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International Journal of Phytopathology

ISSN: 2305-106X (Online), 2306-1650 (Print)

<http://www.escijournals.net/phytopathology>



ROLE OF CERTAIN POTATO TUBERS CONSTITUENTS IN THEIR SUSCEPTIBILITY TO BACTERIAL COMMON SCAB CAUSED BY *STREPTOMYCES SCABIES*

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ABSTRACT

Streptomyces common scab is one of the most bacterial diseases that costs economic losses to potato plants. *Streptomyces scabiei* (Thaxter 1891) Lambert and Loria (syn. *Streptomyces scabies*) is the main causal agent of common scab in potato in the field. Eight potato cultivars namely, Diamont, Lady Rossete, Burna, Lady Belford, Skenta, Flora, Brofent and Everest were tested for their susceptibility to common scab disease under greenhouse conditions. They varied to some extent in the degree of their susceptibility. Lady Rosette appeared to be the highest susceptible one followed by Burna, Everest, and Skenta. Lady Belford, Diamont respectively, and Brofent showed intermediate disease severity. Flora cultivar exhibited the least degree of scab symptoms. The role of certain potato tubers constituents in the physiology of disease susceptibility was investigated (sugar contents, pectin substances and calcium contents), Lady Belford cultivar contained the highest concentration of total sugar followed by Flora, Burna, and Everest but Skenta, Lady Rosette respectively and Diamont were approximately had equal in total sugar contents, while, Brofent cultivar contained the lowest content of total sugar. Skenta cultivar contained the highest concentration of pectin substances and calcium contents followed by Flora and Lady Belford finally Burna cultivars respectively.

Keywords: Potato, Calcium content, common scab, *Streptomyces scabies*.

INTRODUCTION

Potato (*Solanum tuberosum* L.) is one of the most important vegetable crops in the world. In Egypt, potato is the 4th most important staple food; the total cultivated area in Egypt was about 318,214 feddans and the total production was about 3.7 million tons according to Food and Agriculture Organization (FAO, 2010), also it is the second most important crop producing energy, in addition to that it is one of the most important export crop in Egypt. Furthermore, potato is a very important source of complex carbohydrates and contains a variety of phytomaterials that have antioxidant activity. It has more protein than corn and nearly double amount of calcium. It also contains iron, potassium, zinc, copper, manganese, vitamin C, B₆ and dietary fibers (Caprara, 2012 and Bastin, 1997).

Common scab disease of potato can cause significant yield and quality losses, and current control methods for

this disease are inadequate (Fyans, *et al.*, 2016 and Hooker, 1981). Methods used to control common scab include chemical treatments of seed potato tubers (Davis, *et al.*, 1976), irrigation (Adams and Lapwood, 1978), changing soil pH (Pavlista, 1992 and Waterer, 2002), rotation strategies (Li, *et al.*, 1999), use of a tolerant cultivar (Bouchek-Mechiche *et al.*, 2000), and biological control with non-pathogenic streptomycetes (Liu, *et al.*, 1995; Ryan and Kinkel, 1997; Agbessi, *et al.*, 2003 and Arseneault, *et al.*, 2015).

The best management of the disease cannot be achieved without using resistant cultivars, which is easier to apply, cheaper, and acceptable to farmers. (Rich, 1983) mentioned that the most effective method of scab control is to plant resistant cultivars. It has been showed that there is a relationship between levels of some tubers constituents and their susceptibility to common scab disease (Rich, 1983). Many researcher suggested that these constituents such as: calcium, potassium, manganese (Davis *et al.*, 1976) or reducing sugars (Goto, 1981) may play an important role in disease resistance.

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This work is aimed to study the susceptibility of some potato cultivars against common scab, and the role of constituents on their susceptibility to bacterial common scab.

MATERIALS AND METHODS

Source of *Streptomyces scabies* the pathogen of common scab disease: Isolate of *Streptomyces scabies* stc 11 from a diseased potato in Sohag Governorate, Egypt were used. The pathogenicity of this isolate was previously tested by authors (Hosny, *et al.*, 2014).

Susceptibility of certain potato cultivars to common scab disease: Eight potato cultivars namely, Diamont, Lady Rossete, Burna, Lady Belford, Skenta, Flora, Brofent and Everest were tested for their susceptibility to common scab disease under greenhouse conditions. The cultivation of potato tubers were planted in 20th September 2010.

