J. of Plant Production, Vol. 1 (1): 11 - 25, 2010

ALLEVIATING THE HARMFUL EFFECT OF SOME SOILBORNE FUNGI IN WHEAT PLANT GROWN UNDER INFECTED SOIL WITH *Fusarium graminearum* OR *Bipolaris sorokiniana*

Sakr , M. T. *; Z. A. Mohamed * ; M. I. El - Emery and M. S. Abo-El-Dahab ****

*** Agric. Bot. Dept., Agric. Fac., Mansoura Univ.**

**** Seed Technology Research Dep., Field Crops Institute, Agric.Res. Centre, Ministry of Agric.**

ABSTRACT

A pot experiments was carried out to investigate the role of some antioxidant materials (presoaking and foliar spray) in alleviation the harmful effect of biotic stress *Fusarium graminearum* or *Bipolaris sorokiniana* on wheat plant. Applied antioxidants decreased pre and post-emergence damping off, leaf infected percentage and leaf disease severity, fungi transmission from root, stem and grains of wheat plants growing under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana* in comparing with untreated plants growing in the same soil. However, grown infected soil enhanced survived seedlings percentage , emergence percentage of wheat plants with *Fusarium graminearum* or *Bipolaris sorokiniana* compared with untreated plants grown under same condition . SWE and Zn were more effective comparing with other antioxidants in both *Fusarium graminearum* or *Bipolaris sorokiniana*.

Applied antioxidants in high concentration were more effective than other concentrations in this respect.

Keywords: Antioxydants, pathogens, *Fusarium graminearum*, *Bipolaris sorokiniana,* wheat.

INTODUCTION

Several soilborne pathogens of wheat (*Triticum aestivum* L.; poaceae) cause common root rot, with necrosis of basal stems, crowns, subcrown internodes, and roots (Mathre, *et al.,* 2003). Fusarium head blight (FHB), caused by *Fusarium graminearum* , is a major disease of wheat and has resulted in heavy yield losses in many areas of the World (Parry, *et al.,*1995).

Bipolaris sorokiniana is a severe fungal pathogen causing common root rot and leaf spot diseases in wheat, barley, oats and rice, as well as in many other plants. The pathogen occurs throughout the world (Sivanesan, 1987). Plants with common root rot produce fewer tillers and fewer kernels per ear. Grain yield losses due to common root rot and seedling blight for Canada, Scotland (Murray, *et al.,* 1998).

Edgar, *et al.,* (2006), found that *Fusarium oxysporum* is a soilborne fungal pathogen that causes major economic losses by inducing necrosis and wilting symptoms in many crop plants. The interaction between *F. oxysporum* and the model plant *Arabidopsis thaliana* has been investigated to better understand the nature of host defences that are effective against the Fusarium wilt pathogen.

Exogenous salicylic acid treatment prior to inoculation, however, activated defence gene expression in leaves and provided increased *F. oxysporum* resistance as evidenced by reduced foliar necrosis and plant death. Exogenous salicylic acid treatment of the foliar tissue did not activate defence gene expression in the roots of plants. There results suggest that salicylate-dependent defences may function in foliar tissue to reduce the development of pathogen-induced wilting and necrosis.

According to Hammond-Kosack and Jones, (1996), microbes produce a number of cutinases and cell wall hydrolyzing enzymes, such as pectinases, cellulases, xylanases, and polygalacturonases (PGs), that attack the various cell wall polymers. Mechanical pressure may also facilitate microbial entry (Agrios, 1988). Although individually none of the above-mentioned enzymes is crucial for particular modes of pathogenesis (Knogge, 1996), these activities produce cell wall fragments, particularly oligomers of galacturonic acid, that might elicit additional defense responses or amplify the original ones (Levine, *et al.,* 1994).

The toxicity of AOS compounds may contribute to host cell death during the HR as well. Moreover Lipid peroxidation and generation of lipid free radicals after elicitor or pathogen exposure has been extensively documented (Adam, *et al*., 1989). Grant, and Loake, (2000), reported that one of the most rapid defense responses engaged following pathogen recognition is the so-called oxidative burst, which constitutes the production of reactive oxygen intermediates (ROIs), primarily superoxide (O_2) and H_2O_2, at the site of attempted invasion (Apostol, *et al*., 1989).

Severa1 other roles for SA and/or benzel adenine (BA) in plant defense have been proposed (Klessig and Malamy, 1994). Furthermore, exogenous SA application induces the coordinated expression of a subset of PR genes in numerous plant species (Ryals, *et al.,* 1996).

SA also inhibits ascorbate peroxidase (APX), the other key enzyme for scavenging H_2O_2. Furthermore, the ability of SA analogues to block APX activity correlated with their ability to induce defense-related genes in tobacco and enhance resistance to tobacco mosaic virus.

Glutathione, a major low-molecular weight thiol in plant cells, is increased after pathogen infection (Edwards, *et al.,* 1991). In carrot inhibition of glutathione synthesis, triggers phytoalexin accumulation, whereas the addition of H_2O_2 mimics this response (Guo, *et al.,* 1993).

Ascorbate is a substrate for cell wall peroxidases, it may play a role in the regulation of cell wall lignification, particularly during the HR, through its capacity to inhibit the oxidation of phenolic compounds by peroxidases (Mehlhorn, *et al.,* 1996). The pathogen-induced increase in the peroxidase activity of the cell wall would be effective only in the absence of AA.

Huckelhoven, *et al .,*(1999), found that H_2O_2 may play a substantial role in plant defense against the powdery mildew fungus. It did not detect any accumulation of salicylic acid in primary leaves after inoculation of the different barley genotypes, indicating that these defense responses neither relied on nor provoked salicylic acid accumulation in barley. The role of H_2O_2 and SA in the defense responses of plants against parasites is controversial. (Chen, *et al .,* 1993) has argued that SA acts *via* inhibition of a catalase that subsequently results in accumulation of H_2O_2 , which may involve cross-linking reactions leading to cell wall toughening (Brisson, *et al .,* 1994) and/or signaling that results in defense gene activation (Chamnongpol, *et al .,* 1998).

