



INDUCTION OF SYSTEMIC RESISTANCE IN CUCUMBER PLANTS AGAINST POWDERY MILDEW UNDER FIELD CONDITIONS

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ABSTRACT

Cucumber (*Cucumis sativus* L.) plants are liable to infect by many fungal pathogens. However, powdery mildew caused by *Erysiphe cichoracearum* and *Sphaerotheca fuliginea*, is a very damaging. Foliar application of cucumber plants cv. Beit Alpha with the fungicide thiophanate - methyl (Topsin 70% WP) as a comparison treatment, potassium silicate (5 m/l), dry yeast (4000 mg/l) and ascorbic acid (100 mg/l), significantly reduced disease severity by 86.00, 68.31, 50.86 and 42.03%, respectively compared with the untreated plants in 2013 season. The same trend was found in 2014 season (86.02, 71.33, 48.06 and 45.27%), respectively. Moreover, the aforementioned treatments significantly increased each of plant height/plant, number of leaves/plant, dry weight/plant and number of flowers as well as yield components (number of fruits/plant and mean of fruits weight/plant). Also, the treatments increased peroxidase activity and total phenolic content. Reduction in disease severity was positively correlated with both of peroxidase activity ($R^2 = 56.3$, $P < 0.005$) and total phenolic content ($R^2 = 77.2$, $P < 0.005$). Foliar application with potassium silicate, dry yeast and ascorbic acid at the rate of 5ml, 4000 mg and 100 mg/l induced new pathogenesis related proteins (PR-protein) with various molecular weights in the shoots of cucumber plants upon natural infection with the powdery mildew pathogen. Furthermore, these treatments resulted in a significant increase of N, P, K contents of vegetative shoots.

Key words: Cucumber, powdery mildew, foliar spray, induced systemic resistance, peroxidase activity, total phenolic content, pathogenesis-related protein.

INTRODUCTION

Cucumber (*Cucumis sativus* L.) is one of the most important and popular vegetable crops all over the world and Egypt. The crop is mainly cultivated during the summer season in the field, while it could be grown in two other plantations, *i.e.*, autumn and spring under greenhouse conditions.

Powdery mildew of cucumber caused by *Erysiphe cichoracearum* and *Sphaerotheca fuliginea* is considered the most economically important and widespread throughout the world in the greenhouse and fields (Brunell and Davi, 1987). Severe infection can cause premature leaf senescence, resulting in reduction of photosynthesis and transpiration efficiencies by

the plant, leading to stunted and weakened plants (Mc Grath and Thamas, 1996). Fungicides are used to control several plant diseases; however, they increase production cost, health problem and pollution of environment. Therefore, it must be search to an alternative strategies for controlling plant diseases.

Silicon (Si) has been reported as a beneficial element that might enhance plant growth and protecting plants against diseases and abiotic stress (Ma, 2004). The spray with Si was found to be effective in the control of powdery mildew in cucumber; muskmelon and zucchini squash plants (Menziez *et al.*, 1992). Some chemicals were reported as systemic acquired resistance inducers against different plant diseases. In this concern, ascorbic acid (AA) was reported for

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inducing resistance in many plants to fungi (El-Tohamy *et al.*, 2008 ; Sadak *et al.*, 2008). Ziedan and Farrag (2011) showed that yeasts suppressed powdery mildew and cercospora leaf spot disease on sugar beet. Furthermore, Abdel-Kader *et al.* (2012) found that plant sprayed with treatments of chemical inducers and *S. cerevisiae* under the greenhouse conditions led to a reduction in the incidence of powdery and downy mildew diseases on cucumber, cantaloupe and pepper. Ben Rejeb *et al.* (2014) found that plants confronted with both abiotic and biotic stresses showed serious and constant reduced productivity. Plant responses to these stresses are complex and involve numerous physiological, molecular, and cellular adaptations. Treatment with abiotic and biotic inducers activated different defense mechanisms such as accumulation of pathogenesis related protein (PR) in plant tissues (Khan *et al.*, 2011). Also, these inducers increased phenolic content, peroxidase and chitinase activity (Quiroga *et al.*, 2000 ; Cai *et al.*, 2008).

The objectives of this investigation were to determine the extent of powdery mildew disease control in cucumber plants through foliar spraying with each of potassium silicate, ascorbic acid and dry yeast compared with the Topsin - M under the field conditions. The effect of these agents on total phenolic content, peroxidase activity, some probably of defense mechanisms and some plant parameters were also studied.