Healthy potato tubers were planted in 30-cm-diameter sterilized pots containing sterile 1:1 soil/sand mixture. Potato tubers were surface disinfected by soaking tubers in 0.5 % sodium hypochlorite for 5 minutes then rinsed twice in sterilized water directly before planting. Fifty ml of ten days old culture from isolate Stc 11 (10⁸ CFU/ml) were added to each pot before planting. Five replicate pots for each isolate were used. Pots were arranged in a complete randomized block design with five replicates. Control plants were treated similarly by tap water. The plants were grown in greenhouse under natural temperatures (25±3°C). Plants were fertilized every 15 days with urea 46% (20g/pot) and irrigated with water when necessary. After 90 days from planting tubers of each tested cultivar were harvested and scored for scab severity. Tubers from each replicate were examined for scab symptoms as described before (Hosny, *et al.*, 2014).

Role of certain potato tuber constituents in their susceptibility to bacterial common scab

Pectin contents: The anthrone sulphuric acid methods were used in determining the pectin content of potato tubers (Badour, 1959) as follows:

Anthrone sulphuric acid reagent: The reaction mixture consists of 0.2 g anthrone, 8 ml absolute ethyl alcohol, 30 ml distilled water and 100 ml concentrated H₂SO₄. They were successfully mixed in a conical flask under continuous cooling in ice bath. This reagent should be always freshly prepared. Half ml sample + 4.5 ml anthrone reagent were thoroughly mixed and boiled

for 7 minutes. After cooling the absorbency against blank was measured photometrically at 620 nm using spectrophotometer model "Miltonroy Spectronic 601". A standard curve was prepared using pure pectin.

Pectin fractionation: This was conducted essentially according to Galbraith and Shields, (1981) as follows:

Tissues preparations were first extracted twice with 0.5% ammonium oxalate-oxalic acid at 90°C for 24 hours. After each extraction the residue was separated from the supernatant fluid by centrifugation. To each extract, 8 volumes of absolute alcohol were added, slowly with stirring and mixtures were allowed to stand at -10°C overnight. The resulting precipitates were collected by centrifugation, dried in vacuum over anhydrous CuSO₄ for 3 hours, Stored over P₂O₅ and designated as pectin fraction 1 and 2, respectively.

Sugar contents

Extraction and Determination of total sugars: One gram from healthy potato tubers samples was cut into pieces then put in test tube, and 5 ml of 70% ethanol were added, then tubes were subsequently in water bath at 70°C for 72 hrs and left to dry. The residue was dissolved in 5 ml 50% iso-propyl-alcohol. All samples were stored at 4°C until analyzed.

Eight tenth ml of extracted sample was mixed with 5 ml distilled water and 4 ml picric acid reagent in a sterilized test tube. The mixture was boiled on a water bath for 10 minutes then left to cool. One ml Na₂CO₃ 20% (w/v) was added to the mixture and boiled for another time on a water bath for 10 minutes then left to cool. The mixture was completed to final volume (50 ml) using distilled water. Color optical density of the reacted mixture was measured on absorbance spectrophotometer "Miltonroy Spectronic 601" at 540 nm (Thomas and Dutcher, 1924).

Standard curve of sugar values: One gram of glucose was dissolved in a volume of distilled water and the total volume was made up to one liter. Different concentrations of glucose solutions were prepared in 100 ml of distilled water in volumetric flasks. Eight tenth ml of different glucose concentrations was taken separately and mixed with 5 ml distilled water and 4 ml picric acid reagent in sterilized test tube. The mixture was boiled on a water bath for 10 minutes then left to cool. One ml Na₂CO₃ 20% (w/v) was added to the mixture and boiled for another time on a water bath for 10 minutes then left to cool. The mixture was completed to a final volume (50 ml) with distilled

water. Color optical density of the reacted mixture was measured on absorbance spectrophotometer "Miltonroy spectronic 601" at 540 nm and the known concentrations of glucose were plotted for comparison.

Calcium, sodium and potassium contents: Tubers samples were digested using the wet ash with nitric acid (HNO₃) and Bircholoric acid (HCIO₄) method following the next steps:

- a. Sample of 0.21gram of oven dry finally ground plant was transferred to tall beaker and 20 ml of concentrated HNO₃ witting the entire sample. The beaker was covered with a watch glass and was allowed to stand overnight.
- b. Beaker was placed on a hot plate kept inside a fume chamber. Continue heating gently until solid particles nearly disappeared.
- c. The beaker was removed from the hot plate and was allowed to cool.
- d. Ten ml of 72% of HCIO₄ was added to the beaker which was placed on a hot plate. Continue heating gently first and the vigorously until dense white fumes formed during heating subsides, and the contents became colorless clear solution.
- e. The beaker was cooled, distilled water was added then the contents were stirred with a glass rod and were transferred to 50 ml volumetric flask.
- f. Continue transfer of digested residue with portions of distilled water into the same volumetric flask until the entire digest had been quantitatively transferred into the same flask.
- g. The volume was made to 50 ml with distilled water and the flask was covered and the contents were

mixed by turning the flask upside down 4-5 times and the flask was left to stand overnight.