Shao, *et al.* (2008), reported that, antioxidants in plant cells mainly include glutathione, ascorbate, tocopherol, proline, betaine and others, which are also information-rich redox buffers and important redox signaling components that interact with cellular compartments. They added that,as an unfortunate consequence of aerobic life for higher plants, reactive oxygen species (ROS) are formed by partial reduction of molecular oxygen. The above enzymatic and non-enzymatic antioxidants in higher plant cells can protect their cells from oxidative damage by scavenging ROS. According to Chen, *et al.,* (2007), increasing the foliar GSH/GSSG ratio induced *Triticum aestivum* was enhanced resistance to powdery mildew and induced transcript accumulation of other pathogenesis-related genes.

Haffez, (2005), stated that antioxidants are substances that delay or inhibit oxidative to target molecules such as lipids, proteins, nucleic acid and carbohydrates. He added that antioxidants might protect a target by scavenging oxygen-derived species or minimizing the formation of oxygen-derived species.Various antioxidants ascorbic acid and its derivatives, glutathione, proline, trehalose, polyols, tocopherols, as well as pigments such as carotenoids and melanins—are present in fungal cells.

Mohammadi, and Kazemi, (2002), observed a significant increase in POX specific activity in heads of wheat cultivars following the inoculation with *F*. *graminearum* conidia.

MATERIALS AND METHODS

A pot experiments was carried out to investigate the role of some antioxidant materials (presoaking and foliar) in alleviation the harmful effect of biotic stress of *Fusarium graminearum* or *Bipolaris sorokiniana* on wheat plant. All pot were placed in a greenhouse under natural ambient conditions during winter season.

The sterilized seeds were soaked for 6 hours in any of applied antioxidant before sowing. The plants of each biotic stress of *Fusarium graminearum* or *Bipolaris sorokiniana* were sprayed with any of antioxidant used (the same antioxidant used in seed soaking) at three physiological stages (30, 60 and 90 days after sowing) using a hand atomizer.

Antioxidants used were: Ascorbic acid ,(ASA in,100 – 200 - 300 mg/L)., Salicylic acid (SA in, 100 – 200 - 300 mg/L)., Citric acid(100 – 200 - 300 mg/L)., Reduced Glutathione (GSH in,100 – 200 - 300 mg/L)., Seaweed extract (SWE in,1000 – 2000 - 3000 mg/L)., Humic acid (HA, in 1000 – 2000 - 3000 mg/L)., α –Tocopherol (50 – 100 – 150 L).,Putrescince(10 - 20- 30 mg/L)., Zinc sulphate (Zn,in 5 -10 - 15 mM)., Calcium chloride (Ca,in 200 - 300 - 400 mM)., Hydrogen peroxide (H2O2, in 400 - 600 - 800 mg/L)., Distilled water (control).

Seed sowing was carried out on November 20^{th} in pots (30 cm inner diameter) containing 10 Kg of air dried loamy soil at the rate of 15 grains/pot.

The pots were supplied with limit amounts of 15% P_2O_5 in the form of calcium super phosphate (with range of 130 Kg/fed), N 46% in the form of urea (in range of 75 Kg/fed) and K_2O in the form of potassium sulphate 48% . Thirty cm diameter of plastic pots were fill with soil and inoculated with 2% (w /w) of the inocula . The pots inoculated with *Fusarium graminearum* or *Bipolaris sorokiniana* watered and left for 3 days to ensure the distribution of inoculated fungi.

This experiment contained three factors, the first factor was infection fungi and included three treatments (*Fusarium graminearum* and *Bipolaris sorokiniana* and (control) without infection). The second factor was twelve antioxidant treatments in addition to the control .The third factor was antioxidants concentrations which were three concentrations for each treatment of antioxidants solutions except for distilled water treatment which was a control in the present study. Each treatment was carried out in three replicates.

Pre and post-emergence damping off ,survived seedlings , emergence percentage, severity disease and infected leaves percentage and fungi transmission were calculated.

Transmission of both pathogenic fungus from various plant parts (Root, lower first stem and middle stem node , upper stem node and grains) by planting them on sterile moist blotters and incubated for 7-10 days at 20 ± 2 ^oC . Fungi recorded from each treatment were identified and the transmission percentage was recorded according to the follow formula :
Mean of incidence % = Number of tissue infected / total number of tissue sections examined × 100 , (Abou-Table, 2006).

RESULTS

1- Pre-emergence damping off :

Data in the table (1) show the effect of applied antioxidants on pre-emergence damping off of wheat seedlings grown under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana.*

Data show that applied antioxidants decreased pre-emergence damping off of wheat seedlings growing in infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana* in comparing with untreated seedlings growing in infected soil. SWE and Zn were the most effective in decreasing pre-emergence damping off in both infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana*.

Higher concentration of ASA, GSH, SWE, HA, Toco, H_2O_2 while moderate concentration of SA, Zn were more effective in the case of *Fusarium graminearum* and *Bipolaris sorokiniana*.

2- Post-emergence damping off :

Data in the table (1) show the effect of applied antioxidants on post-emergence damping off of wheat seedlings growing under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana* fungi .

Data show that applied antioxidants slightely decreased post-emergence damping off of wheat seedlings grown under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana* when compared with untreated seedlings grown under same condition . SWE and Zn were mos effective in decreasing post-emergence damping off of wheat seedlings in both case of *Fusarium graminearum* or *Bipolaris sorokiniana*.

Applied antioxidants in high concentration were more effective than other concentrations in this respect.

Table (1): Effect of antioxidants concentrations on Pre and post- emergence damping off, Survived seedling (S.S) and emergence percentage (E.P) of wheat plant grown under infection soil by *Fusarium graminearum* or *Bipolaris sorokiniana*.