MATERIALS AND METHODS

Evaluation of some plant resistance inducers against powdery mildew of cucumber was performed under field conditions at Etay El-Baroud Agricultural Research Station, Beheria Governorate, during 2013 and 2014 growing seasons using cucumber cultivar Beit Alpha sown on the first week of May. The chemical inducers used in the present study were solution of potassium silicate (K_2SiO_3), as Si [PQ Corporation-USA], composed of 29% SL. potassium silicate was applied at the rate of 2.5 and 5.0 ml/l water and ascorbic acid ($C_6H_8O_6$) (50 and 100 mg/l water) produced by (Sigma Aldrich Chemical Co. St. Louis, Mo, USA). In addition, active dry yeast which was prepared by

adding to sugar at ratio 1:1 and dissolves in warm water at 32°C, kept overnight, and left three hours for activation and reproduction of dry yeast then used at the rate of 2000 and 4000 mg/L water. Plants were sprayed twice at 15 days interval beginning after 30 days from sowing. Topsin 70% WP (70 g/100 l water) was used as control. The selected concentrations were mixed with adhesive surfactant biofilm 1265 at 30 ml/100 l water and hand homogenized before spraying on the upper and lower leaf surface of cucumber plants (15 days old). Another set of plants were sprayed with water mixed with the adhesive surfactant and served as control treatment.

The experimental plot contained 5 ridges (1.10 × 3.5 m). Seeds were soaked in the water for six hours and kept in cheesecloth for 12 hours before sowing. Seeds were planted in hills (2 seeds/hill) on one side of ridge with 50 cm between hills. All agricultural practices were carried out according to the recommendations of the Ministry of Agricultures, Egypt. Complete randomized block design with four replicates for each treatment was used.

Disease Severity Determination

Disease severity scale of Lewellen and Schrandt (2001) was followed; percentage of disease severity (DS) was calculated as follows:

$$DS (\%) = \frac{\sum (\text{disease grade} \times \text{number of plants in each grade})}{(\text{total number of plants} \times \text{highest disease grade})} \times 100$$

Also, the efficiency percentage was recorded weekly for five weeks and calculated as follows:

$$\text{Efficiency (\%)} = \frac{\text{Disease severity in the control} - \text{disease severity in the treatment}}{\text{Disease severity in the control}} \times 100$$

Growth Parameters

Cucumber plants were randomly selected from the middle ridge of each plot, leaving two ridges from each side to avoid border effects. Plant length/ plant; number of leaves/plant and dry weight/plant were recorded 75 days after sowing.

Average number of fruits/plant was also estimated by harvesting fruits at marketable size of each treatment and were recorded, and

accumulated yield was expressed as number and weight of fruits/plant.

Biochemical Changes

Effect of resistance inducers on peroxidase activity and total phenolic content

In the 2nd season, the peroxidase activity and total phenolic content were determined after 10 days from the second foliar spray application.

Leaf samples from each treatment were collected for peroxidase activity assay, three grams of fresh leaf tissues was ground in a precooled mortar and pestle containing 9 ml of 0.1 M phosphate buffer (pH 7.1). The extract was centrifuged at 3000 rpm at 6°C for 20 mins. Peroxidase activity was expressed as changes in absorbance min⁻¹ at 425 nm, according to the method proposed by Allam and Hollis (1972).

Total phenolic content in fresh leaves extract was determined using the colourimetric method described by (Anonymous, 1985).

Protein extraction and electrophoretic analysis

Leaf samples were collected from different treatments and kept frozen at approximately -80°C. Three grams of each tested sample were ground using a mortar in a liquid nitrogen until the sample was completely homogenized. These homogenates were transferred into Eppendorf tubes (1 ml), each containing 200 µl of extraction buffer (50 mM tris-HCl buffer, pH 6.8, glycerol 10% W/V, ascorbic acid 0.1%, cysteine hydrochloride 0.1% W/V), then centrifuged at 18,000 rpm for one min to remove debris. The protein content in the supernatant was estimated according to Bradford (1976) using bovine serum albumin as a standard protein.

Sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE) was performed by the method described by Laemmli (1970). Proteins were analyzed on 1.5-mm thick and 15-cm long gels run in a dual vertical slab unit (Hofer Scientific Instruments, San Francisco, CA, USA). Twenty-microlitre samples (40 µg of protein), loaded on the gel, were subjected to electrophoresis. The separation gel (10%) and stacking gel (3.5%) were prepared from an acrylamide monomer solution (Roth, Karlsruhe,

Germany). Protein was electrophoresed at a constant current of 30 mA through the stacking gel, and at 90 mA through the separation gel at room temperature, the gels were stained by silver nitrate (Rabilloud *et al.*, 1988).

NPK contents of cucumber shoots

Determination of total nitrogen by microkjeldahl methods, potassium using flame photometer (Kalra, 1998) and phosphorus using ammonium molybdate (Cooper, 1977) were carried out in cucumber dry shoots.

