

BIOCHEMICAL RESPONSE IN POTATO CULTIVARS TO INFECT WITH *Rhizoctonia solani* THE CAUSAL OF STEM CANKER AND BLACK SCURF DISEASE IN EGYPT.

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ABSTRACT

Potato cultivars differ in their susceptibility to stem canker and black scurf disease caused by *Rhizoctonia solani*. Cultivar reactions of the most nine common commercial cultivars of potato were tested under greenhouse conditions of plant pathology research Institute, ARC-Giza. The nine tested cultivars were divided to three groups . Lady Rosetta was the most susceptible cultivar followed by Monalisa and Mondial cvs. Spounta , Nicola and Hernez cultivars showed moderate susceptibility , While Draga followed by Kara and Diamond were the most resistant . The two potato cultivars Draga the resistant cv.and lady Rosetta , the highly susceptible one were selected to study the nature of disease resistance among potato cultivars . Fraction of phenolic compounds by HPLC, oxidative enzymes and thio amino acid contents were determined in leaves and root of these potato cultivars. The resistance cultivar Draga showed higher amounts of free , conjugated and total phenols , fraction of phenolic compounds (Coumarin ,Caffeic acid,p-Coumaric acid ,Benzoic acid ,Resorcinol and Apigenin), oxidative enzymes (peroxidase and polyphenoloxidase) and thio amino acids (Cysteine , Cystine and Methionine) compared with lady Rosetta , the highly susceptible cultivar.

Keywords: Potato, *Rhizoctonia solani*, Stem canker, Black scurf, Oxidative enzymes, Phenolic compounds, Fraction of phenolic compound, HPLC.

INTRODUCTION

Misaghi (1982) recorded that phenolic compounds, their oxidative products (quinones) and derivatives (certain phytoalexins) exhibited antibiotic properties and therefore were considered by some to play a role in disease resistance. Halloin (1994) found that a histochemical test employing nitrous acid, that produces red nitroso derivatives or some phenolic compounds, was modified for use in the field and was used to demonstrate localization of phenolic compounds in healthy and *Rhizoctonia solani* infected sugar beet (*Beta vulgaris*) plants. Nitroso-reactive materials were present in petioles and around mature crown cavities or healthy plants. They were absent from tap roots, except that they occasionally were observed in lateral root tissue contained within tap roots.

El-Shaer (2002) found that infection of tolerant/susceptible lentil cultivars by *F. oxysporum*, *F. solani*, *Sclerotium bataticola* and *R. solani* led to a considerable increment in thio (Sulphur) amino acids, methionine, cystine and cysteine were obtained in all tested cultivars as a result of infection. The lowest decrease in thio-amino acids was recorded after infection in

susceptible cultivars compared with the tolerant and moderately susceptible cultivars.

Paranidharan *et al* (2003) recorded that changes in the activities of peroxidase, ascorbate peroxidase, catalase and superoxide dismutase in rice in response to infection by *Rhizoctonia solani* were studied. A significant increase in peroxidase activity was observed in *R. solani* inoculated rice leaf sheaths 1 day after inoculation and the maximum enzyme activity was recorded 3 days after inoculation at which period a 3 fold increase in peroxidase activity was observed compared to the untreated control. Ascorbate peroxidase and catalase activities significantly increased 1-2 days after inoculation and the maximum enzyme activities were recorded 5 days after inoculation.

The aim of the present work was to study the cultivar reactions of the most nine common commercial cultivars of potato to infect with *Rhizoctonia solani* under greenhouse conditions. Fractions of phenolic compounds by HPLC, oxidative enzymes and thio amino acid contents in resistant and susceptible potato cultivars were determined.

