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Effect of bacterial isolates and phosphite compounds on disease incidence of late blight (*Phytophthora infestans*) and improve productivity of some potato cultivars

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Abstract: Late blight of potato is caused by *Phytophthora infestans* and led to economic loss in large areas of potato production. In green house and field experiments, we used Four bacterial strains as biocontrol agents (*Brevibacillus brevis, Pseudomonas putida, Pseudomonas aerogenosa, Bacillus subtilis*), potassium phosphite (kPhi), copper as well as difenoconazole (Score) for disease control. The highest reduction in disease was achieved by applying difenoconazole where led to high reduction of diseases incidence compared to control, followed by potassium phosphite and *Bacillus subtilis* by 26.6% reduction with significantly increasing of potato yield compared to other treatments. Regarding to the response of cultivars to biological and chemical treatments compared with the non-treated control. Results clarified that cultivars used differed in their susceptibility to *Phytophthora infestans* under both green house and natural infection. Burren cv. was the less susceptible followed with cv. Spunta. Control treatment gave the lowest yield due to high late blight incidence. The coefficients of determination and regression coefficients for yield characters were analyzed, highly significant increasing effect on potato yield was obtained according to decreasing of disease incidence and severity. It is recommended that using of *Bacillus subtilis* as biocontrol agents and with integrated management program with potassium and pesticide may be useful in controlling potato late blight disease under field conditions.

Keywords: Late blight, biological control, potassium phosphite, potato cultivars, fungicides, disease severity, potato production.

1. Introduction

Potato (*Solanum tuberosum* L.) is the number one of non-grain food crop worldwide and the third most consumed food crop in the world, after wheat and rice **[1].** Egypt is on of top twenty five potato producing countries with a total production 4.5 M. ton in 2012 **[2]; [3]**. Among all the crops grown worldwide, potato is known to suffer the greatest losses from disease attack **[4].** Late blight of potato, which is caused by *Phytophthora infestans* (Mont) de Bary is still the most destructive of all potato disease worldwide **[5]; [6]**. This disease is known as the most devastating disease of potatoes where the losses of potato produced due to late blight are more than five billion USD annually **[7].** Late blight can completely destroy all parts of plants. When environmental conditions are appropriate for the pathogen there are no procedures taken to stop the progression of the disease **[8]; [9].** Late blight of potato has seriously infected potato and rapidly destroys potato in the field and after harvest. In endemic areas of this disease is difficult to combat it, but because of its epidemiology and life cycle with continually development of new strain of pathogens the single management strategy almost be not effective. Therefore, a number of management techniques have been developed and used. Integrated disease management (IDM) are most effective, economically, and environmentally safe **[10];[11]; [12]** where many approaches implemented such as chemical and biological control, use of resistant cultivars and fertilizing applications. IDM has helped to achieved increasing production and reduce the need of chemical controls. Fungicide is not effective to control of late blight if used individually but must be used as one tool in an integrated management approaches **[13]; [14]**. Economical and effective management of late blight was resulted by integrating resistant varieties of potato with the lowest rate of pesticide application **[15]; [12]**. Biological control of this disease has attracted the attention of researchers in last years and still need more concern because *Phytophthora infestans* is highly variable therefore contributed little to controlling late blight **[16]**. Biological control of plant diseases to reduce the pollution to environment is more important. Use of biocontrol agents against *Phytophthora infestans* in potato has more attention than in other crops **[17].**

Various bacterial species have shown great potential for control of several plant pathogens. Although biological control is not commonly used for foliar diseases, numerous organisms capable of antagonizing fruit and leaf pathogens have been reported in recent years. Significant studies on biological control of potato late blight focus on evaluate the efficacy of local isolates of microorganisms and aims to provide the alternative tools to reduce application of synthesis chemical pesticides to control this disease. The purpose of this work is to study the efficacy of some chemical, metal salts of potassium phosphite and cupper as mineral components, as well as some bacterial strains and one commercial pesticide to control of *P. infestans*.

2. Materials and Methods

Isolation, identification of pathogen

During winter growing seasons of 2013 and 2014, infected potato fruits and leaves were collected from Baloza experimental station, Desert research center, North Sainai governorate, Egypt. Infected leaf portions were placed in petri dishes with moist filter paper and incubated in darkness at 18 °C for 2 days. Once sporulation was evident, a small piece of DNA medium was wiped over sporangia to pick up few sporangia and directly transferred onto DNA plates. The plates were incubated at 25 °C for 7 days and examined for *P. infestans* colonies. Isolates were confirmed to be pathogenic using Koch's Postulate as preliminary test on potato plants under greenhouse conditions **[18]**.

Bacterial strains tested as a biocontrol agents:

Four bacterial isolates which used in this study were identified and provided by Dr. Amal M. Omar, Soil Microbiology Unit, Department of Soil Fertility and Microbiology, Desert Research Center. The preliminary identification was carried out according to Bergey's Manual of Determinative Bacteriology **[19]**, and molecular identification was carried out based on partial 16S rRNA gene sequence technique according to **[20]** in Sigma Scientific Services Co. Bacterial 16S rRNA gene sequences were amplified by PCR using the eubacterial primer pair 27f (5'-AGA GTT TGA TCC TGG CTC AG-3') and 1492r (5'-TAC GGY TAC CTT GTT ACG ACT T-3') according to **[21]**.The bacterial strains which used in this study are *Brevibacillus brevis* (Bbr), *Pseudomonas putida* (Ppu), *Pseudomonas aerogenosa* (Pae), *Bacillus subtilis* (Bsu).