Statistical Analysis

Analysis of variance was carried out using MSTAT-C (1991) program version. Least significant difference (LSD) was employed to test the significant differences between treatments at P ≤ 0.05% (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

Effect of Resistance Inducers on Disease Severity of Cucumber Powdery Mildew

The obtained results presented in Table 1 reveal that all treatments have positive effect on decreasing disease severity compared with the control. The disease severity was significantly reduced in case of the treatments tested in the first season. Fungicide Topsin 70% WP (0.7 g/l) and potassium silicate (5 and 2.5 ml/l) reduced disease severity by 7.25, 16.40 and 19.15%, respectively followed by dry yeast (4000 g/l) and ascorbic acid (100 mg/l) compared with the control (51.75%). The same trend was recorded with the data obtained in the second season. These results are in harmony with the findings of Wolff *et al.* (2008). They found that foliar application of silicon 8.6% as a fine mist to cucumber leaf blades twice per week reduced powdery mildew infection by 87%. Moreover, Ziedan and Farrag (2011) found that fungicide Topsin was the best treatment; it completely suppressed powdery mildew on sugar beet plants followed by *S. cerevisiae*. Bowen *et al.* (1992) and Menzies *et al.* (1992) reported that leaf application with potassium silicate controlled powdery mildew disease on leaves of cucumber, grape, muskmelon and zucchini squash plants. Furthermore, Cacique *et al.* (2013) reported that blast symptoms were

Table 1. Effect of foliar spraying with potassium silicate, ascorbic acid, dry yeast and Topsin on percentage of disease severity on cucumber plants during two tested growing seasons (2013 and 2014)

Treatment	Concentration	Percentage of disease severity (%)			
		1 st season	Efficiency (%)	2 nd season	Efficiency (%)
Potassium silicate (ml/l)	2.5	19.15	63.00	16.45	64.62
	5.0	16.40	68.31	13.33	71.33
Ascorbic acid (mg/l)	50	34.97	32.43	32.20	30.75
	100	30.00	42.03	25.45	45.27
Dry yeast (mg/l)	2000	33.38	35.50	28.10	39.57
	4000	25.43	50.86	24.15	48.06
Topsin (g/l)	0.7	7.25	86.00	6.50	86.02
Control	0.0	51.75	-	46.50	-
LSD at 5%		2.11		3.331	

reduced on leaves of rice plants that were root or foliar supplied with soluble Si, indicating the importance of this element to increase resistance against *Pyricularia oryzae* infection. It was demonstrated by X-ray microanalysis that Si deposition was very similar on the leaf epidermis of plants sprayed with soluble Si or growing in soil amended with calcium silicate contributing. In addition, El-Gamal Nadia (2003) reported that applied yeast (*S. cerevisiae*) on cucumber plants reduced severity of powdery mildew with an average of 41.7% compared with the control. The mode of action of yeast (*S. cerevisiae*) was observed on sugar beet surfaces by scanning electron microscope as complete malformation and lyses of *Erysiphe betae* hyphae with high yeast colonization (Ziedan and Farrag, 2011). Abdel-Kader *et al.* (2012) found that plants sprayed with ascorbic acid only or combined with *S. cerevisiae* under the greenhouse conditions, showed a reduction in the disease incidence of powdery and downy mildews on cucumber, cantaloupe and pepper plants.

Effect of Resistance Inducers on Some Growth Parameters

Data presented in Table 2 indicate that spraying plants by potassium silicate, ascorbic acid and yeast extract, significantly increased all

studied parameters including plant length, number of leaves and dry weight as well as yield components of cucumber plants (number and weight of fruits). In the first season, the highest value of plant length was found in case of plant treated with fungicide Topsin, being 98.75 cm. This might be due to reduction of the disease severity in addition to the increase of vegetative characters. In this respect, applications of dry yeast (4000 and 2000 mg/l) was the best treatment followed by ascorbic acid (100 mg/l). The analogous values were 92.75, 83.75 and 82.75 cm, respectively. These findings are in agreement with the results obtained by El-Lethy *et al.* (2011) who reported that the increase in vegetative growth of geranium plants as a result of ascorbic acid and dry yeast treatments might essentially be due to ascorbic acid is currently considered to be a regulator of cell division and differentiation involved in wide range of important functions as antioxidant, photoprotection and regulation of photosynthesis and growth. Yeast has also been considered as a natural source of cytokinins that stimulate cell division and enlargement as well as the synthesis of protein, nucleic acid and chlorophyll (Fathy and Farid, 1996). In the second season, Topsin, dry yeast (4000 mg/l)

Table 2. Effect of foliar spray with potassium silicate, ascorbic acid, dry yeast and Topsin on some growth parameters and yield components of cucumber plants during two growing seasons (2013 and 2014)