MATERIALS AND METHODS

1. Varietal reactions under greenhouse conditions:

Nine potato cultivars were screened for their reaction to black scurf and stem canker disease. The tested cultivars were Draga, Kara, Diamont, Hermez, Nicola, Spounta, Mondial, Monalisa, and Lady Rosetta. Pure cultures of the selected *R. solani* isolates (10-days-old) were used for cultivars reaction tests. Soil infestation was conducted with inoculums of each tested isolate on corn-meal sand medium. Loamy soil was sterilized with formalin solution 5 %, and left for two weeks, then soil infestation was carried out at the level of 5 % w/w and left for week before planting. Potato tubers of the nine cultivars were surface sterilized by immersing in 0.2 % Sodium hypochlorite for 2 min. and putting in 50 cm. diam. plastic pots containing 5 tubers and soil previously infested with *Rhizoctonia solani* with 4 replicates under greenhouse conditions were planted with each tested cultivar. Infection percentage was recorded 90 days after planting.

2. Biochemical studies:

Biochemical studies associated with healthy and inoculated plants with *R. solani* on two cultivars Draga, as low susceptible cv. and Lady Rosetta as highly susceptible cv. were determined. The investigated parameter included changes in phenolic compounds, fraction of phenolic compounds, oxidative enzymes, and fraction of thio amino acid at 20, 40 and 60 days after the planting.

2.1. Determination of the phenolic compounds content:

The phenolic compounds were determined in leaves and roots of the tested cultivars of potato which were inoculated separately with isolate of *R. solani* to determine total, free and conjugated phenolic compounds. The phenolic compounds content was colorimetrically determined using the follin reagent according to Snell and Snell (1953).

2.2 Fractionation of phenolic compounds

HPLC system (HP1050) was used to detect and determine some phenolic compounds from the plant tissue (Coumarin, Caffeic acid, P.coumaric acid, Benzoic acid, Resorcinol and Apegenin). Ten gms of fresh tissues of each sample were homogenized with methanol 40% and stirred on a shaker. The extract was filtered through a Whatman filter paper No.1 and the solvent was evaporated in vacuum. The dried residues containing phenol compounds were dissolved in a solution consisted of methanol: water: acetic acid, 40: 59.3: 0.7, v: v: v) and stored in vials. The separation and determination were performed on C18 column according to the method suggested by Christian (1990).

2.3 Determination of oxidative enzymes:

The oxidative enzymes were determined in leaves and roots of the tested cultivars of potato which were inoculated separately with isolate of *R. solani* to determine peroxidase (PO) and polyphenoloxidase (PPO) activity. The enzyme extraction from all samples was prepared as recommended by Maxwell and Bateman (1967). Peroxidase activity was estimated according to the method of Allam and Hollis (1972) and the activity of polyphenoloxidase was measured using the colorimetric method of Maxwell and Bateman (1967).

2.4. Fractionation of thio amino acids:

HPLC system (HP1050) was used to detect and determine thio amino acids from the plant tissue (Cysteine, cystine and Methionine). Free amino acids were extracted according to the method proposed by Shad *et al.*, (2002). The separation and determination were performed on C18 column. The method suggested by Christian (1990).

3. Statistical analysis:

The statistical analysis was computed using analysis of variance procedure described by Sendecor and Cochran (1980), the significant mean differences between treatment means were separated by Duncan's Multiple Range Test (Duncan, 1955).

RESULTS

1. Reactions of different potato cultivars to *R.solani*:

Nine potato cultivars, *i.e.* Diamond, Draga, Hermez, Kara, Lady Rosetta, Monalisa, Mondial, Nicola and Spounta were evaluated, under greenhouse conditions, for their reactions against the most pathogenic *R. solani* Sharkia's isolates code no. 18. Data presented in Table (1) show significant differences between nine cultivars, the highest percentage of infection by stem canker was obtained on Lady Rosetta (18.6 %) and followed by Monalisa (18.1%). The lowest percentage of infection was obtained by Draga (5.3%), Kara (6.4%) and Diamond (6.6%). On the other hand, the same trend was observed in reaction to black scurf symptom for the disease. The highest percentage of infection by black scurf was obtained on Lady Rosetta (10.3 %) followed by Monalisa (9.6%). The lowest percentage of infection was obtained on Draga (3.3%) and Kara (3.5%).