Treatments	Pre - emergence damping off		post- emergence damping off		Survived seedling percentage		Emergence percentage	
	F.g	B.s	F.g	B.s	F.g	B.s	F.g	B.s
Control	5.9	5.9	-	-	94.1	94.1	94.1	94.1
Inf. Soil + water	25.1	26.7	9.7	8.9	65.1	64.4	74.8	73.3
Inf. Soil + ASA	12.6	15.5	8.2	7.4	78.5	77.0	86.7	84.5
Inf. Soil + SA	14.8	15.5	7.4	6.7	77.7	77.8	85.2	84.5
Inf. Soil + Citric	14.1	19.3	8.2	8.9	76.2	71.8	84.4	80.7
Inf. Soil + GSH	17.7	15.5	7.4	11.1	74.5	73.3	81.9	84.4
Inf. Soil + SWE	8.2	8.2	5.2	6.7	86.6	85.1	91.8	91.8
Inf. Soil + HA	12.6	15.5	6.7	7.4	80.7	77.0	87.4	84.5
Inf. Soil + Put	15.5	13.3	7.4	11.8	77.8	74.0	85.2	85.9
Inf. Soil + Toco	18.5	15.5	9.6	10.4	71.8	73.3	81.5	83.7
Inf. Soil + Zn so_4	8.9	8.2	6.7	6.7	84.4	85.1	91.1	91.8
Inf. Soil + $CaCl_2$	14.0	16.3	6.7	8.9	78.5	74.0	85.1	82.9
Inf. Soil + H2O2	18.5	17.0	8.2	10.4	73.3	71.8	81.4	82.2
Mean	14.3	14.8	7.0	8.1	78.4	76.8	85.4	84.9
LSD at 5%	3.41	3.49	1.0	1.05	2.33	2.30	3.79	3.60

ASA : Ascorbic acid **Toco : α- Tocopherol** **SWE : seaweeds extract**
SA : Salicylic acid **Zn so_4 : Zinc sulfite** **HA : Humic acid**
GSH : Glutathione **$CaCl_2$: Calcium chloride** **H_2O_2 : Hydrogen peroxide**
Citric : Citri acid **F.g : *Fusarium graminearum*** **Inf. Soil : Infected soil with Conc.**
Put : Putrescine **B.s : *Bipolaris sorokiniana*.**

3- Survived seedlings :

Data in the table (1) show the effect of applied antioxidants on survived seedlings percentage of wheat plants grown under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana.*

Applied antioxidants enhanced survived seedlings percentage of wheat plants grown under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana.* compared with untreated plants grown under same condition . SWE , HA , and Zn were most effective in this respect.

Higher concentration of SWE , ASA , HA , Put , Toco and H_2O_2 while the moderate concentration of SA , Zn and Ca were mor effective than other concentrations in the case of *Fusarium graminearum*.

Moreover higher concentrations of ASA , Citric , GSH , Put , Toco , Ca , H_2O_2 while moderate concentrations of SA and Zn were more effective in *Bipolaris sorokiniana* case.

4- Emergence seedlings percentage :

Data in the table (1) show that applied antioxidants slightely increased emergence percentage of wheat seedlings grown under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana* compared with untreated seedlings grown under same condition .

SWE and Zn were most effective in this respect. In most cases the higher concentration of applied antioxidants were more effective comparing with other concentrations.

5- Leaves infection and leaves disease severity percentage :

Data in the tables (2) show that applied antioxidants markedly decreased both infection leaves percentage and leaves disease severity of wheat plants grown under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana* , when compared with untreated plants grown under the same conditions . SWE and Zn were most effective antioxidants in decreasing both leaves infection or leaves disease severity percentage.

Table (2): Effect of antioxidants concentrations on leaves infection , disease severity, transmission percentage of fungi from root and stem of wheat plant grown under infection soil by *Fusarium graminearum* or *Bipolaris sorokiniana*.

Treatments	Leaves infection.		disease severity		Transmission percentage of fungi from root.		Transmission percentage of fungi from lower first stem node	
	F.g	B.s	F.g	B.s	F.g	B.s	F.g	B.s
Control	00.0	00.0	1.0	1.0	0.0%	0.0%	0.0%	0.0%
Inf. Soil + water	70.0	75.0	4.0	4.0	100%	100%	100%	80.0%
Inf. Soil + ASA	44.0	45.7	3.0	3.3	60.0%	60.0%	60.0%	46.7%
Inf. Soil + SA	53.3	42.3	3.7	3.3	73.3%	66.7%	73.3%	53.3%
Inf. Soil + Citric	60.0	44.0	4.0	3.0	86.7%	73.3%	86.7%	66.7%
Inf. Soil + GSH	61.3	44.7	4.0	3.3	86.7%	86.7%	86.7%	66.7%
Inf. Soil + SWE	28.0	31.0	2.7	2.7	40.0%	46.7%	40.0%	40.0%
Inf. Soil + HA	47.0	48.3	3.3	3.3	66.7%	80.0%	66.7%	66.7%
Inf. Soil + Put	56.0	58.3	4.0	4.0	86.7%	86.7%	86.7%	66.7%
Inf. Soil + Toco	64.3	59.7	4.0	4.0	93.3%	93.3%	93.3%	73.3%
Inf. Soil + Zn so_4	31.7	25.7	3.0	2.3	60.0%	53.3%	60.0%	46.7%
Inf. Soil + $CaCl_2$	54.0	62.0	3.7	4.0	86.7%	93.3%	86.7%	66.7%
Inf. Soil + H2O2	50.3	53.7	3.7	3.7	80.0%	80.0%	80.0%	66.7%
Mean	47.7	45.4	3.38	3.23	70.8%	70.8%	70.8%	56.9%
LSD at 5%	6.5	6.1	-	-	12.6	12.0	13.0	7.9

ASA : Ascorbic acid **Toco : α- Tocopherol** **SWE : seaweeds extract**
SA : Salicylic acid **Zn so_4 : Zinc sulfite** **HA : Humic acid**
GSH : Glutathione **$CaCl_2$: Calcium chloride** **$H2O_2$: Hydrogen peroxide**
Citric : Citri acid **F.g : *Fusarium graminearum*** **Inf. Soil : Infected soil with Conc.**
Put : Putrescine **B.s : *Bipolaris sorokiniana*.**

6- Transmission percentage :

Data in the tables (3) show the effect of applied antioxidants on fungi transmission percentage from roots , lower first stem node, middle stem node, upper stem node and grains of wheat plants grown under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana*.