- h. The diluted digest was filtered through a dry Whatman No. 42.
- i. The subsequent filtrate was collected and preserved as a sample test solution for the determination.
- j. A blank test solution was similarly prepared without plant sample.

Sodium and potassium were measured by flame photometer (CL378- ELICO) while calcium was measured by Atomic Adsorption Spectrophotometer according to (Jackson, 1973).

Statistical analysis: Analysis of variance (ANOVA) was carried out using MSTAT-C program. The Least Significant Difference (LSD) at P≤0.05 was applied to detect differences among treatments Gomez and Gomez, (1984).

RESULTS

Susceptibility of certain potato cultivars to common scab disease:

The susceptibility of certain potato cultivars to infect with common scab was evaluated. Isolate Stc 11 the highest pathogenic one used in this test. The experiment was conducted in 2010 growing season under greenhouse condition. Data of this experiment presented in figure 1 indicated that the tested potato cultivars susceptible differently for *S. scabies* isolate Stc 11. They varied to some extent in the degree of their susceptibility. Lady Rosette exhibited the highest susceptibility one followed by Burna, Everest, and Skenta. Lady Belford, Diamont respectively and Brofent showed intermediate disease severity. Data also indicated that Flora cultivar exhibited the least degree of scab symptoms.

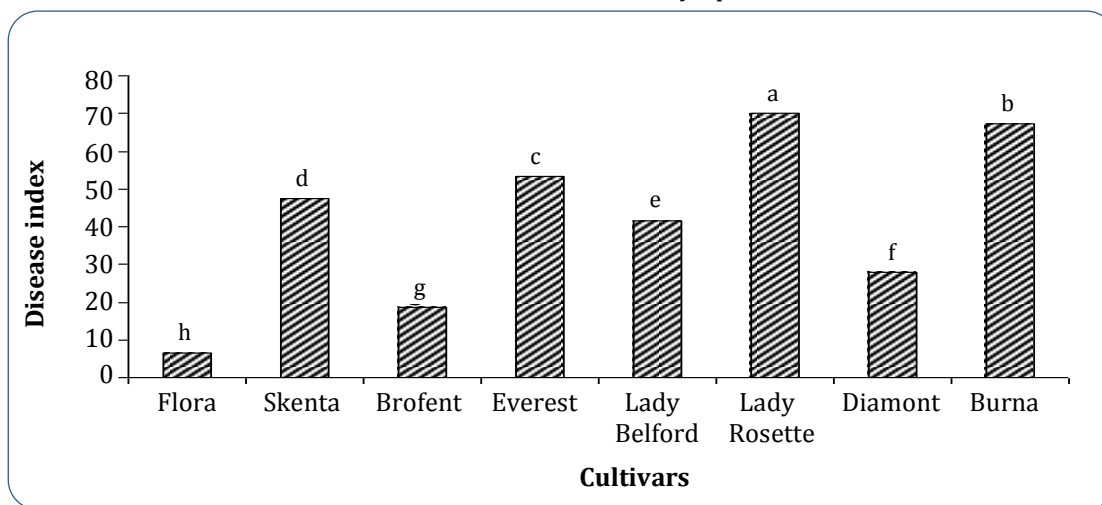


Figure 1. Susceptibility of certain potato cultivars to common scab disease.

Role of certain potato tuber constituents in their susceptibility to bacterial common scab:

The role of total sugar, pectin, calcium, sodium, and potassium contents of potato tubers on their susceptibility to common scab disease was investigated. Healthy potato tubers of eight cultivars were used.

Total sugars: Data in Table (1) indicated that Lady Belford cultivar contained the highest concentration of total sugar followed by Flora, Burna, and Everest but Skenta, Lady Rosette respectively and Diamont were approximately equal amount of total sugar contents. Whereas Brofent cultivar contained the lowest content of total sugar.

Pectin contents: Data in Table (1) indicated that the percentages of pectin substances differed by cultivars. Skenta cultivar contained the highest concentration of pectin substances followed by Flora and Lady Rosette, while Burna cultivar contained the lowest concentration of pectin substances.

Table (1). Effects of pectin and sugar potato cultivars contents on susceptibility of bacterial common scab.