Treatments	Concentrations	1 st season						2 nd season					
		Plant length (cm)	Number of leaves /plant	Dry weight (g/plant)	Number of flowers/plant	Number of fruits/plant	Yield (Kg/plant)	Plant length (cm)	Number of leaves /plant	Dry weight (g/plant)	Number of flowers/plant	Number of fruits/plant	Yield (Kg/plant)
Potassium silicate (ml/l)	2.5	80.00	27.0	13.05	17.5	6.4	0.58	98.50	31.5	18.46	20.0	9.4	0.83
	5.0	81.00	30.0	13.68	20.5	9.4	0.84	106.0	38.5	20.27	23.3	12.2	1.21
Ascorbic acid (mg/l)	50	74.00	24.5	10.18	17.5	6.6	0.56	90.00	27.0	14.48	18.5	7.4	0.80
	100	82.75	29.0	13.27	18.0	8.6	0.81	99.00	31.5	17.38	20.8	9.6	0.92
Dry yeast (mg/l)	2000	83.75	35.0	14.52	20.8	9.6	0.90	100.8	33.8	20.29	24.5	14.4	1.28
	4000	92.75	39.5	16.63	23.3	9.6	0.97	107.3	42.8	22.65	28.5	17.4	1.32
Topsin (g/l)	0.7	98.75	43.3	18.37	31.8	13.2	1.22	110.5	50.0	28.60	39.8	20.0	1.82
Control	0.0	70.00	20.8	8.45	12.0	5.4	0.50	80.00	23.0	10.61	15.0	7.2	0.64
LSD at 5%		5.89	2.18	1.34	2.44	0.69	0.09	9.79	3.00	2.88	2.11	0.68	0.07

and potassium silicate (5 ml/l) increased plant length. The corresponding values were 110.5, 107.3 and 106.0 cm, respectively. Number of leaves/plant was significantly increased in plants sprayed with Topsin, dry yeast (2000 and 4000 mg/l) and potassium silicate (5 ml/l), followed by ascorbic acid (100 ml/l). The analogous values were 43.3, 39.5, 35.0, 30.0 and 29.0 leaves plant on the average, respectively. Shalaby and El-Ramady (2014) reported that number of leaves and bulb quality of garlic plants increased by foliar application with ascorbic acid, yeast, amino acids and seaweed extract. Silicon positively influences plant growth and biomass production, especially monocotyledons as a consequence of improve tissue rigidity, better angle of leaves and light interception, improving photosynthesis (Ma, 2004). Si might be involved in cell elongation and division processes, as well as in the hormone balance (Elawad *et al.*, 1982). Similar results were obtained in the second season. Also, the number of flowers per plant were highest when the fungicide and dry yeast (4000 mg/l) were used being 31.8 and 23.3 flowers/plant.

The other concentration of yeast extract gave a similar value as the application with potassium silicate (5 ml/l) (20.8 and 20.5 flowers), on the average, respectively. In the second season, Topsin, dry yeast (4000 and 2000 mg/l) and potassium silicate (5 ml/l) increased number of flowers. The corresponding values were 39.8, 28.5, 24.5 and 23.3 flowers, on the average, respectively. Data in the same table also indicate that yield of fruits (number and weight) increased in different treated plants; clear negative relation was observed between disease severity and yield. The highest number of fruits was recorded in the second season with treatments of Topsin, dry yeast (2000 and 4000 mg/l) and potassium silicate (5 ml/l). The corresponding values were 20.0, 17.4, 14.4 and 12.2 fruits per plant on the average, respectively. In this respect, using *S. cerevisiae* as single treatment or combined with K_2HPO_4 or KH_2PO_4 increased number of fruits per plant by 63.8, 57.1 and 42.8%, respectively (El-Gamal, 2003). Generally, dry weight of plants were significantly increased in the plants sprayed with Topsin, dry yeast, ascorbic acid (100 mg/l) and

potassium silicate (5 ml/l) in both seasons. El-Tohamy *et al.* (2008) reported that eggplant, sprayed with dry yeast and ascorbic acid increased its vegetative growth. Application of ascorbic acid on wheat plants caused an increase of dry weight compared with the untreated plants (Sadak *et al.*, 2008).

Effect of Resistance Inducers on Peroxidase Activity and Total Phenolic Content

Data in Table 3 indicate that the two applied concentrations of potassium silicate, ascorbic acid and dry yeast, significantly increased peroxidase activity (PO) and total phenolic content in cucumber leaves under the field conditions compared with control. Peroxidase activity was also increased, in general, as the concentration of the tested compounds increased. Foliar treatments with the highest concentration of potassium silicate (5.0 ml/l), dry yeast (4000 mg/l) and Topsin led to the highest increase of PO activity. The analogous values were 0.624, 0.573 and 0.570 on the average, respectively followed by ascorbic acid (100 mg/l), being 0.486. In this concern,