Data show that applied antioxidants slightely decreased fungi transmission from root, stem and grains compared with untreated plants grown under the same condition.

SWE and Zn were most effective in this respect. High concentration of applied antioxidants was more effect compared with other of concentrations

High concentration of ASA , SWE , HA , Toco , H_2O_2 were mor effect than the other concentrations in the case of decreasing both leaves infection percentage or leaves disease severity of wheat plants grown under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana*.

Table (3): Effect of antioxidant concentrations on Transmission percentage of fungi from stem and grainsof wheat plant grown under infection soil by *Fusarium graminearum* or *Bipolaris sorokiniana*.

Treatments	Transmission percentage of fungi from middle stem node		Transmission percentage of fungi from upper stem node		Transmission percentage of fungi from wheat grains	
	F.g	B.s	F.g	B.s	F.g	B.s
Control	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
Inf. Soil + water	80.0%	60.0%	80.0%	60.0%	9.6%	4.8%
Inf. Soil + ASA	46.7%	40.0%	40.0%	33.3%	5.6%	3.0%
Inf. Soil + SA	53.3%	46.7%	46.7%	40.0%	7.1%	3.2%
Inf. Soil + Citric	66.7%	53.3%	53.3%	46.7%	7.9%	3.3%
Inf. Soil + GSH	66.7%	53.3%	53.3%	46.7%	8.1%	3.6%
Inf. Soil + SWE	33.3%	33.3%	26.7%	26.7%	4.5%	2.7%
Inf. Soil + HA	53.3%	46.7%	46.7%	46.7%	5.7%	3.7%
Inf. Soil + Put	66.7%	53.3%	60.0%	60.0%	7.8%	3.8%
Inf. Soil + Toco	73.3%	60.0%	73.3%	60.0%	8.7%	4.3%
Inf. Soil + Zn so_4	40.0%	33.3%	33.3%	26.7%	4.9%	2.5%
Inf. Soil + $CaCl_2$	60.0%	60.0%	60.0%	53.3%	6.6%	4.0%
Inf. Soil + H2O2	60.0%	53.3%	53.3%	53.3%	6.3%	3.8%
Mean	53.8%	45.6%	48.2%	42.6%	6.4%	3.4%
LSD at 5%	16.7	14.2	14.0	11.0	0.8	0.6

ASA : Ascorbic acid **Toco : α- Tocopherol** **SWE : seaweeds extract**
SA : Salicylic acid **Zn so_4 : Zinc sulfite** **HA : Humic acid**
GSH : Glutathione **$CaCl_2$: Calcium chloride** **$H2O_2$: Hydrogen peroxide**
Citric : Citri acid **F.g : *Fusarium graminearum*** **Inf. Soil : Infected soil with Conc.**
Put : Putrescine **B.s : *Bipolaris sorokiniana*.**

DISCUSSION

Applied antioxidants decreased pre and post -emergence damping off , infected leaves percentage and leaves disease severity, fungi transmission from root, stem and grains of wheat seedlings growing under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana* in comparing with untreated seedlings growing in the same soil. While enhanced survived seedlings percentage , emergence percentage of wheat plants grown under infected soil with *Fusarium graminearum* or *Bipolaris sorokiniana* compared with untreated plants grown under same condition . SWE and Zn were more effective comparing with other antioxidants in both case of *Fusarium graminearum* or *Bipolaris sorokiniana*.

Exogenous salicylic acid treatment prior to inoculation, however, activated defence gene expression in leaves and provided increased *F. oxysporum* resistance as evidenced by reduced foliar necrosis and plant death. This suggests that salicylate-dependent defences may function in foliar tissue to reduce the development of pathogen-induced wilting and necrosis. (Edgar, *et al.,* 2006).

Durner and Klessig, (1995), reported that salicylic acid (SA) plays an important role in plant defense responses to pathogen attack. One of SA's mechanisms of action is the inhibition, of catalase, resulting in elevated levels of H_2O_2, which activate defense-related genes. SA also inhibits ascorbate peroxidase (APX), the other key enzyme for scavenging H_2O_2.

Glutathione, a major low-molecular weight thiol in plant cells, is increased after pathogen infection (Edwards, *et al.,* 1991). Because ascorbate is a substrate for cell wall peroxidases, it may play a role in the regulation of cell wall lignification, particularly during the HR, through its capacity to inhibit the oxidation of phenolic compounds by peroxidases (Mehlhorn, *et al.,* 1996). The pathogen-induced increase in the peroxidase activity of the cell wall would be effective only in the absence of AA.

The role of H_2O_2 and SA in the defense responses of plants against parasites is controversial. (Chen, *et al .,* 1993) has argued that SA acts *via* inhibition of a catalase that subsequently results in accumulation of H_2O_2 , which may involve cross-linking reactions leading to cell wall toughening (Brisson, *et al .,* 1994) and/or signaling that results in defense gene activation (Chamnongpol, *et al .,* 1998).