Preparation of Biocontrol agents:-

For determine the ability of bacterial isolates to control of *Phytophthora infestans* under greenhouse and field conditions, four bacterial isolates were selected for investigation. The bacterial isolates were obtained as fresh bacterial cells from cultures grown in nutrient broth medium under shaking conditions. 100 mL of nutrient broth was inoculated and incubated for 48h at 24-26 °C in a rotary shaker (100 round/min). The supernatant was discarded after the bacterial culture was centrifuged (10000 rpm for 10 min). The cell pellet was centrifuged again under the same conditions in sterile 0.85% NaCl. The bacterial cells were resuspended in sterile distilled water. The concentration of cells in the suspension was counted spectrophotometricaly and adjusted to 10^8CFU/mL and used for greenhouse and field pot experiments **[22]**.

Evaluation of bacterial isolates and chemical treatments to control late blight of potato in greenhouse

The experiment was set in the greenhouse of Plant protection department, Desert Research Centre, Egypt as a randomized complete block design (RCBD) with three replicates (Five plants/replicate) for each treatment. Tubers of four potato cultivars (Burren , Diamond, Picasso and Spunta) were used. Healthy potato seed tubers (locally produced) were planted in plastic pots (50 cm diameter) containing sandy loam soil at 22–25c and RH 75–80%. The treatments were added as follows:

- Inoculated of potato plants treated with water were used as control.

- 30 ml of fungus suspension were added to plants had 4–5 leaves then the pots kept covered under plastic sheet for two days.

- 90 ml of bacterial cells concentration (108CFU/ml.) for each isolate.

- while the concentration of Phosphites compound and fungicide were (2 ml/L, 2.5 gm/L and 0.5 ml / L) for Potafore, Oxy plus and Score respectively.

- Disease incidence and severity were rated based on percentage of damaged potato leaf area and affected number of plants.

Efficacy of biological and chemical treatments under field conditions

During winter growing seasons of 2013 and 2014 the field experiment was conducted at Baloza experimental station, Desert research center, North Sinai governorate, Egypt, This experiment was conducted in split plot design with three replicates, since potato cultivars assessed in main plot while foliar spry treatments were assessed in sub plot. Local potato seed tubers saved by the farmers from the previous summer harvest were planted at 1st of October in rows 1 m width, 30 cm apart under drip irrigation system in both seasons, and the plot area was 10.5 m^2. In both seasons, all cultural practices (irrigation, fertilization and weeding) were performed according to the recommendations of the Egyptian Ministry of Agriculture. Potassium phosphite (kPhi) , copper (Cu) , difenoconazole (Score) and four bacterial strains (Bbr, Ppu, Pae and Bsu) were used to evaluate their efficacy on late blight severity and incidence on four potato cultivars (Burren, Diamond, Picasso and Spunta) under natural infection conditions. All treatments were applied used as a foliar spray at 30 days after emergence (4 –5 true leaf stage) four times with 10-12 days intervals. The bacterial and chemical compound concentrations prepared and applied as described above.

Data recorded:

- Disease assessment

Late blight disease incidence and severity were recorded with the appearance of natural disease symptoms on control plants (after 60 days of planting). The average of records of the surveyed replicates for each particular treatment was calculated. Disease severity and incidence was estimated as follows; 0 = no leaf lesion; 1= lesions on < 25% of leaf area; 2 = lesion on 26–50% of leaf area; 3 = lesion on 51–75% of leaf area and 4 = lesions on 76 up to 100% of leaf area according to the scale from 0 to 4 suggested by **[23],** then the traits calculated using the following formula:

$$D.S = \frac{\sum (n \times c)}{N}$$

Where; D.S. = Intensity of attack, n = Number of infected plants per category, c = Category number and N = Total examined plants.

- Vegetative growth:

A random sample of five plants of each experimental plot was taken at 90 days after planting for vegetative growth data. Plant height, leaves number/plant, leaves chlorophyll content and plant fresh weight were recorded.

- Yield and its component:

120 days after planting all experimental plot area was harvested and tubers were counted, and weighed to record total yield, tuber number and average tuber weight.

Statistical analysis:

The recorded data was subjected to statistical analysis by M-STAT C **[24].** The differences among means were performed using least significant difference (LSD) at 5% level.