increasing activity of (PO) in plants treated with various agents was recorded by many investigators. Kombrink and Somssich (1995) found that the increase in the activities of PO and polyphenoloxidase (PPO) have been thought to be key components in local and systemic disease resistance. Another explanation was advanced by Quiroga *et al.* (2000) who found that the activities of PO and PPO are important in defense mechanism against pathogens, through their role in the oxidation of phenolic compounds to quinones, with higher antimicrobial activity. Therefore, they might directly involved in stopping pathogen development. Moreover, Silicon nutrition increased lignin content and enhanced activities of peroxidase, polyphenoloxidase, and phenylalanine ammonialyase (PAL) in the infected rice leaves by blast (Cai *et al.*, 2008). So, ascorbic acid protect metabolic process against H₂O₂ and other toxic derivatives of oxygen affect enzyme activities as well as minimize the damage caused by oxidative process through synergistic function with other antioxidant and stabilize membranes (Shao *et al.*, 2008).

Table 3. Peroxidase activity and total phenolic content in cucumber plants grown after foliar spray with potassium silicate, ascorbic acid, dry yeast and Topsin in the 2nd season of 2014

Treatment	Concentrations	Peroxidase activity*	Total phenolic content (mg/g fresh weight of leaves)
2nd season			
Potassium silicate (ml/l)	2.5	0.420	0.451
	5.0	0.624	0.614
Ascorbic acid (mg/l)	100	0.361	0.262
	200	0.486	0.427
Dry yeast (mg/l)	2000	0.382	0.389
	4000	0.573	0.587
Topsin (g/l)	0.7	0.570	0.625
Control	0.0	0.268	0.203
LSD at 5%		0.015	0.007

* Peroxidase activity expressed as change in absorbance at 425nm/g fresh weight/20 mins.

Concerning to the amount of total phenolics, the data in Table 3 show that Topsin and potassium silicate (5m l/l) caused a significant increase in total phenolic content, followed by dry yeast (4000 mg/l) and ascorbic acid (100 mg/l). The corresponding values were 0.625, 0.614, 0.587 and 0.427 mg/g fresh weight on the average, respectively. These results are in agreement with those obtained by Belanger *et al.* (2002) who found that silicon amendments to wheat plants mediate resistance in more complex ways than the original. The production of phenolic materials in powdery mildew-infected wheat leaves of Si, suggesting that these phenolics were likely in a fungitoxic form. These results strongly suggest that silicon mediates active localized cell defenses against *Blumeria graminis* f.sp. *tritici*. In addition, Rodrigues *et al.* (2003) found that the soluble Si in plant tissue might be associated with an increase in rice resistance to blast through the production of phenolic-like compounds, diterpenoid phytoalexins and the activation of some PR-genes. Further biochemical analysis identified some of these metabolites as flavonoids and phenolic acids that accumulated specifically and strongly in a manner typical of phytoalexin (Fawe, *et al.*, 1998). El-Lethy *et al.* (2011) state that foliar spray of geranium (*Pelargium graveolens* L.) with ascorbic acid, riboflavin and dry yeast, increased total phenolics and flavonoids which were known to possess significant antimicrobial activities and utilized as natural plant protectants

Data in Table 4 and Fig. 1 indicate that all foliar treatments with the resistance inducers recorded the lowest percentages of disease severity (DS) of powdery mildew on cucumber plants. Also, negative linear relationship between the reduction in DS of powdery mildew and PO and total phenolic content was found. Regression analysis showed that PO activity compounds contribute to about 56.3% ($R^2 = 56.3$, $P < 0.005$) and total phenolic content 77.2% ($R^2 = 77.2$, $P < 0.005$). These results were corroborated with the finding of Wei *et al.* (2004) who found that silicon supplements reversed the powdery mildew induced metabolic changes in cucumber seedlings, decreasing production of free radicals and increasing defense-related enzyme activity. Meantime,

Hassan *et al.* (2007) showed a positive relation between reduction of chocolate spot disease severity on faba bean plants and the increase in peroxidase activity and peroxidase isozymes as the results of application with chemical inducers.