It induces the expression of pathogenesis related proteins and initiate the development of systemic acquired resistance and hypersensitivity (Metwally *et al*., 2003). Moreover, White, (1979) demonstrated that application of exogenous salicylic acid or its derivatives induces synthesis of pathogenesis related proteins and partial resistance to pathogens. On the other side, citric acid decrease growth of *Macrophomina phasolaina* and *Rhizoctonia solani* consequently decrease damping-off as well as charcoal rot disease percentage. These findings are in agreement with the finding of Elwakil and El-Metwally, (2000), who shows that the most potent antioxidant on the linear growth of *Cephalosporium* sp., *F. moniliforme*, *F. oxysporum*, *F. solani*, *Rhizoctonia solani*, *Sclerotium bataticola* and *Verticillium* sp. was hydroquinone. Galal, *et al*., (2000) the investigated sensitivity of *Alternaria radicina* and *A. tenuissima in vitro* against five antioxidants (ascorbic acid, benzoic acid, hydroquinone, salicylic acid and tannic acid), they found that the mycelial dry weights of both tested fungi were completely inhibited at 10 mM benzoic acid and salicylic acid.

Evaluation under artificial infection in greenhouse conditions indicated that all natural compounds (mannitol, oxalic acid, citric acid and ascorbic acid), ImM polyamines spermine, omithine and 1% antitranspirants were effective against both pathogens., *Phytophthora infestans* (*Alternaria solani*) Haggag, and EL-Khair, (2007).

The present study demonstrated that these natural compounds not only inhibit the blight pathogens but also have an effect in improving the growth and tuber yield of potato plants.Addition of exogenous ascorbic acid lowered lipid peroxidation in fungal cells and inhibited sclerotial differentiation. GSH plays an important antioxidant role in cells by decreasing ROS level (Lee, *et al.,* 2001).

HA enhanced natural resistance against plant diseases (Scheuerell & Mahaffee, 2004 and 2006), stimulation plant growth through increased cell division, as well as optimized uptake of nutrients and water, (Chen *et al*., 2004). Moreover, HA stimulated the soil microorganisms (Atiyeh *et al*., 2002). Several reports indicated the efficiency of HA in reducing some plant diseases(Bush, 1993).

The role of HA in overcoming the harmful effects of chocolate spot and rust diseases in faba bean plant may be due to the increase in chitinase activity (Abd-El- Kareem, 2007) and stimulation plant growth through increased cell division, as well as optimized uptake of nutrients and water (Atiyeh *et al*., 2002 and Chen *et al*., 2004), regulate hormone level, improve plant growth and enhance stress tolerance (Piccolo *et al*., 1992). Foliar application of HA enhanced antioxidants such as á-tocopherol, â-carotene, superoxide dismutases, and ascorbic acid concentrations in turfgrass species (Zhang, 1997).

El-Ghamry, *et al.,* (2009) stated that growth and yield components and chlorophyll content significantly increased by the application of HA (2000 ppm) interacted with AA (2000 ppm). The maximum reduction of disease severity of chocolate spot was recorded with the interaction between HA at1000 ppm + AA at 1000 ppm .

The results in the present investigation show that antioxidant zinc caused significant reductions in linear growth of both pathogenic fungi . The role of antioxidants on overcoming the injurious effects of both may be attributed to the regulation of plant development and chilling of disease resistance (Achuo, *et al*., 2004). In addition, antioxidants may neutralize the harmful oxygen radicals released during the infections (Shahda, 2002). Also, zinc has a marked effect on the level of auxins due to its important for the synthesis of intermediates in the metabolic pathway, through tryptophan to auxin, which encourage the meristemic activity (Devlin and Witham, 1983). In this investigation, all tested antioxidants and Zn increased photosynthetic pigments in turn, it will increases carbohydrate content in plant tissues. Carbohydrates are the main substances of photosynthetic energy, they comprise structurally polysaccharides of plant cell walls, principally cellulose, hemicelluloses and pectin that consider a barrier against plant pathogens invasion and phenolic compounds are associated with structural carbohydrates, which play a major and important role in plant defense (Hahlbrock and Scheel, 1989).

Zinc is co-factors of Super Oxide Dismutase (SOD), which considered enzymatic antioxidant, hence alleviate the harmful effect of Reactive Oxygen Species (ROS free radicals) caused by fungal stress. (Kostas and Christos, 2006), who found that, the foliar application of microelements (zinc) can be used to reduce the severity of tan spot disease on durum wheat . The positive effect of zinc on increasing the vegetative growth which lead to an increase in plant tolerance and yield components may be due to the role of zinc as essential constituent of three enzymes (Carbonic anhydrase, Alcohol dehydrogenase and superoxide dismutase).). In addition, the enhancement in chlorophyll content by Zn is resulting from stimulating pigment formation and increasing the efficiency of photosynthetic apparatus with a better potential for resistance as well as decreasing phtotophosphorylation rate, which occurred after infection (Amaresh and Bhatt, 1988).

The results in the present investigation demonstrate that calcium salts directly suppress the bitter rot pathogens ,suppressive effects include reduced germ tube growth, reduced mycelial growth *in vitro*, and reduced severity of infection of host tissues pretreated with calcium. Calcium salts also have been shown to reduce mycelial growth *in vitro* and reduce incidence and severity of infection of peach fruits and shoots by *Monilinia fructicola* and *Leucostoma persoonii*, respectively (Biggs, *et al.,* 1997; Biggs, and Peterson, 1990). The mechanisms by which calcium salts inhibit germ tube and mycelial growth are not known. One hypothesis is that high external concentrations of Ca2+ may lead to increased concentration of Ca2+ in the cytosol (Droby, *et al.,* (1997) . Since maintenance of low basal concentrations of internal Ca2+ is essential for normal cell functions, organisms with the inability to regulate intracellular Ca2+ may exhibit compromised growth and development. Calcium ions may reduce the incidence of fungal infection by directly inhibiting fungal growth and by inhibiting cell wall–degrading enzymes produced by the pathogens (Conway, and Sams, 1984 ,Droby, *et al.,* 1997 and Biggs, *et al.* (1997) .

