عن كلا الفطرين. أوضحت النتائج ان المعدلات المنخفضة من مبيدات الفطريات عند استخدامها مخلوطة مع السيلينيوم كان لها نفس تأثير المعدلات المرتفعة بالاضافة الى تقليل تلوث البيئة بالمبيدات.