Protein Extraction and Electrophoretic Analysis

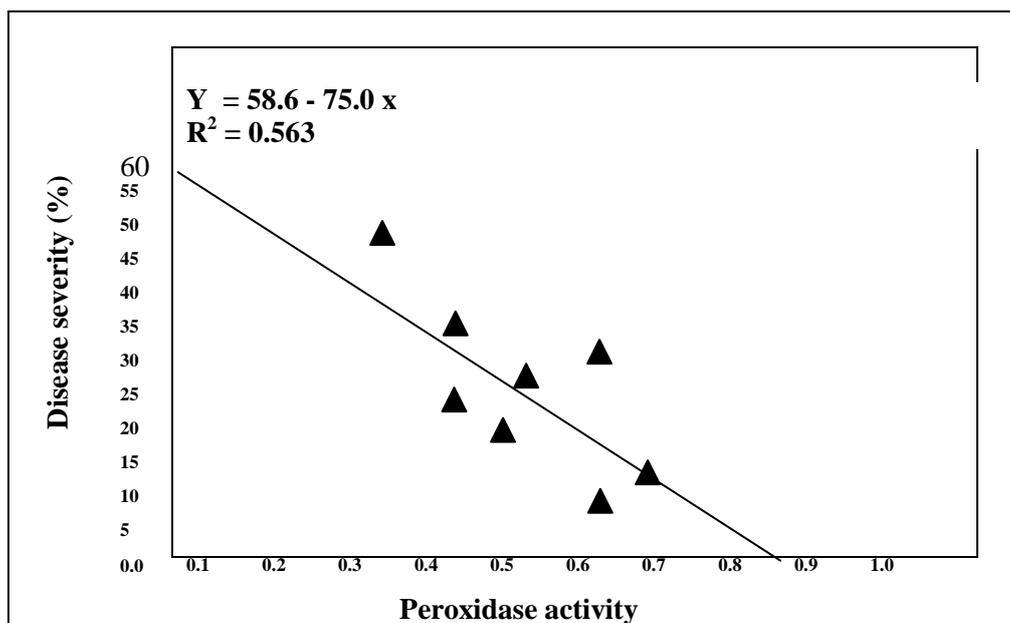
Results presented in Table 5 and Fig. 2, show the effect of potassium silicate (5ml/l), dry yeast (4000 mg/l), ascorbic acid (100mg/l), Topsin (0.7 g/l) on the induced of new proteins, which were not found in the untreated plants under natural infection by powdery mildew as shown by different bands separated by electrophoresis. New proteins of molecular weight (MW) 55.68, 42.55, 29.92, 16.03 and 14.00 KD were noticed in the plants treated with potassium silicate. Also, new proteins of molecular weight (MW) 46.75, 39.65, 33.21, 31.20, 23.13 and 13.27 KD were found in the plants treated with dry yeast. In addition, new proteins of molecular weight (MW) 40.75, 30.55, 27.28 and 21.13 KD were found in the plants treated with ascorbic acid. The following similar new protein of molecular weight (MW) 19.54 was found in the treated plants with dry yeast and ascorbic acid. Cucumber plants treated with potassium silicate, ascorbic acid and Topsin resulted in new protein of molecular weight (MW) 66.00 KD, which was not found in the untreated plants under natural infection. Furthermore, the new protein with molecular weight (MW) 15.78 KD was found in all the plants treated with biotic and abiotic inducers. These induced proteins have been defined as pathogenesis related proteins, where they implicated in plant defense for their anti pathogenic activities. Liang *et al.* (2005) found that the lower disease index and higher activity of pathogenesis –related proteins (PRs) of cucumber powdery mildew in the resistance cultivar was generally in time with silicon (Si) treatment. El-Gamal (2003) showed that increase in chitinase activity in the treated plants, was recorded when *S. cerevisiae* was applied on cucumber under the greenhouse conditions. Hassan *et al.* (2006) and Al-Sohaibani *et al.* (2011) concluded that the induction of plant resistance with chemical

Table 4. Correlation coefficient between percentage of disease severity and each of peroxidase activity and total phenolic content

Variable	Regression equation	Correlation coefficient (r)**	F. value
Peroxidase activity X_1	$Y = 58.6 - 75.0 X_1$	-0.750*	37.03***
Total phenolic content X_2	$Y = 54.5 - 68.4 X_2$	-0.879***	91.03***
X_1+X_2	$Y = 60.2 - 26.2 X_1 - 54.0 X_2$	-0.886**	53.12***

All correlations are significant at the $p < 0.005$ (***)

(A)



(B)