Seaweed extracts have been shown to enhance plant defense against pest and diseases (Allen, *et al.,* 2001). Besides influencing the physiology and metabolism of plants, seaweed products promote plant health by affecting the rhizosphere microbial community. Fungal and bacterial pathogens SWE can serve as an important source of plant defense elicitors (Cluzet, *et al.,* 2004 Khan, *et al.,* 2009). Plants protect themselves against pathogen invasion by the perception of signal molecules called elicitors which include a wide variety of molecules such as oligo and polysaccharides, peptides, proteins, and lipids, often found in the cell wall of attacking pathogens (Boller, 1995). A variety of polysaccharides present in algal extracts include effective elicitors of plant defense against plant diseases (Kloareg and Quatrano, 1988). Although red algae typically(SWE) contain agars and carrageenans in their cell walls, extracts of brown algae contain alginates, laminarans, sulfated fucans, and other complex mucilages, and green algae . contain mucilages composed of units such as rhamnose, uronic acid, and xylose (Cluzet, *et al.,* 2004). Laminaran, a linear b-(1,3)-glucan, and sulfated fucans from brown algae elicit multiple defense responses in alfalfa and tobacco (Kobayashi, *et al.,* 1993; Klarzynski, *et al.,* 2000, 2003). Simailarly, carrageenans, a family of sulfated linear galactans, are effective elicitors of defense in tobacco plants (Mercier, *et al.,* 2001). Foliar sprays of SWE reduced Phytophthora capsici infection in Capsicum and Plasmopara viticola in grape (Lizzy, *et al.,* 1998).

Seaweeds are a rich source of antioxidant polyphenols with bactericidal properties (Zhang, *et al.,* 2006). Who added that the application of A. nodosum extract and humic acid to bentgrass (*Agrostis stolonifera*) increased SOD activity, which in turn significantly decreased dollar spot disease caused by *Sclerotinia homoeocarpa.* Treatment of alfalfa with the algal extracts prior to pathogen challenge resulted in an increased resistance to *Colletotrichum*..

REFERENCES

Abd El-Kareem, F. (2007). Induced Resistance in Bean Plants Against Root Rot andAlternaria leaf spot Diseases Using Biotic and Abiotic Inducers under Field Conditions. Res.J. Agric. and Biol. Sci., 3(6): 767-774.

Abou- Table, A. H.A. (2006).Further studies on seed-borne disease of wheat. Ph. D. Thesis, Faculty of Agric., Mansoura Univ.

Achuo, E.A., Audenaert, K., Meziana, H. and Hofte, M. (2004). The salicylic acid-dependent defense pathway is effective against different pathogens in tomato and tobacco. Plant Pathol., 53: 65-72.

Adam, A., Farkas, T., Somlyai, G., Hevesi, M., and Kiraly, Z. (1989). Consequence of 02- generation during a bacterially induced hypersensitive reaction in tobacco: deterioration of membrane lipids. Physiol Mo1 Plant Pathol, 34: 13-26.

Agrios, G.N. (1988). Plant Pathology. (London: Academic Press).

Allen, R.D., Webb, R.P., and Schake, S.A. (1997). Use of transgenic plants to study antioxidant defences. Free Radical Biology and Medicine, 23: 473–479.

Amaresh, C. and Bhatt, R.K. (1988). Biochemical and physiological response to salicylic acid in reaction to systemic acquired resistance Photosynthetica, 35: 255-258.

Apostol, I., Heinstein, P.F. and Low, P.S. (1989). Rapid stimulation of an oxidative burst during elicitation of cultured plant cells. Role in defense and signal transduction. Plant Physiol, 90: 109-116.

Atiyeh, R.M., Lee, S., Edwards, C.A., Arancon, N.Q. and Metzger, J.D. (2002). The influence of humic acids derived from earthworm processed organic wastes on plant growth. Bioresource Technology, 84: 7-14.

Biggs, A. R., and Peterson, C. A. (1990). Effect of chemical applications to peach bark wounds on accumulation of lignin and suberin and susceptibility to *Leucostoma persoonii*. Phytopathology 80:861-865

Biggs, A. R., El-Kholi, M. M., El-Neshawy, S., and Nickerson, R.(1997). Effects of calcium salts on growth, polygalacturonase activity, and infection of peach fruit by *Monilinia fructicola*. Plant Dis. 81:399-403.

Boller, T. (1995). Chemoperception of microbial signals in plant cells.Annu. Rev. Plant Physiol. Plant MOI. Biol. 46, 189-214.

Brisson, L.F., Tenhaken, R., and Lamb, C, (1994). Function of oxidative cross-linking of cell wall structural proteins in plant disease resistance. Plant Cell, 6: 1703–1712.

Bush, D.R., (1993). Proton coupled sugar and amino acid transporters in plants. Annual Review of Plant.

Chamnongpol, S., Willekens, H., Moeder, W., Langebartels, C., San-Dermann, H., van Montagu, M., Inze, D., and van Camp, W. (1998). Defense activation and enhanced pathogen tolerance induced by H2O2 in transgenic tobacco. Proc Natl Acad Sci, 95: 5818–5823.

Chen, Y., De Nobili, M. and Aviad, T. (2004). Stimulatory effect of humic substances on plant growth. In "Soil organic matter in sustainable agriculture".(Eds F. Magdoff, R.R. Weil). 103-130, Boca Raton, FL.

Chen, Y.P., Xing1, L.Ping., Wu, G.J., Wang, H.Z., Wang1, X.E., Cao1,A.Z., and Chen1, P.D. (2007). Plastidial glutathione reductase from Haynaldia villosa is an enhancer of powdery mildew resistance in Wheat (Triticum aestivum). Plant and Cell Physiology , 48 (12): 1702-1712.

Chen, Z., Silva, H., and Klessig, D.F. (1993). Active oxygen species in the induction of plant systemic acquired resistance by salicyiic acid. Science, 262: 1883-1886.

Cluzet, S., Torregrosa, C., Jacquet, C., Lafitte, C., Fournier, J., Mercier, L., Salamagne, S., Briand, X., Esquerre-Tugaye, M.T.and Dumas, B. (2004). Gene expression profiling and protection of Medicago truncatula against a fungal infection in response to an elicitor from the green alga Ulva spp. Plant Cell Environ 27:917–928.