3. Results and Discussion

Efficacy of bacterial isolates and chemical agents against late blight disease in potato plants under greenhouse conditions:

The biocontrol potential of bacterial isolates Bbr, Ppu, Pae and Bsu as well as the antifungal activity of Potassium phosphite (kPhi), copper (Cu), difenoconazole (Score) against potato late blight pathogen, *Phytophthora infestans,* were studied under greenhouse conditions. Data showed in (Table 1) regarding the percent disease incidence due to different treatments confirmed that minimum disease was recorded (40 %, 46.7%, 53.3% and 46.7%) in cultivars Burren, Diamond,Picasso and Spunta respectively, while the maximum 53.3% , 80%, 73.3%, 70% was recorded in Burren, Diamond, Picasso and Spunta cultivars respectively, Accordingly, the most effective treatment was pesticide when it led to reduction of diseases incidence by 36.3% compared to control, as mean of records in tested cultivars flowed by Bsu bacterial strain and potassium phosphite (kPhi) by 26.6%. regarding to the response of cultivars to biological and chemical treatments, highly significant differences were observed among the cultivars. The best response to treatment was in Burren cultivar with all treatments, while the less response was in case of Diamond. The response of all the other cultivars to treatments was among the fungicide followed by Bsu and least by Bbr isolate. Also the disease incidence induction rates were compatible with disease severity results where the most effective treatment was fungicide with records (2.3, 2.7, 3, 2.7) on Burren, Diamond, Picasso and Spunta respectively with significant reduction compared to control. This treatment followed by Bsu isolates where minimum disease index was recorded 2.3 while there are no differences between other cultivars in their response to Bsu isolaes where recorded index value (3) with significant reduction as compared to control. Generally, late blight of potato rapidly infected plants, spreading to all parts of potato plants and destroying the plants within 3-4 weeks in control as compared to all treatments,where the treated plants were healthier. According to these results, Burren cv. was less susceptible to infection of *phytophthora infestans* with records (73.3 and 3.0) disease incidence and severity respectively. Picasso cv. Show high susceptibility (93.3% and 4) disease incidence and severity. All treatments decreased disease incidence and severity compared to control. Also kPhi and Oxy plus caused best results in decreasing of disease incidence and severity for *P. infestans*. These results agreed with some previous investigations where Phosphite (Phi) has been recommended to enhance plant resistance against *Phytophthora spp*. **[25]; [26]; [27]**, and *B. subtilis* Bsu also recommended to control of *P. infestans* **[28]; [29]; [30]; [31].** These results suggests that we need to continue studying the effective treatments which used in this experiment for late blight disease control in potato to conceder these compounds as control agents of *P. infestans.*

Evaluation of chemical and bacterial agents to control late blight of potato under field conditions:

The field experiments were carried out at Baloza experimental station, Desert research center, North Sinai governorate, Egypt, during winter growing seasons of 2013 and 2014. kPhi, Cu, Score and four bacterial strains Bbr, Ppu, Pae and Bsu were used to evaluate their antifungal effects on late blight disease on four potato cvs., Burren, Diamond, Picasso and Spunta under natural infection conditions. Data revealed that all cultivars were susceptible to late blight but they significantly difference in their susceptibility according to their disease incidence (Table 2) and disease severity (Table 3) recorded during the two successive seasons. Burren cultivar exhibit less susceptibility to late blight according to disease incidence and severity compared to control treatment in both seasons (21.7% and 29.2%) for disease incidence and (2.7 and 3.3) for disease severity with significant difference compared to other cultivars. On the other hand, all treatments revealed significant results with Burren cultivar in both seasons. Spunta cultivar was moderate susceptible in both seasons (30.3% and 37.0%) (2.7 and 3.3) for both disease incidence and severity in first and second seasons respectively. Cultivars Dimond and Picasso showed more susceptibility to late blight in both seasons compared with other cultivars.

Table (1): Evaluation of biological and chemical control agents against *Phytophthora infestans* under greenhouse conditions.

Treatments	Burren		Diamond		Picasso		Spunta		X¯	
	D I	D S	D I	D S	D I	D S	D I	D S	D I	D S
Bbr	53.3	3.7	73.3	4.0	80.0	3.7	73.3	3.3	70.0	3.7
Ppu	53.3	3.7	73.3	3.7	73.3	3.3	73.3	3.3	68.3	3.5
Pae	53.3	3.0	73.3	3.7	66.7	3.7	66.7	3.7	65.0	3.5
Bsu	46.7	2.3	66.7	3.0	53.3	3.0	60.0	3.0	56.7	2.8
KPhi	53.3	3.3	53.3	2.7	60.0	3.3	60.0	3.7	56.7	3.3
Cu	53.3	3.0	60.0	3.0	60.0	3.3	60.0	3.3	58.3	3.2
Fungicide	40.0	2.3	46.7	2.7	53.3	3.0	46.7	2.7	46.7	2.7
Control	73.3	3.0	86.7	4.0	93.3	4.0	80.0	3.7	83.3	3.7
X¯	53.3	3.0	66.7	3.3	67.5	3.4	65.0	3.3		

LSD at 5%			
	Var.	Treatments	Interaction
D I	5.3	7.6	15.3
D S	0.2	0.4	0.8

Table (2): Effect of cultivars and foliar spray treatments and their interactions on potato disease incidence during two successive growing seasons (2014 and 2015).