Table 1: Reactions of potato cultivars against the highly pathogenic *Rhizoctonia solani* isolate under green house conditions.

Cultivars	Percentages of infection (Mean ± SE)	
	stem canker	black scurf
Draga	5.3±0.386 ^f	3.3±0.407 ^d
Kara	6.4±0.450 ^f	3.5±0.418 ^d
Diamond	6.6±0.438 ^f	5.0±0.581 ^{cd}
Hermez	8.4±0.669 ^e	5.3±0.684 ^c
Nicola	10.1±0.526 ^d	6.8±0.584 ^b
Spounta	13.5±0.785 ^c	6.6±0.669 ^{bc}
Mondial	16.5±0.787 ^b	8.3±0.697 ^b
Monalisa	18.1±0.475 ^a	9.6±0.584 ^a
Lady Rosetta	18.6±0.515 ^a	10.3±0.493 ^a
LSD .05 =	1.7061584821	1.7087116575

- Each value represents the mean ± S.E (Standard Error) and mean of 4 replicates.

- Values in the same column with the same letter are not significantly at ($p \leq 0.05$).

2. Biochemical changes in susceptible and resistant potato Cultivars:

2.1. Free, conjugate and total phenolic contents in resistant cv. Draga and susceptible cv. Lady Rosetta inoculated or uninoculated with *R.solani*:

Data presented in Table (2), reveal that leaves and roots attained higher levels of free, conjugate and total phenol contents in inoculated plants compared with uninoculated plants, in roots compared with leaves and in resistant cultivar (Draga) compared with susceptible cultivar (Lady Rosetta). Twenty days after inoculation the free, conjugate and total phenols content were higher in inoculated leaves and roots compared with the uninoculated plants in each of Draga and Lady Rosetta cultivars.

Fourty days after inoculation , a similar trend to that of 20 days was observed, high content of free, conjugate and total phenols in Draga, the inoculated leaves showed activity (9.0), (7.7) and (16.7) respectively than the healthy (8.3), (7.0) and (15.3) respectively. A same trend was observed in the inoculated root. In Lady Rosetta the same trend was observed in the inoculated leaves and roots.

Sixty days after inoculation, high content of free, conjugate and total phenols in Draga was observed and the inoculated leaves showed activity (9.6), (7.6) and (17.2) respectively than the healthy (9.1), (6.8) and (15.9) respectively. A same trend was observed in the inoculated roots. In Lady Rosetta, the same trend was observed in the inoculated leaves and roots.

2.2. Fractionation of phenolic compound contents in resistant cv. Draga and susceptible cv. Lady Rosetta inoculated or uninoculated with *R.solani*:

Data presented in Table (3), reveal that leaves attained higher levels of six phenolic compounds content in inoculation plants compared with the uninoculated. Twenty days after inoculation coumarin and caffic acid content were higher in inoculated leaves compared with the uninoculated plants in each of Draga and Lady Rosetta cultivars. In the same time , Draga the resistant cv . had higher percentage of phenolic compound contents than that in the susceptible cv Lady Rosetta ., Fourty days after inoculation,, a different

trend to that of 20 days was observed by appearance of four phenolic compounds (p-coumaric acid, benzoic acid, resorcinol and apigenin) in Draga and two phenolic compounds (p-coumaric acid, benzoic acid) in Lady Rosetta. High percentage of p-coumaric acid and Benzoic acid in Draga, the inoculated leaves showed (6.79) and (0.92) respectively than the healthy (4.83) and (0.74) respectively. In Lady Rosetta the same trend was observed in the inoculated leaves.

Table 2: Free, conjugate and total phenol contents in inoculated and uninoculated resistant potato cv. Draga(R) and susceptible cv.Lady Rosetta(S) cultivars 20, 40 and 60 days after sowing under green house conditions.