Conway, W. S., and Sams, C. E. (1984). Possible treatment reduces decay in apples. Phytopathology 74:208-210.

Devlin, R.M. and Witham, F.H. (1983). Plant Physiology. 4th Edn., A Division of Wads Worth, Inc., Wadads Worth Publishing Co., Belmont, California.

Droby, S., Wisniewski, M. E., Cohen, L., Weiss, B., Touitou, D., Eilam, Y., and Chalutz, E. (1997). Influence of CaCl2 on *Penicillium digitatum*, grapefruit peel tissue,and biocontrol activity of *Pichia guilliermondii*. Phytopathology 87:310-315.

Durner, J., and Klessig, D.F. (1995). Inhibition of ascorbate peroxi-dase by salicylic acid and 2,6-dichloroisonicotinic acid, two inducers of plant defense responses. Proc. Natl. Acad. Sci. USA, 92: 11312-1 1316

Edgar, C. I., McGrath, K. C., Dombrecht , B. , Manners, J. M., Maclean, D. C. Schenk, P. M., and Kazan , K. (2006). Salicylic acid mediates resistance to the vascular wilt pathogen *Fusarium oxysporum* in the model host Arabidopsis thaliana. Australasian Plant Pathology, 35(6): 581–591.

Edwards, R., Blount, J.W., and Dixon, R.A. (1991). Glutathione and elicitation of the phytoalexin response in legume cell cultures. Planta, 184: 403–409.

El-Ghamry,A.M., Abd El-Hai, K.M. and Ghoneem, K.M. (2009). Amino and Humic Acids Promote Growth, Yield and Disease Resistance of Faba Bean Cultivated in Clayey Soil. Australian Journal of Basic and Applied Sciences, 3(2): 731-739.

Elwakil, M.A. and M.A. El-Metwally, (2000). Hydroquinone, a promising antioxidant for managing seed-borne pathogenic fungi of peanut. Pak. J. Biol. Sci., 3: 374-375

Galal, A.A.; Shaat, M.M.N and El-Bana, A.A. (2000). Sensitivity of *Alternaria radicinia* and *A. tenuissima* to some antioxidants compounds. J. Agric. Sci. Mamsoura Univ., 25:1553-1562.

Grant, J. J., and Loake, G. J. (2000). Role of Reactive Oxygen Intermediates and Cognate Redox Signaling in Disease Resistance. Plant Physiology,١٢٤ : 21–29.

Guo, Z.J., Nakagawara, S., Sumitani, K., and Ohta, Y. (1993). Effect of intracellular glutathione level on the production of 6-methoxymellein in cultured carrot (Daucus carota) cells. Plant Physiol, 102: 45–51.

Haffz, Y.M. (2005). Biochemical and molecular studies on role of reactive oxygen species and antioxidants in plant disease resistance. PH. D. Thesis, Fac. of Szwnt Istvan Univ.

Haggag, W. M., and EL-Khair, H. A. (2007). Application of some natural compounds for management of potato late and early blights. International journal of food, agriculture and environment, vol. 5 (2): 157-163.

Hahlbrock, K. and Scheel, D. (1989). Phsiology and molecularbiology of phenyl propanoid metabolism. Annu. Rev. Plant Physiol. Plant Mol. Biol., 40: 347-369.

Hammond-Kosack, K. E., and Jones' J. D. G. (1996). Resistance Gene-Dependent Plant Defense Responses. The Plant Cell, Vol. 8: 1773-1791.

Huckelhoven, R., Fodor, J., Preis, C., and Kogel, K.H. (1999) . Hypersensitive Cell Death and Papilla Formation in Barley Attacked by the Powdery Mildew Fungus Are Associated with Hydrogen Peroxide but Not with Salicylic Acid Accumulation1. Plant Physiology, Vol. 119: pp. 1251–1260.

Khan, W., Rayirath , U. P., Subramanian, S., Jithesh , M. N., Rayorath, P., Hodges, D. M., Critchley, A. T., Craigie, J. S., Norrie, J. and Prithiviraj, B. (2009). Seaweed Extracts as Biostimulants of Plant Growth and Development. J Plant Growth Regul (2009) 28:386–399

Klarzynski, O., Descamps, V., Plesse, B., Yvin, J.C., Kloareg ,B. and Fritig, B. (2003). Sulfated fucan oligosaccharides elicit defense responses in tobacco and local and systemic resistance against tobacco mosaic virus. Mol Plant Microbe Interact 16:115–122

Klarzynski, O., Plesse, B., Joubert, J.M., Yvin, J.C., Kopp, M., Kloareg ,B. and Fritig, B. (2000). Linear beta-1, 3 glucans are elicitors of defense responses in tobacco. Plant Physiol 124:1027–1038.

Klessig, D.F., and Malamy, J. (1994). The salicylic acid signal in plants. Plant MOI. Biol. 26: 1439-1458.

Kloareg, B., Quatrano, R.S. (1988). Structure of the cell walls of marine algae and ecophysiological functions of the matrix polysaccharides. Oceanogr Mar Biol Annu Rev 26:259–315.

Knogge, W. (1996). Funga1 infection of plants. Plant Cell, 8:1711-1722.

Kobayashi, A., Tai, A., Kanzaki, H. and Kawazu, K. (1993). Elicitor-active oligosaccharides from algal laminaran stimulate the production of antifungal compounds in alfalfa. Z Naturforsch 48c:575–579.

Kostas, B.S. and Christos, D. (2006). Effect of foliar applied boron, manganese and zinc on tan spot in winter durum wheat. Crop Prod., 25: 657-663.

Lee, J. H., Choi, I. Y., Kil, I. S., Kim, S. Y., Yang, E. S., and Park, J. (2001). Biochim. Biophys. Acta, 1526, 191- 198.