Treatments	**Burren**		**Diamond**		**Picasso**		**Spunta**		X¯	
	1St S*	2nd S**	1St S	2nd S	1St S	2nd S	1St S	2nd S	1St S	2nd S
Bbr	23.7	31.5	40.3	51.8	47.3	61.5	26.0	34.2	34.3	44.7
Ppu	23.3	30.9	36.7	48.3	43.3	56.3	23.7	31.7	31.8	41.8
Pae	19.7	27.1	40.0	52.7	42.0	55.3	24.3	32.3	31.5	41.9
Bsu	13.7	18.5	33.7	45.7	35.0	46.6	21.0	28.6	25.8	34.8
KPhi	13.0	17.9	34.0	44.3	36.7	44.0	20.0	26.6	25.9	33.2
Cu	12.0	16.9	38.0	50.3	36.7	49.0	21.3	32.0	27.0	37.1
Fungicide	6.3	8.9	23.3	33.5	20.7	27.9	14.0	18.8	16.1	22.3
Control	21.7	29.2	37.0	48.9	46.7	59.6	30.3	37.0	33.9	43.7
X¯	16.7	22.6	35.4	47.0	38.6	50.0	22.6	30.1		

LSD at 5%			
	Var.	Treatments	Interaction
1St S	**1.6**	1.2	2.4
2nd S	1.2	2.1	4.3

*, ** Means the first (1st S) and second (2nd S) growing seasons; KPhi = Potassium phosphite; Cu = Cupper

Picasso was the more susceptible cultivar in both seasons (46.7% and 59.6%) (4 and 4) as disease incidence and severity, while all treatments revealed lowest reduction in disease incidence and severity with cv. Picasso in both seasons and the significant differences resulted compared with other cultivars. All treatments revealed significant differences. Bacterial isolates *Brevibacillus brevis* (Bbr) *Pseudomonas putida* (Ppu), *Pseudomonas aerogenosa* (Pae) and *Bacillus subtilis* (Bsu) gave significant results if compared with control but not with fungicide, Potassium and copper phosphite which gave significant reduction in disease incidence and severity than bacteria isolates. (Bsu) was more effective agent compared with all bacteria isolates through two seasons followed by (Pae) isolate. Isolats (Ppu) and (Bbr) exhibit the lowest reduction of disease incidence and severity with all cultivars through the two seasons.

Table (3): Effect of cultivars and foliar spray treatments and their interactions on potato diseases severity during two successive growing seasons (2014 and 2015).

Treatments	Burren		Diamond		Picasso		Spunta		$\bar{X}$	
	1St S*	2nd S**	1St S	2nd S	1St S	2nd S	1St S	2nd S	1St S	2nd S
Bbr	2.3	3.0	3.0	3.3	3.3	4.0	2.7	3.3	2.8	3.4
Ppu	2.3	3.0	3.3	3.7	3.7	4.0	2.7	3.3	3.0	3.5
Pae	2.7	3.3	3.7	4.0	3.7	4.0	3.0	3.3	3.3	3.7
Bsu	1.7	2.3	2.7	3.0	2.7	3.0	2.3	3.0	2.3	2.8
KPhi	1.7	2.0	2.3	2.7	2.3	3.0	2.3	3.0	2.2	2.7
Cu	2.0	2.7	2.7	3.0	2.7	3.3	2.0	3.0	2.3	3.0
Fungicide	1.0	1.3	1.3	1.7	2.0	2.7	1.0	1.7	1.3	1.8
Control	2.7	3.3	3.3	4.0	4.0	4.0	2.7	3.3	3.2	3.7
$\bar{X}$	2.0	2.6	2.8	3.2	3.0	3.5	2.3	3.0		

LSD at 5%			
	Var.	Treatments	Interaction
1St S	0.4	0.4	0.8
2nd S	0.4	0.3	0.7

*, ** Means the first (1st S) and second (2nd S) growing seasons. Late Blight Disease Severity Recorded according to the Scale 0-4;

Efficacy of chemical and biological agents against late blight of potato under natural infection:

Efficacy of treatments on vegetative growth:

The treatments were applied under field conditions during two successive growing seasons. Data presented in Table (4) showed that potato shoot length, shoot fresh weight, leaf number and leaf chlorophyll content significantly affected by potato cultivars and foliar spray treatments. Moreover, the effect of foliar spray on shoot length and all interactions effects were not significant in both seasons. Diamond cultivar produced the highest significant shoot length, shoot weight and leaf number compared with other cultivars, followed by cvs Burren then Spunta, while Picasso cultivar give the lowest significant values in both seasons. Burren cultivar give the highest leaf chlorophyll content in both seasons compared with other cultivars.

Concerning foliar spray treatments, potassium phosphite and score treatments give the highest shoot weight compared with other treatments. Leaf chlorophyll content increased significantly by potassium phosphite, score and cupper treatments compared with bacterial strains and control treatments in both seasons.