Duration days	Treatments		Phenols (mg/g fresh weight)					
			Draga			Lady Rosetta		
			Free	Con.	Total	Free	Con.	Total
20	Inoculated	Leaves	7.9	6.9	14.8	6.1	5.8	11.9
		Root	109	37	146	65	27	92
	Healthy uninoculated	Leaves	7.2	6.3	13.5	5.6	5.7	11.3
		Root	62	33	95	36	42	78
40	Inoculated	Leaves	9.0	7.7	16.7	6.6	6.6	13.2
		Root	168	66	234	125	44	169
	Healthy uninoculated	Leaves	8.3	7.0	15.3	5.9	6.5	12.4
		Root	124	46	170	94	41	135
60	Inoculated	Leaves	9.6	7.6	17.2	6.9	7.4	14.3
		Root	246	49	295	173	73	246
	Healthy uninoculated	Leaves	9.1	6.8	15.9	6.2	7.3	13.5
		Root	189	78	267	123	81	204

Table 3: Phenolic compounds in inoculated and uninoculated resistant potato cv.Drage and susceptible cv. Lady Rosetta 20, 40 and 60 days after sowing under green house conditions.

Phenolic Compound	Relative concentration (%) of Phenolic compounds											
	Draga (R)						Lady Rosetta(S)					
	Days											
	20		40		60		20		40		60	
I*	H**	I	H	I	H	I	H	I	H	I	H	
Coumarin	23.6	11.4	35.2	23.5	19.3	13.5	8.99	5.62	16.1	11.8	8.16	7.53
Caffic acid	8.55	6.91	17.9	12.4	11.2	8.63	3.46	2.87	6.83	4.77	2.90	1.48
p-Coumaric acid	-	-	6.79	4.83	2.87	1.92	-	-	3.09	2.45	1.64	0.82
Benzoic acid	-	-	0.92	0.74	0.59	0.48	-	-	0.62	0.55	Tras.	Tras.
Resorcinol	-	-	0.87	0.69	0.71	0.57	-	-	-	-	-	-
Apigenin	-	-	0.81	0.52	0.62	0.43	-	-	-	-	-	-

I: inoculated ** H: healthy (uninoculated)

Sixty days after inoculation, a same trend to that of 40 days was observed by appearance of the four phenolic compounds (p-coumaric acid, benzoic acid, resorcinol and apigenin) in Draga and two phenolic compounds (p-coumaric acid, benzoic acid) in Lady Rosetta. High percentage of p-coumaric acid and benzoic acid in Draga, the inoculated leaves showed (2.87) and (0.59) respectively while the uninoculated recorded (1.92) and (0.48) respectively. In Lady Rosetta the same trend was observed in the inoculated leaves, but the amount and its increase were limited when compared with Draga cultivar.

2.3. Peroxidase and polyphenoloxidase activities in resistant cv. Draga and susceptible cv. Lady Rosetta inoculated or uninoculated with *R.solani*:

Data presented in Table (4), show that roots exhibited higher levels of PO and PPO activities than shoots in all treating dates, in both cultivars and in inoculated and uninoculated samples. The activities of PO and PPO were higher in inoculated leaves or roots compared with the healthy uninoculated plants 20 days after inoculation in each of Draga and Lady Rosetta cultivars. Also, in general, Draga, the resistant cv., had higher levels of activities regarding PO and PPO than Lady Rosetta the susceptible one.

Fourty days after inoculation, a similar trend to that of 20 days was observed in activity of PO and PPO in Draga as their activities were higher in root samples than in the leaves after inoculated. A same trend was observed in Lady Rosetta. Where the inoculated leaves showed higher activity (2.52) than the healthy uninoculated (2.23) regarding PO activity. On the other hand, PPO activities were almost alike in inoculation and uninoculated plants either in leaves or root samples.