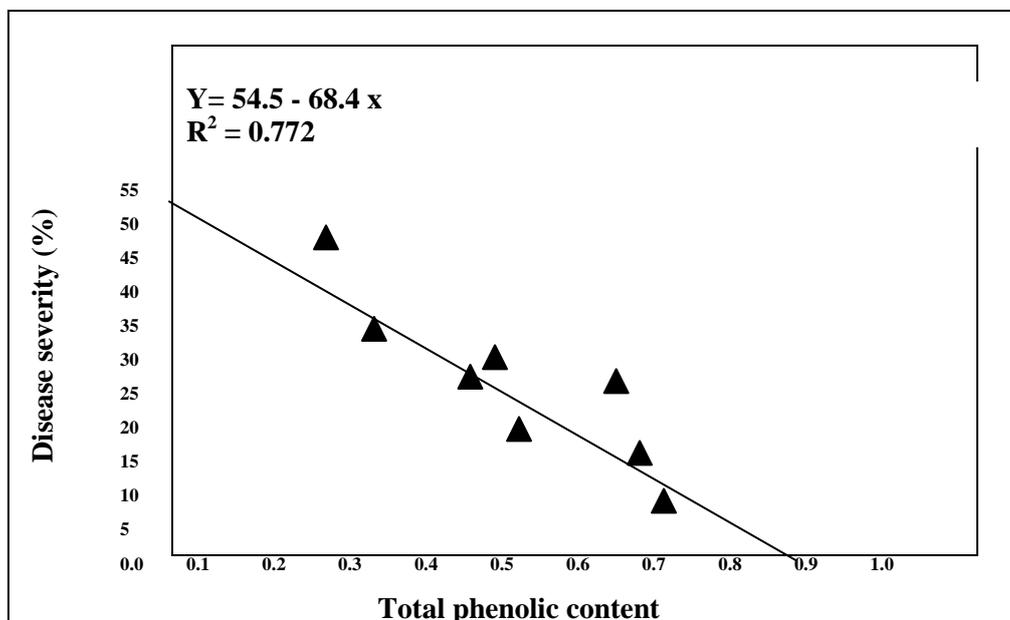


Fig. 1. Relationship between disease severity of powdery mildew and each of peroxidase activity (A) and total phenolic content (B) as mg/g of cucumber leaves sprayed with potassium silicate, ascorbic acid, dry yeast and Topsin in the 2nd season of 2014

Table 5. Proteins in cucumber plants treated with potassium silicate, dry yeast, ascorbic acid and Topsin

S/N	MW K.D	CO.	Si	DY	AA	F
1	69.24	+	-	-	-	-
2	67.62	+	-	+	-	-
3	66.00	-	+	-	+	+
4	65.85	+	-	+	-	-
5	57.84	-	-	-	-	+
6	55.68	-	+	-	-	-
7	54.51	+	-	+	+	+
8	46.75	-	-	+	-	-
9	42.55	-	+	-	-	-
10	40.75	-	-	-	+	-
11	39.65	-	-	+	-	-
12	33.21	-	-	+	-	-
13	31.20	-	-	+	-	-
14	30.55	-	-	-	+	-
15	29.92	-	+	-	-	-
16	27.28	-	-	-	+	-
17	26.73	+	+	-	+	-
18	25.95	-	-	-	-	+
19	24.48	+	-	-	-	+
20	23.13	-	-	+	-	-
21	22.92	+	+	-	-	-
22	21.13	-	-	-	+	-
23	20.94	+	+	-	-	+
24	19.54	-	-	+	+	-
25	16.03	-	+	-	-	-
26	15.78	-	+	+	+	+
27	14.00	-	+	-	-	-
28	13.27	-	-	+	-	-
29	12.00	+	-	-	-	-
30	0.83	+	-	-	-	-

S/N = Serial number; MW= Molecular weight; + = Present protein; - = Absent protein; Co. = Control without any treatment; Si = potassium silicate (5.0 ml/l); DY = Dry yeast (4000 mg/l); AA = Ascorbic acid (100 mg/l) and F = Topsin

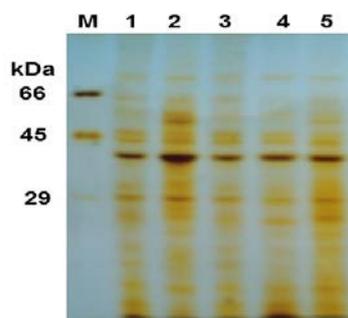


Fig. 2. Electrophoretic proteins profile of cucumber plants. M= Marker (Molecular weight 66, 45 and 29 Kd); 1= Potassium silicate (5m/l), 2 = Dry yeast (4000mg/l), 3 = Ascorbic acid (100mg/l), 4 = Topsin and 5= Control

inducers was associated with accumulation of pathogenesis-related proteins. Khan *et al.* (2011) found that ascorbic acid regulated stress response as a result of a complex sequence of biochemical reaction such as activation or suppression of key enzymatic reactions, induction of stress responsive proteins synthesis, and the production of various chemical defense compounds.

Effect of Foliar Spray with Resistance Inducers on the Content of N, P and K mg/g (dry weight) in Cucumber Leaves

Results in Table 6 show that all the tested compounds recorded high content of N, P and K in cucumber shoots, especially at the higher concentrations. The highest nitrogen content was obtained due to using dry yeast (4000 and 2000 mg/l) followed by ascorbic acid (100 mg/l). These results are in harmony with the findings of El-Tohamy *et al.* (2008) who indicated that the foliar application with yeast and ascorbic acid (vitamin C) to eggplant (*Solanum melongena* L.) plants resulted in a significant increase of nitrogen, phosphorus and potassium. The improvement of plant growth in response to the foliar application with active dry yeast might be attributed to its different nutrient, higher percentage values of vitamins which played an improving growth and controlling of

fungal diseases. The increment in N concentration due to ascorbic acid treatments could be explained by the finding of Talaat (2003) who indicated that accumulation of nitrate by ascorbic acid foliar application might be due to the positive effect of ascorbic acid on root growth, which consequently increased nitrate absorption. Moreover, increasing of nitrogen in plant stimulates synthesis of phenol and lignin contents, which are considered defense system of plant against infection (Marschner, 1995). The maximum of phosphorus (0.725 and 0.638) and potassium (1.863 and 1.785) were recorded from plants treated with dry yeast (4000mg/l) and potassium silicate (5ml/l), respectively. Under P-deficiency internal accessibility of P is controlled by other metals, such as Mn and Fe. Therefore, Si can increase P accessibility indirectly by decreasing the availability of Fe and Mn in the plants (Ma, 2004).