Efficacy of treatments on yield and its component:

Data in Table (5) showed that potato yield per plot, tuber number, average tuber weight and tuber dry matter content significantly affected by potato cultivars and foliar spray treatments, while all interaction effects were not significant in both seasons. Burren cultivar produced the highest significant potato yield per plot and average tuber weight compared with other cultivars. Also, Burren, Diamond and Picasso produced the highest tuber number compared with Spunta cultivar in both seasons. On the other hand, Burren showed the lowest dry matter percent compared with other cultivars especially Diamond cultivar which give the highest value in both seasons (Table 6).Regarding foliar spray treatments, highly significant increasing effect on potato yield was obtained with score treatment, followed by potassium phosphite then (Bsu) bacterial strain. While cupper, (Bbr), (Ppu), (Pae) strains and control treatments produced the lowest potato yield in both seasons. Moreover, tuber number per plot, average tuber weight and dry matter percent increased by Score treatment followed by potassium phosphie then (Bsu) bacterial strain compared with cupper, (Bbr), (Ppu), (Pae), and control treatments which give the lowest values in this respect. Lowest treatment in decreasing disease incidence and severity with all cultivars in both seasons are potassium phosphite (kPhi) and Oxy plus which exhibit superior results in significantly decreasing disease incidence and severity comparing with bacterial isolates and control but no differences between them these results agreed with **[32].** The highly decreasing in disease incidence and disease severity were obtained by treated potato plants with fungicide, where, gave highly significant differences with all treatments, control throw seasons and cultivars.

Table (4) Effect of cultivars and foliar spray treatments and their interactions on potato shoot length and fresh weight, leaves number and leaves chlorophyll content of two seasons.

Characters		Shoot length (cm)		Shoot fresh weight (g)		Leaves number		chlorophyll (SPAD)	
Cultivars	Seasons	1St S	2nd S	1St S	2nd S	1St S	2nd S	1St S	2nd S
Burren		75.87	73.62	228.06	208.17	16.56	17.69	42.66	39.53
Diamond		85.59	82.02	261.63	235.64	19.14	19.78	39.28	36.22
Picasso		53.14	40.66	122.44	94.99	10.63	10.63	39.94	37.04
Spunta		68.85	61.92	213.09	193.52	14.11	15.74	40.61	37.38
LSD at 0.05		**6.18**	**6.34**	**12.44**	**13.39**	**0.82**	**0.82**	**1.61**	**1.71**
Foliar spray									
Bbr		69.53	62.99	199.95	180.03	14.96	15.82	39.87	36.98
Ppu		70.33	64.05	199.96	178.88	14.93	16.04	39.36	37.22
Pae		69.22	63.17	197.90	175.09	14.71	15.74	39.17	36.28
Bsu		69.51	63.97	201.23	181.64	14.65	16.13	39.91	37.35
K (Phi)		76.02	68.32	226.75	199.64	16.08	16.86	42.98	40.08
Cu		72.17	64.97	209.33	185.31	15.24	15.77	42.00	39.52
Score		71.59	66.16	216.47	186.18	15.82	16.19	42.62	38.48
Control		68.53	62.81	198.88	177.87	14.49	15.12	39.06	36.42
LSD at 0.05		**NS**	**NS**	**16.58**	**14.18**	**NS**	**NS**	**2.00**	**1.43**
Interaction									
Burren	Bbr	73.43	71.85	220.80	203.05	16.28	16.95	41.88	39.41
	Ppu	74.30	72.44	217.52	209.01	16.55	17.25	41.50	39.03
	Pae	73.62	72.02	220.50	200.41	16.25	17.28	40.37	38.23
	Bsu	74.42	72.82	226.80	209.71	16.11	18.14	41.65	38.51
	K (Phi)	83.58	79.05	251.44	222.66	17.88	19.58	45.93	41.46
	Cu	75.33	73.74	229.50	208.39	16.33	17.70	43.91	40.11
	Score	78.78	76.84	232.15	208.75	16.83	17.56	44.38	40.24
	Control	73.50	70.23	225.80	203.38	16.28	17.01	41.67	39.21
Diamond	Bbr	84.22	79.96	254.10	232.35	18.72	20.08	38.95	36.48
	Ppu	85.40	81.47	257.10	230.26	18.30	20.34	37.89	35.75
	Pae	84.66	81.06	253.38	226.75	18.33	20.04	38.83	35.69
	Bsu	85.36	83.44	259.80	233.71	18.86	20.67	39.62	37.48
	K (Phi)	88.11	85.51	277.28	249.61	21.02	19.72	39.90	38.43
	Cu	86.71	82.51	263.10	238.33	19.13	18.85	39.93	33.46
	Score	86.57	81.09	274.50	241.72	20.42	19.45	41.69	37.55
	Control	83.70	81.10	253.80	232.38	18.36	19.06	37.38	34.90
Picasso	Bbr	52.40	39.50	113.10	93.01	10.64	10.64	39.25	35.11
	Ppu	52.18	39.94	116.40	89.44	10.91	10.91	39.27	37.13
	Pae	51.45	39.21	112.50	85.75	10.25	10.25	37.96	35.48
	Bsu	50.62	38.72	109.80	89.71	10.03	10.03	38.83	36.69
	K (Phi)	59.92	45.36	154.12	111.35	11.38	11.38	43.00	41.18
	Cu	55.70	41.47	129.90	103.15	11.03	11.03	41.97	38.13
	Score	53.40	42.50	133.27	97.18	11.43	11.43	40.94	37.13
	Control	49.48	38.58	110.40	90.30	9.41	9.41	38.30	35.48
Spunta	Bbr	68.08	60.65	211.80	191.71	14.23	15.59	39.39	36.91
	Ppu	69.43	62.34	208.80	186.81	13.98	15.68	38.77	36.96
	Pae	67.16	60.40	205.20	187.44	14.01	15.38	39.53	35.72
	Bsu	67.65	60.89	208.50	193.41	13.62	15.69	39.54	36.73
	K (Phi)	72.47	63.37	224.17	214.92	14.03	16.74	43.07	39.26
	Cu	70.93	62.17	214.80	191.38	14.48	15.51	42.18	38.37
	Score	67.63	64.20	225.96	197.08	14.62	16.32	43.49	39.01
	Control	67.43	61.33	205.50	185.42	13.91	14.99	38.91	36.10
LSD at 0.05		**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**