Sixty days after inoculation, in Draga the activity of PO in inoculated leaves was higher (2.68) than that in healthy uninoculated leaves (2.47), and the same trend in inoculated and uninoculated roots. In Lady Rosetta the activity of PPO in inoculated leaves was higher (2.48) than that in healthy uninoculated leaves (2.27), and the same trend in the inoculated and uninoculated roots.

Generally, the activity of PO was higher in the 3 dates and in inoculated and uninoculated samples .while activity of PPO showed the same trend but it was very limited.

Table 4: Activity of peroxidase and polyphenoloxidase in inoculated and uninoculated Draga,the resistant cv.and Lady Rosetta ,the susceptible one with *R.solani* 20,40 and 60 days after inoculation .

Duration days	Treatments		Activity/min.			
			Draga		Lady Rosetta	
			PO*	PPO**	PO	PPO
20	Inoculated	Leaves	2.45	0.09	2.16	0.06
		Root	3.74	0.11	3.44	0.08
	uninoculated	Leaves	2.23	0.07	1.92	0.05
		Root	3.54	0.09	3.17	0.07
40	Inoculated	Leaves	2.88	0.12	2.52	0.10
		Root	4.21	0.15	3.74	0.12
	uninoculated	Leaves	2.31	0.08	2.23	0.07
		Root	3.34	0.13	3.16	0.11
60	Inoculated	Leaves	2.68	0.08	2.48	0.06
		Root	3.50	0.12	3.25	0.11
	uninoculated	Leaves	2.47	0.05	2.27	0.05
		Root	3.26	0.10	3.09	0.10

*PO:Peroxidase.

**PPO:polyphenoloxidase.

2.4. Thio-amino acids content in resistant cv. Draga and susceptible cv. Lady Rosetta inoculated and uninoculated with *R. solani*:

Three thio-amino acids cysteine, cystine and methionine were chromatographically identified in the leaves and root of Draga and Lady Rosetta cultivars using the high performance liquid chromatography (HPLC) technique. These thio-amino acids cysteine, cystine and methionine were determined at 20, 40 and 60 days after inoculated and uninoculated plants with *R.solani*.

Data presented in Table (8), reveal that leaves and roots showed higher levels of thio-amino acids cysteine, cystine and methionine contents in inoculated plants compared with uninoculated plants. Twenty days after inoculation, the thio-amino acids contents were higher in inoculated leaves and roots compared with the uninoculated plants in each of Draga and Lady Rosetta cultivars.

Forty days after inoculation, a similar trend to that of 20 days was observed as higher content of cysteine, cystine and methionine in Draga, the inoculated leaves showed higher activity (0.78), (0.64) and (0.59) respectively than the healthy uninoculated (0.67), (0.55) and (0.42) respectively, same trend was observed in the inoculated roots. In Lady Rosetta, the same trend was observed also in the inoculated leaves and roots.

Sixty days after inoculation, high content of cysteine, cystine and methionine was observed in Draga, the inoculated leaves showed the activity values of (0.51), (0.41) and (0.29) respectively than the healthy uninoculated (0.43), (0.36) and (0.22) respectively. A same trend was observed in the inoculated roots. In Lady Rosetta the contents of cysteine, cystine and methionine were very low amount (trace) in the inoculated and uninoculated leaves and root.

Table 8: Relative concentration of cysteine, methionine and cystine in inoculated or uninoculated potato cultivars differed in their susceptibility to *R.solani* 20, 40 and 60 days after sowing under green house conditions.