Potato cultivars	Sugar (mg/kg)	Pectin %
Flora	2.38 b	0.439 b
Skenta	1.35 f	0.528 a
Brofent	0.90 h	0.225 e
Everest	1.65 d	0.227 e
Lady Belford	3.53 a	0.293 d
Lady Rosette	1.45 e	0.314 c
Diamont	1.20 g	0.210 f
Burna	1.90 c	0.181 g
LSD	0.054	0.0071

Different letters indicate significant differences among treatments within the same column according to L.S.D (Least Significant difference test 5%).

Calcium, sodium and potassium contents: Data presented in figure 2 indicated that tested cultivars showed different concentrations of calcium, sodium and potassium. Diamont cultivar contained the highest concentration of sodium followed by Burna and Skenta.

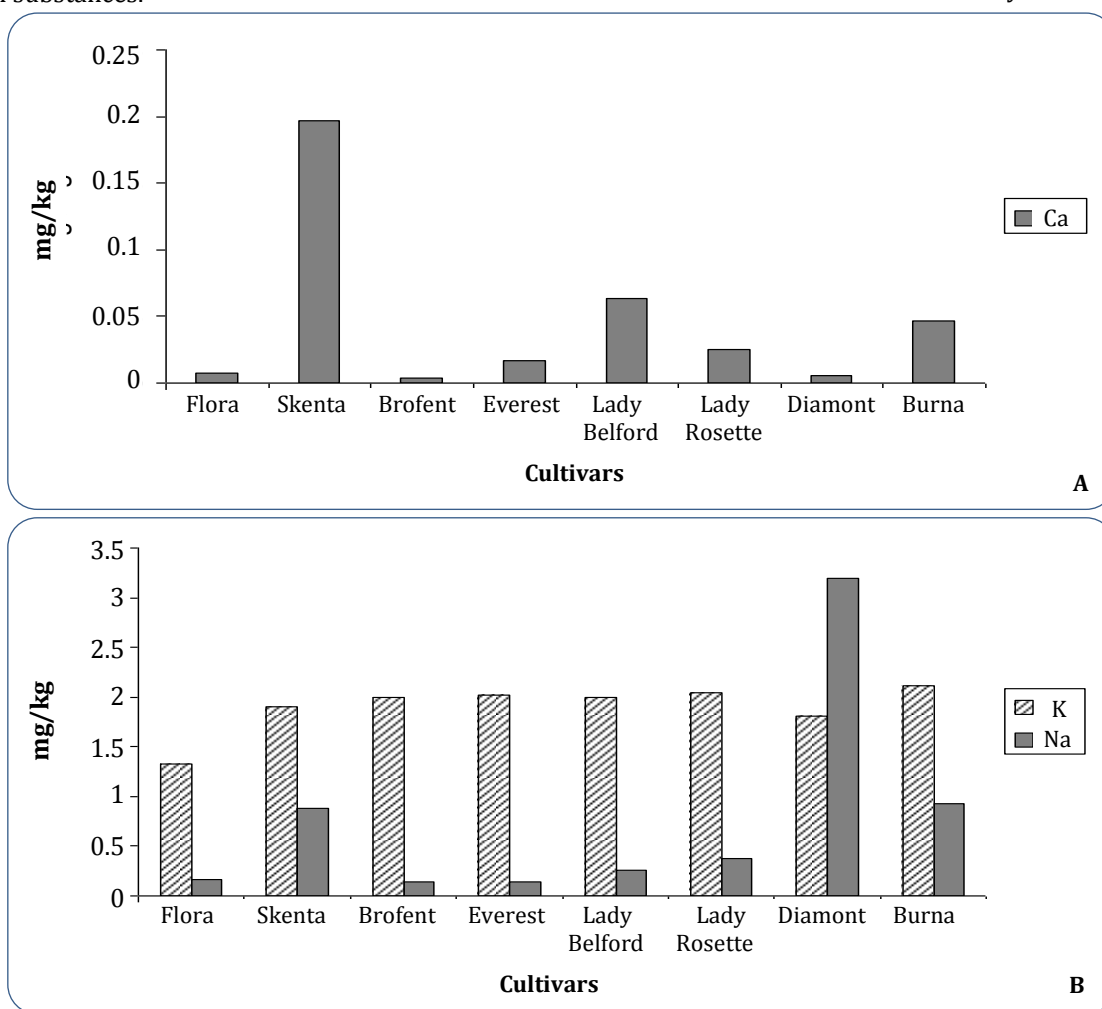


Fig. 2. Potato cultivars contents of sodium and potassium in total tubers (A and B).

Finally Brofent and Everest contained the lowest concentration of sodium. Also, data indicated that Burna cultivar contained the highest concentration of potassium followed by Lady Rosette, Everest and Lady Bellford. Whereas Flora contained the lowest concentration of potassium. Skenta cultivar contained the highest concentration of calcium followed by Lady Bellford and Burna respectively, whereas Flora, Diamont and Brofent contained the lowest concentration of calcium.