Table (5) Effect of cultivars and foliar spray treatments on potato yield (kg/plot), tubers number per plot, average tuber weight (g) and dry matter % in both of two growing seasons.

Cultivars	Potato yield (kg/polt)		Tubers number/plot		Average tuber weight (g)		Dry matter %	
	1St S*	2nd S**	1St S	2nd S	1St S	2nd S	1St S	2nd S
Burren	35.602	24.827	384.3	319.8	92.94	77.65	18.65	16.57
Diamond	29.996	20.891	384.7	331.8	77.79	62.90	21.94	19.22
Picasso	19.134	10.819	400.2	360.3	47.85	33.38	20.16	17.96
Spunta	22.025	14.335	255.3	202.6	85.87	70.18	20.30	17.83
LSD at0.05	3.488	2.137	45.7	31.6	2.65	3.64	1.07	0.61
Foliar spray								
Bbr	24.834	15.394	342.8	256.2	73.56	59.98	19.62	17.20
Ppu	24.452	15.264	339.9	258.1	73.41	59.31	19.98	17.60
Pae	25.328	15.805	349.1	276.6	73.82	58.08	20.16	17.63
Bsu	27.058	17.783	367.9	307.4	75.30	60.03	19.98	17.54
K (Phi)	29.328	20.555	367.6	320.9	80.90	65.22	20.50	18.58
Cu	25.317	16.878	347.2	293.4	74.28	59.01	20.25	17.80
Score	33.079	24.222	398.3	362.0	84.41	68.73	21.39	19.22
Control	24.118	15.844	335.6	274.6	72.79	57.88	20.22	17.58
LSD at 0.05	2.467	1.910	34.8	35.9	4.01	3.63	0.70	0.57

*, ** Means the first (1St S) and second (2nd S) growing seasons; k(Phi) = Potassium phosphite; Cu = Cupper

Determination of the relationship between late blight disease control and yield components of the potato cultivars:

It worth to notice that potato plant fresh weight (g/plant), average tuber weight (g) and potato tuber yield (kg/plot) were highly negative correlated with late blight disease incidence in both seasons Fig (1). A linear correlation coefficients (r) were (-0.401,-0.399), (-0.796,-0.779) and (-0.671,-0.628) for potato plant fresh weight, average tuber weight and tuber yield in the first and second seasons, respectively. The corresponding coefficients of determination (r^2) were (0.161,0.159), (0.603,0.607) and (0.381,0.394) which indicated that (16.1,15.9), (60.3,60.7) and 38.1,39.4) of the variation in potato plant fresh weight, average tuber weight and tuber yield were related to late blight disease incidence percent in the first and second seasons, respectively. Regarding of regression coefficients (-1.95,-1.57), (-1.25,-0.98) and (-0.411,-0.291) were obtained for plant fresh weight, average tuber weight and tuber yield in the first and second seasons, respectively. This indicated that when late blight disease incidence increased by one percent, plant fresh weight (g), average tuber weight (g) and tuber yield (kg/plot) decreased by (1.95g, 1.57g), (1.25g, 0.98g) and (0.411kg, 0.291kg) in the first and second seasons, respectively.

Table (6): The interactions between cultivars and foliar spray treatments on potato yield (kg/plot), tubers number per plot and average tuber weight (g) of two seasons