Duration days	Treatments		Relative concentration (%) of thio-amino acids					
			Draga			Lady Rosetta		
			Cysteine	Cystine	Methionine	Cysteine	Cystine	Methionine
20	Inoculated	Leaves	0.67	0.51	0.36	0.41	0.27	0.33
		Root	7.65	6.71	2.35	2.84	1.36	3.12
	Healthy uninoculated	Leaves	0.57	0.41	0.28	0.29	0.17	0.21
		Root	4.34	3.52	1.84	1.47	0.91	2.55
40	Inoculated	Leaves	0.78	0.64	0.59	0.49	0.32	0.45
		Root	8.71	8.64	2.52	2.67	2.11	4.37
	Healthy uninoculated	Leaves	0.67	0.55	0.42	0.38	0.24	0.32
		Root	6.74	4.61	1.96	1.91	1.23	3.19
60	Inoculated	Leaves	0.51	0.41	0.29	trace	trace	trace
		Root	2.95	trace	1.15	trace	trace	trace
	Healthy uninoculated	Leaves	0.43	0.36	0.22	trace	trace	trace
		Root	2.87	trace	0.92	trace	trace	trace

In general, 20,40 and60 days after planting , leaves and roots of Draga and Lady Rosetta cultivars recorded higher levels of thio-amino acids (cysteine, cystine and methionine) in the inoculated plants, generally in the resistant cultivar Draga comparing with both healthy uninoculated plants and the susceptible cultivar (Lady Rosetta) ,respectively.

DISCUSSION

Potato cultivars differed in their susceptibility to stem canker and black scurf diseases caused by *R.solani*. Cultivar reactions of the most common commercial cultivars of potato were tested. Data obtained from the nine tested cultivars were divided to three groups. Lady Rosetta was the most susceptible cultivars followed by the cultivars Monalisa and Mondial. Spounta, Nicola and Hermez cultivars showed moderate susceptibility, while Draga, Cara and Diamond, were most resistant. Cultivar variation may be attributed to the differences in the morphology, anatomy and chemical components of the tubers and plants. The former results are in accordance with that mentioned by Hide *et al.*, (1973). At the same time, Carling and Leiner (1990) and Demirci and Doken (1993) showed that most isolates of *R. solani* AG-3 were moderately high virulent than King Edward and Resy potato cultivars.

For that, this study was carried out to find the different changes in chemical components of the inoculated tubers and plants comparing with the healthy uninoculated tubers and plants.

Free, conjugated and total phenol contents were considered in leaves and root samples of the highly resistant cv. Draga and the highly susceptible cv. Lady Rosetta 20, 40 and 60 days after planting and inoculated with *R.solani*. Results showed that free, conjugated and total phenol contents were increased in the two potato cultivars but the increasing in the cv. Draga was higher than the cv. Lady Rosetta.

One of the ways by which plants express resistance is through plant secondary metabolites. Among them, phenolic compounds are known to impart resistance against fungal diseases. This could be explained on base of the fact that phenolic compounds are toxic to several plant pathogens (Vidhyasekaran, 2004).

Phenols and oxidation products are involved in various stages of host parasite relationship and are associated with disease syndrome of plant and plant resistant phenomena. There was a clear correlation between the oxidative enzymes, phenolic compound contents and resistance (Jindal, 2000).

Frakas and Kiraly (1962) found that the participation of an endogenous supply of phenolic compounds in plant disease resistance depends upon the activity of oxidative enzymes (polyphenol oxidase, peroxidase and catalase). Phenols are oxidized to quinone or semi-quinones which play a great role as antimicrobial. Some phenols have been found in all plants after infection as response to the ingress of pathogen, and their appearance is considered as part of an active defense response. Since the first suggestion that phenolic

intermediates have a role in the active expression of resistance the localization and timing of the host response. (Stoessl, A. 1983).

Oxidative enzymes, *i.e.* peroxidase and polyphenoloxidase were considered in leaves and root samples of the highly resistant cv. Draga and susceptible cv. Lady Rosetta 20, 40 and 60 days after planting and the inoculation with *R.solani*, obtained results showed that inoculated potato plants had a higher peroxidase and polyphenoloxidase activity than the healthy uninoculated ones. Also, the moderately resistant cultivar had higher peroxidase and polyphenoloxidase activity than the susceptible one. These differences in enzymatic activities might be attributed to that oxidative enzymes play a partial role in activating inducible defense of plant (Vera-Estrella *et al.*, 1994).