Conclusion

The obtained results suggested that beside the ability of the previous agents to induce systemic resistance in cucumber plants against powdery mildew disease they could be used as protective treatments. In addition, they are easy to employ, environmentally safe and can be used through the integrated management programs.

Table 6. Effect of foliar spray with potassium silicate, ascorbic acid, dry yeast and Topsin on the content of N, P and K (mg/g) of shoots

Treatment	Concentration	N (%)	P (%)	K (%)
2 nd season				
Potassium silicate	(ml/l)	2.5	0.638	1.785
		5.0	0.544	1.530
Ascorbic acid	(mg/l)	50	0.395	1.428
		100	0.474	1.698
Dry yeast	(mg/l)	2000	0.453	1.668
		4000	0.725	1.863
Topsin	(g/l)	0.7	.0408	1.378
Control		0.0	0.344	1.110
LSD at 5%			0.053	0.108

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إستحثات المقاومة الجهازية فى نباتات الخيار ضد مرض البياض الدقيقى تحت الظروف الحقلية

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يصاب نبات الخيار بالعديد من الأمراض الفطرية وعلى وجه الخصوص البياض الدقيقى المتسبب عن الفطر *Erysiphe cichoracearum* و *Sphaerotheca fuliginea*، أجريت هذه الدراسة بغرض تقييم بعض المستحثات وهى سليكات البوتاسيوم ومستخلص الخميرة وحمض الأسكوربيك وذلك لبيان تأثير هذه المعاملات على شدة الإصابة بمرض البياض الدقيقى على نباتات الخيار صنف بيت ألفا تحت الظروف الحقلية، أدى رش نباتات الخيار بالمبيد الفطرى توبسين ٧٠% (٠.٧ جم/لتر) كمعاملة مقارنة و بالمواد المستحثة وهى سليكات البوتاسيوم (٥ مللى/لتر) ومستخلص الخميرة (٤٠٠٠ ملليجرام/لتر) وحمض لأسكوربيك (١٠٠ ملليجرام/لتر) إلى انخفاض ملحوظ فى شدة الإصابة بالبياض الدقيقى بنسبة ٨٥.٩٩ و ٦٨.٣١ و ٥٠.٨٦ و ٤٢.٠٣%، على التوالى وذلك مقارنة بالنباتات غير المعاملة فى الموسم الأول ٢٠١٣، وقد أوضحت النتائج بالمعاملات السابقة زيادة معنوية فى النمو الخضرى المتمثلة فى (طول النبات، عدد الأوراق، الوزن الجاف، عدد الأزهار، عدد الثمار ووزن الثمار لكل نبات) كما أظهرت نتائج الموسم الثانى ٢٠١٤ نتائج مقارنة للمتحصل عليها فى الموسم الأول من حيث تقليل شدة الإصابة بالبياض الدقيقى بنسبة ٨٦.٠٢ و ٧١.٣٣ و ٤٨.٠٦ و ٤٥.٥٩%، على التوالى، أوضحت نتائج البحث المتحصل عليها زيادة فى النشاط الفسيولوجى المتمثل فى نشاط الإنزيم بيروكسيداز وكمية الفينولات الكلية تحت ظروف العدوى الطبيعية وكانت هناك علاقة عكسية مع شدة الإصابة بالبياض الدقيقى والمتمثلة إحصائيا بمعامل الارتباط R^2 بنسبة ٥٦.٣% لإنزيم بيروكسيداز و ٧٧.٢% للفينولات الكلية، أوضحت نتائج أيضا البحث زيادة فى تركيز عناصر النتروجين والفسفور والبوتاسيوم فى المجموع الخضرى، كما أدى استخدام المستحثات إلى استحداث تكوينات بروتينية جديدة فى النباتات المعاملة، أظهرت نتائج هذه الدراسة إلى إمكانية استخدام بعض المستحثات الكيماوية والحيوية كطريقة وقائية ضد مرض البياض الدقيقى حيث تتميز هذه المستحثات بكونها رخيصة الثمن وأمنة للبيئة وسهلة الإستخدام مما يؤهلها للإدراج ضمن برنامج مكافحة المتكاملة لمرض البياض الدقيقى فى الخيار.

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