Cultivars/ treatments	Characters/ seasons	Potato yield (kg/polt)		Tubers number/plot		Average tuber weight (g)		Dry matter %	
		1St S*	2nd S**	1St S	2nd S	1St S	2nd S	1St S	2nd S
Burren	Bbr	33.443	23.253	368.3	297.0	90.84	78.49	17.89	15.58
	Ppu	33.077	23.220	367.0	303.0	90.63	76.62	18.44	16.45
	Pae	35.243	23.720	389.1	316.3	91.04	75.12	18.80	16.55
	Bsu	35.243	23.713	381.6	308.9	92.27	76.58	18.26	16.50
	K (Phi)	38.410	27.553	406.3	348.8	94.53	78.85	18.68	16.95
	Cu	35.910	24.387	393.7	324.8	91.50	75.82	18.42	16.42
	Score	40.743	29.217	403.8	342.5	100.96	85.28	20.18	18.30
	Control	32.743	23.553	364.3	317.4	90.11	74.43	18.53	15.78
Diamond	Bbr	29.743	19.887	398.2	323.6	75.18	62.83	20.33	18.38
	Ppu	29.580	19.397	391.2	306.1	75.23	62.88	21.56	18.99
	Pae	28.750	19.227	379.8	321.9	75.64	59.96	21.30	18.68
	Bsu	29.743	19.887	381.6	319.7	77.87	62.18	20.85	17.90
	K (Phi)	31.580	22.057	381.7	329.4	82.80	67.12	23.13	20.33
	Cu	27.743	19.557	361.0	323.7	76.10	60.42	22.74	19.67
	Score	36.587	29.063	432.0	420.9	84.90	69.21	23.66	20.83
	Control	26.243	18.053	351.9	308.8	74.58	58.63	21.97	18.97
Picasso	Bbr	16.410	7.553	367.7	242.8	45.09	31.17	20.02	17.22
	Ppu	16.577	8.057	377.5	268.7	44.88	30.53	19.53	17.16
	Pae	17.577	8.720	390.8	295.9	45.29	29.61	20.56	17.99
	Bsu	20.410	12.220	438.2	378.3	46.52	32.50	20.78	18.23
	K (Phi)	21.910	13.720	406.2	357.4	54.12	38.44	20.50	18.90
	Cu	18.703	10.513	408.9	332.1	45.75	31.74	19.45	17.35
	Score	25.077	16.887	441.6	410.6	56.88	41.20	20.99	19.50
	Control	16.410	8.883	370.2	276.9	44.23	31.88	19.49	17.37
Spunta	Bbr	19.740	10.883	237.0	161.3	83.11	67.43	20.24	17.63
	Ppu	18.573	10.383	224.1	154.7	82.90	67.22	20.38	17.79
	Pae	19.743	11.553	236.9	172.2	83.31	67.63	19.98	17.31
	Bsu	22.833	15.313	270.5	222.5	84.54	68.85	20.05	17.53
	K (Phi)	25.413	18.890	276.5	247.9	92.14	76.46	19.71	18.16
	Cu	18.910	13.053	225.4	193.0	83.77	68.09	20.41	17.76
	Score	29.910	21.720	316.1	274.1	94.90	79.22	20.73	18.26
	Control	21.077	12.887	256.0	195.1	82.25	66.57	20.89	18.22
LSD at 0.05		NS	NS	NS	NS	NS	NS	NS	NS

Concerning of the same characters via late blight disease severity, highly negative correlation were found fig (2). Where, a linear correlation coefficients (r) were (-0.386, -0.372), (-0.629,-0.582) and (-0.607,- 0.699) in the first and second seasons, respectively. Moreover, the corresponding coefficients of determination (r^2) were (0.149, 0.138), (0.395, 0.339) and (0.368, 0.489) for plant fresh weight, average tuber weight and tuber yield, which indicated that (14.9, 13.8), (39.5, 33.9) and (36.8, 48.9) of the variation related to late blight disease severity in the first and second seasons, respectively. Regarding regression coefficients (-27.47, -29.03), (-14.98, -14.59) and (-5.90, -6.41) were obtained in the first and second seasons. This indicated that when late blight disease severity increased by one grade on the scale, plant fresh weight, average tuber weight and tuber yield decreased by (27.47g, 14.98g), (14.98g, 14.59g) and (5.90kg, 6.41kg) in the first and second seasons, respectively. Such decrement in potato yield per plot due mainly to average tuber weight which, in turn, highly decreased when plants especially leaves (assimilative organs) significantly affected by late blight disease. Similar results were reported by **[33]; [34]; [35]; [36]; [37]**. Also, **[38] and [39]** reported that primary damage by late blight is attributed to premature defoliation of the potato plants, resulting in tuber yield reduction in most potato growing regions of the world. Results clarified that cultivars used differed in there susceptibility to *P. infestans* under both green house and natural infection Burren cv. was the less susceptible fowled with cv. Spunta. Bacterial isolates *Bacillus subtilis* and *Pseudomonas aerogenosa* respectively had significant differences in reducing disease incidence and severity than control. *Bacillus* able to suppressing disease of *P. infestans* and enhancing different growth parameters **[40]** also *Pseudomonas aeruginosa* had the ability to reduce disease severity of *Phytophthora infestans* when used as bacterial cell based formulation, culture supernatant and bacterial cell suspension and it holds promise as biological control agent against *P. inefstans* in field **[41]**. As integrated management for *P. inefstans* using resistant cultivars, biocontrol agent, and eco-friendly compound are good in reducing benefits and pollution. But in case the pathogen infects the crop, the epidemic must be stopped by using more powerful formulations. **[42]** revealed that chemical control remains the most important control measure against late blight. Growing potatoes without using fungicides has become unthinkable in most parts of the world. The control strategy is primarily preventive but in case the pathogen infects the crop, the epidemic must be stopped by using more powerful formulations. “Metalaxyl” in the trade name of Ridomil Gold was most effective in reducing Late blight disease severity and concluded that host resistance when integrated with fungicide application enhanced the control of late blight and reduced losses