The high increment in enzymatic activity happened in early stages in resistant cultivar and hindered infection progress successfully, and by time, that enzymatic activity retreated to the normal levels (like in the healthy). On the contrary, in the susceptible cultivar enzymatic activity was increased gradually by time after inoculation but with low levels of activity which failed to hinder the infection progress.

Fractionation of thio amino acids in leaves and root samples of resistant cv. Draga and susceptible cv. Lady Rosetta 20, 40 and 60 days after planting and inoculating with *R.solani*. showed that thio amino acid contents increased in the two potato cultivars but the increasing in cv. Draga was higher than cv. Lady Rosetta.

The increasing in free amino acids contents in the inoculated tissues may be ascribed to the decomposition of host protein or to the decrease in protein synthesis as explained by (Sempio and Marte, 1968).

On the other hand, the increase in amount of tyrosine and phenylalanine were observed with inoculation, don't result of protein breakdown but the increase in concentration of the aromatic amino acids due to specific synthesis for phenolic compounds. (Vir and Grewal, 1975).

Fractionation amino acids showed that the infection in general, caused several changes in these fractions such as absence or presence of certain components and increase or decrease in the amount of other ones. These data are in harmony with those obtained by (Farag *et al.* 1986).

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"الاستجابة الكيموحيوية لبعض اصناف البطاطس للاصابة بالفطر
Rhizoctonia solani مسبب مرض تقرح الساق والقشرة السوداء في
مصر"

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تختلف اصناف البطاطس في مدي قابليتها للاصابة بمرض تقرح الساق والقشرة السوداء
المتسبب عن الفطر *Rhizoctonia solani* حيث يؤثر المرض علي بعض الصفات
المورفولوجية والبيوكيميائية علي اصناف البطاطس وفقا لطبيعة المقاومة .
تم اختبار رد فعل اهم ٩ اصناف تجارية من البطاطس للاصابة بالمسبب تحت ظروف الصوبة وتم
تقسيم تلك الاصناف الي ٣ مجاميع :

الصف *Lady Rosetta* كان اكثر الاصناف قابلية للاصابة يليه الصف *Monalisa*
والصف *Mondial*. الاصناف *Spounta* ، *Nicola* ، *Hermez* اظهرت قابلية متوسطة
للاصابة . بينما الصف *Draga* يليه الصف *Kara* والصف *Diamond* اكثر الاصناف
مقاومة للمرض .تم اختيار الصف *Draga* كصف مقاوم والصف *Lady Rosetta* لدراسة
طبيعة المقاومة من بين اصناف البطاطس المختبرة ولدراسة هذا التأثير تم تقدير المواد الفينولية (
الحررة والمرتبطة والكلية) وعمل تفريد لها كما تم تقدير الانزيمات المؤكسدة والاحماض الامينية
الكبريتية وذلك علي ثلاث فترات بعد ٦٠، ٤٠، ٢٠ يوم من الزراعة في تربة محقونة بالفطر المسبب

يحتوي الصف *Draga* المقاوم علي كميات مرتفعة من الفينولات (الحررة والمرتبطة
والكلية) وكذلك المركبات الفينولية التي تم تفريدها (*Coumarin* , *Caffic acid* , *p-Coumaric acid* ,
Resorcinol , *Benzoic acid* , *Apigenin*) مقارنة بالصف *Lady Rosetta*
الاكثر قابلية للإصابة بالفطر المسبب . كذلك وجد ان الصف *Draga* المقاوم يحتوي علي كميات
مرتفعة من الانزيمات المؤكسدة بيروكسيداز وبولي فينول اكسيداز والاحماض الامينية الكبريتية
سستين ، سستاين والميثايونين مقارنة بالصف *Lady Rosetta* الاكثر قابلية للاصابة .