Levine, A., nhaken, R., ixon, R., and Lamb, C. (1994). H2O2 from the oxidative burst orchestrates the plant hypersensitive disease esistance response. Cell,79: 583–593.

Lizzi, Y., Coulomb, C., Polian, C., Coulomb, P.J. and Coulomb, P.O. (1998). Seaweed and mildew: what does the future hold? Laboratory tests have produced encouraging results [L'algue face au mildiou: quel avenir? Des resultats de laboratoire tres encourageants].Phytoma 508:29–30

Mathre, D. E., Johnston, R. H., and Grey, W. E. (2003). Diagnosis of common root rot of wheat and barley. Online. Plant Health Progress doi:10.1094/PHP-2003-0819-01-DG.

Mehlhorn, H., Lelandais, M., Korth, H.G., and Foyer, C.H. (1996). Comparison of ascorbate-dependent peroxidase activity in horseradish peroxidase types I and II and in leaf extracts. FEBS Letts, 378: 203 – 206.

Mercier, L., Lafitte, C., Borderies, G., Briand, X., Esquerre-Tugaye, M.T. and Fournier, J. (2001). The algal polysaccharide carrageenans can act as an elicitor of plant defence. New Phytol 149:43–51

Metwally, A., Finkemeier, I., Georgi, M. and Dietz, K.J. (2003). Salicylic acid alleviates the cadmium toxicity in barly seedlings. Plant Physiology, 132: 272-281.

Mohammadi, M. and Kazemi, H. (2002). Changes in peroxidase and polyphenol oxidase activities in susceptible and resistant wheat heads inoculated with Fusarium graminearum and induced resistance. Plant Science, Vol. 162, Issue 4, pp. 491-498.

Murray, T.D., Parry, D.W., and Cattlin, N.D. (1998). A color handbook of diseases of small grain cereal crops. Iowa State University Press, Ames, Iowa.

Parry, D.W., Jenkinson, P., and McLeod, L. (1995). Fusarium ear blight (scab) in small grain cereals—a review. Plant Pathol. 44: 207–238.

Piccolo, A., S. Nardi and G. Concheri, (1992). Structural characteristics of humic substances as regulated to nitrate uptake and growth regulation in plant systems. Soil Biochem., 24: 373-380.

Ryals, J.A., Neuenschwander, U.H., Willits, M.G., Molina, A., Steiner, H. Y., and Hunt, M.D. (1996). Systemic acquired resistance. Plant Cell, 8: 1809-1819.

Scheuerell, S.J. and Mahaffee, W.H. (2004). Compost tea as a container medium drench for suppressing seedling damping off caused by Pythium ultimum. Phytopathology, 94: 1156-1163.

Scheuerell, S.J. and Mahaffee, W.H. (2006). Variability associated with suppression of Gray Mold (Botrytis cinerea) on Geranium by foliar applications of nonaerated and aerated compost teas. Plant Dis., 90: 1201-1208.

Shahda, W.T. (2001). Antifungal activety of salicylic acid for controlling Alternaria alternata of tomato. J. Agric Sci., Mansoura Univ., 26: 6117-6129.

Shao, H.B., Chu, L.Y., Lu, Z.H., and Cong-Min Kang, C.M. (2008). Primary antioxidant free radical scavenging and redox signaling pathways in higher plant cells. International Journal of Biological Sciences, 4 (1): 8-14.

Sivanesan, A. (1987). Graminicolous Species of *Bipolaris*, *Curvularia*, *Drechslera*, *Exserohilum* and their Teleomorphs. Mycological Papers 158, 261.

Zhang, Q., Zhang, J., Shen, J., Silva, A., Dennis, D.A., Barrow, C.J. (2006). A simple 96-well microplate method for estimation of total polyphenol content in seaweeds. J Appl Phycol 18:445–450

Zhang, X. (1997). Influence of plant growth regulators on turfgrasss growth, antioxidant status, and drought tolerance. Ph.D. thesis, Fac. Of Virginia Polytechni (Institute and State University).

التغلب على الاثار الضارة لبعض فطريات التربة على نبات القمح النامى تحت ظروف التربة المعدية بفطر الفيوزاريوم أو البايبولارس.

محب طه صقر* ، زين العابدين عبد الحميد محمد*، محمود ابراهيم العميرى و مجدى سعد الدين أبو الدهب****

*** قسم النبات الزراعى – كلية الزراعة – جامعة المنصورة**

**** وحدة بحوث تكنولوجيا البذور – معهد بحوث المحاصيل الحقلية – مركز البحوث الزراعية**

أجريت تجربة أصص لدراسة دور بعض مضادات الأكسدة (نقعا ورشا) للتغلب على الاثار الضارة الناجمة عن العدوى بفطرى الفيوزاريوم و البايبولارس لنبات القمح.

أتضح أن المعاملة بمضادات الأكسدة أدت الى نقص واضح فى كل من البادرات الميتة قبل و بعد انبثاقها فوق سطح التربة، نسبة إصابة الأوراق، شدة إصابة أوراق نبات القمح ، نسبة انتقال الفطر من الجذر والساق والحبوب لنباتات القمح النامية تحت ظروف التربة المعدية بأى من الفطرين تحت الدراسة بينما أدت المعاملة بمضادات الأكسدة المستخدمة إلى زيادة كل من نسبة انبثاق البادرات وكذلك النسبة النهائية للبادرات التى استمرت فى النمو لنباتات القمح مقارنة بالنباتات النامية تحت نفس الظروف والغير معاملة بمضادات الأكسدة كما أتضح أيضا أن التركيز الأعلى لمضادات الأكسدة فى معظم الأحوال كان هو الأكثر فعالية مقارنة بالتركيزات الأقل.

قام بتحكيم البحث

أ. د/ محمد نصر الدين هلالى	**كلية الزراعة – جامعة المنصورة**
أ. د/ حسنى محمد عبد الدايم	**كلية الزراعة – جامعة بنها**