Role of certain potato peel constituents of some potato cultivars on their susceptibility to bacterial common scab:

Data in Table (2) indicated that the tested cultivars varied in their peel constituents of calcium, sodium and potassium. Peels of Skenta and Flora cultivars contained the highest concentration of calcium followed by those of Diamont then Lady Bellford and Everest respectively. Whereas peels of Burna, Lady Rosette and Brofent respectively contained the lowest concentration of calcium.

Table (2). Effects of Ca, K and Na potato cultivars peels contents on susceptibility of bacterial common scab (mg/kg).

Potato Cultivars	Ca	K	Na
Flora	0.214 a	2.310 e	0.857 d
Skenta	0.214 a	2.381 d	0.881 c
Brofent	0.010 e	2.548 b	0.167 g
Everest	0.052 c	2.524 b	0.905 b
Lady Bellford	0.058 c	2.452 c	0.143 h
Lady Rosette	0.017 e	2.500 b	0.524 f
Diamont	0.116 b	3.214 a	1.000 a
Burna	0.034 d	2.095 f	0.667 e
LSD	0.0061	0.0269	0.0143

Different letters indicate significant differences among treatments within the same column according to L.S.D (Least Significant difference test 5%).

Peels of Diamont cultivar contained the highest concentration of potassium followed by those of Brofent, Everest, Lady Rosette, and Lady Bellford respectively. Whereas peels of Burna contained the lowest concentration of potassium.

Peels of Diamont cultivar contained the highest concentration of sodium followed by peels of Everest then peels of Skenta and Flora. Finally peels of Brofent and Lady Bellford contained the lowest concentration of sodium.

DISCUSSION

In the present study the tested potato cultivars were susceptible to common scab disease with some differences in their susceptibility. Tubers of cultivar

Lady Rosette appeared to be the highest susceptible one followed by Burna, Everest, and Skenta. Lady Bellford, Diamont respectively and Brofent showed intermediate disease severity. Data also indicated that Flora cultivar exhibited the least degree of disease severity. Such results are in agreement with those reported by several previous studies (Hiltunen, *et al.*, 2005; Wanner, 2009 and Sedláková, *et al.*, 2013) who reported that potato varieties have a variable response to *S. scabies* presence. However, no variety was symptomatic over all trial.

It should be noted that differences in susceptibility of potato cultivars to common scab disease may be due to certain physiological aspects as well as their constituents of certain antimicrobial contents. To study the role of total sugars, pectin, calcium, sodium, and potassium contents of potato tubers on their susceptibility to common scab disease, healthy potato tubers of eight cultivars were tested. Results indicated that Lady Bellford cultivar contained the highest concentration of total sugar followed by Flora, Burna, and Everest but Skenta, Lady Rosette and Diamont were approximately equal in total sugar contents. Whereas Brofent cultivar contained the lowest content of total sugar.

Data indicated that there is no correlation between tuber contents of total sugars and common scab severity. Also obtained results are disagree with those reported by Goto, (1981) they reported that tubers of scab-susceptible cultivars had more scab and a higher reducing sugar content than those of resistant cultivars. Data indicated that there is a positive correlation between tuber contents of potassium and common scab severity. Burna cultivar contained the highest concentration of potassium followed by Lady Rosette, Everest and Lady Bellford. Whereas there is no correlation between tuber contents of sodium and common scab severity. these results are disagree with those reported by Davis, *et al.*, (1976) who reported that there is a negative correlations between potassium and scab severity and Kristofek, *et al.*, (2000) they reported that significant differences in accumulation of some elements such as Ca, P and K were found between cultivars differing in susceptibility to common scab.

Data indicated that there is a positive correlation between tubers contents of calcium and common scab severity. Skenta cultivar contained the highest concentration of calcium followed by Lady Bellford and Burna, whereas Flora, Diamont, and Brofent contained the lowest concentration of calcium. Such results are in

agreement with those reported by several workers among them Davis, *et al.*, (1976) who reported that nutrient analysis of petioles collected early in season showed a highly significant positive linear correlation between calcium and scab severity. Also, Horsfall, *et al.*, (1954) and Goto, (1985) mentioned that increased tuber calcium was positively correlated with scab severity.

Data also indicated that there is a negative correlation between peels contents of calcium and common scab severity. Such results are in disagreement with those reported by Kristofek, *et al.*, (2000) and Lambert and Manzer, (1991) they reported that concentrations of mineral elements including calcium in the tuber periderm may not be related to scab incidence or severity.

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