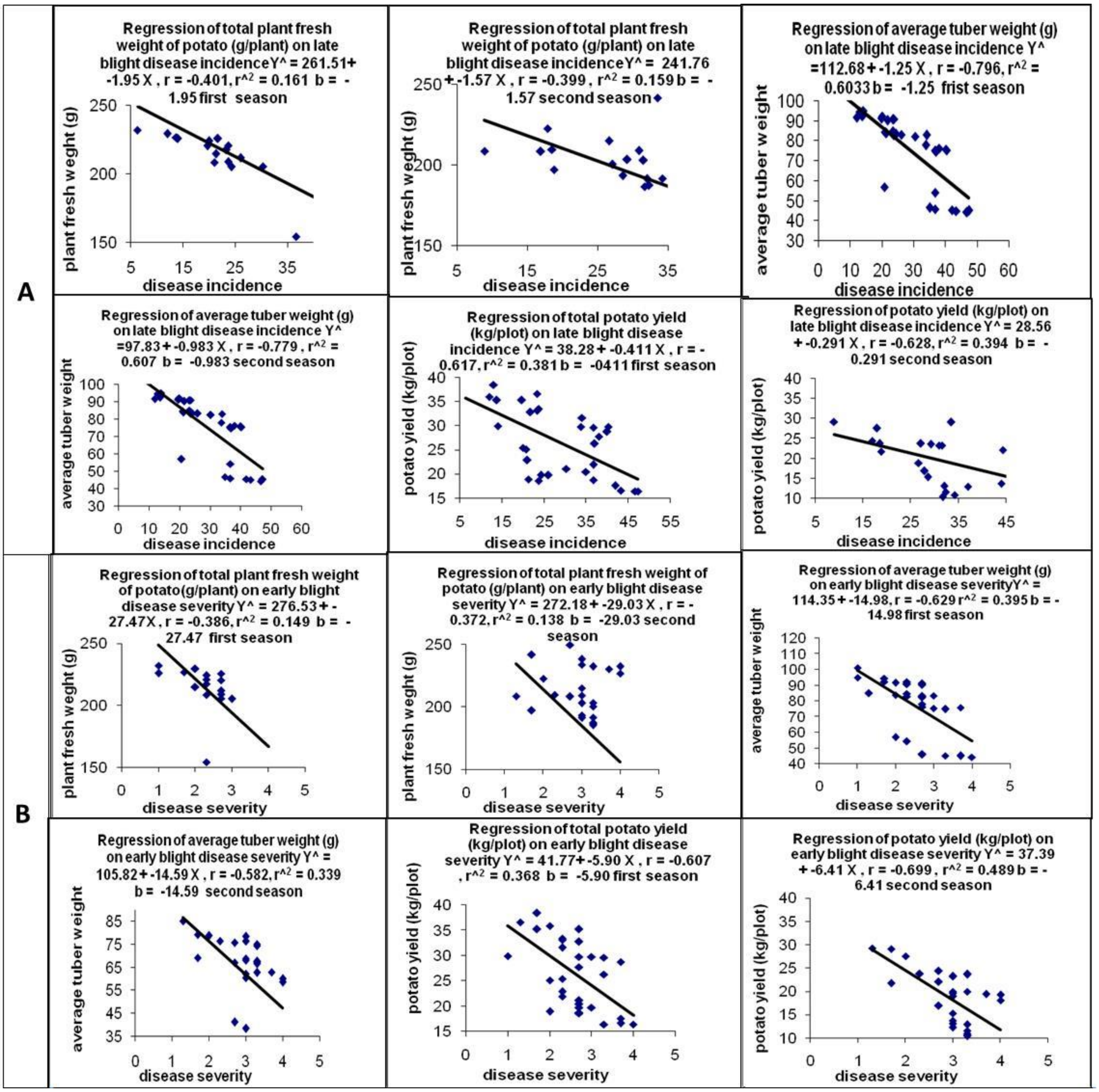

Fig.(2) regression lines, coefficients of determination (r^2) and regression coefficients (b) for plant fresh weight (g/plant), total yield(kg/plot) and average tuber weight (g) of potato vs. late blight (A) disease incidence % and (B) disease severity for 2014 and 2015 seasons.

4. Conclusion

To the best of our knowledge, the current study presents the data on response of four potato cultivars to bacterial isolates (*Brevibacillus brevis, Pseudomonas putida* , *Pseudomonas aerogenosa, Bacillus subtilis*) and chemical components (Potassium phosphite, copper, difenoconazole) under green house and field condition in Egypt. Furthermore, the outcomes of this study to determine the best treatments could be considered as alternatives to pesticides to use it in late blight disease control. Also the obtained data helps to determine the cultivars could be grown under studied condition and their response to studied components. The variation of potato cultivars response to control agents which used helps to determine the most effective agents and the percentage of disease incidence and severity reduction and its impacts on potato yield. Regarding foliar spray treatments, highly significant increasing effect on potato yield was obtained with Score treatment, followed by potassium phosphite then (Bsu) bacterial strain with the correlation between disease control and increasing of yield. It could be suggested that the most effective bacterial isolates which could be partially exploited as alternative fungicides and might be used for controlling late blight disease of potato plants under field conditions.

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