

(Original Article)



## Occurrence of New Pathogenic Species of *Streptomyces* Causing Common Scab Disease on Potato Tubers in Egypt

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### Abstract

This study was aimed to identify major species of *Streptomyces* that are able to cause potato common scab in Egypt. Potato tubers showing typical symptoms of common scab disease were collected from three different regions of Upper Egypt. The causal pathogens were isolated, and morphological, biochemical and pathogenicity tests were conducted *in vitro* to preliminary identify the isolated *Streptomyces* isolates. Moreover, the partial 16S rDNA gene sequencing was used to genetically identification of the isolates. Four new pathogenic species that have not previously been reported in Egypt were identified. Four species of *Streptomyces rochei*, *S. rutgersensis*, *S. lateritius*, and *S. bottropensis* were identified. Isolates S1, S10, S14, S17, S18, S20, and S21 were identified as *Streptomyces rochei*, isolates S11 was identified as *S. rutgersensis*, isolates S13 identified as *S. lateritius* and isolates S15 was identified as *S. bottropensis*. To the obtained data, this is the first report that *S. rochei*, *S. rutgersensis*, *S. lateritius*, and *S. bottropensis* cause of potato common scab disease in Egypt. The different of obtained 16S rDNA sequences were deposited at NCBI GenBank with the following accession numbers: (S1) MZ267260, (S10) MZ267264, (S14) MZ267268, (S17) MZ267271, (S18) MZ267272, (S20) MZ267274, (S21) MZ267275, (S11) MZ267265, (S13) MZ267267, and (S15) MZ267269. The four newly identified isolates showed different pathogenicity, morphological and biochemical characteristics.

**Keywords:** *Potato (Solanum tubersum)*, *Common scab*, *16S rDNA gene sequencing*, *Streptomyces spp.*

### Introduction

Potato (*Solanum tubersum* L.) is the fourth most important food crop worldwide and the first economically important vegetable crop. It is cultivated in several regions worldwide, especially in Europe, America, and Asia. Europe and Asia are major producers and account for about 80% of world potato production and the main consumers are Europe, North America, and Asia. Potato is cultivated in an area of 20 million hectares and produces close to 400 million tons annually that are consumed fresh or processed (Medina *et al.*, 2017). Potato is one of the

most important crops grown in Egypt for local consumption, export, and processing. Potatoes are the second most important vegetable after tomatoes, both in terms of cash value and total tonnage produced. Potatoes are Egypt's largest horticultural export. In most recent years the EU has accounted for about 70% - 90% of Egyptian potato exports. The total cultivated area in Egypt was about 289,926 feddans (299.936 Acres) and the total production was about 5,078,374 tons (FAO, 2020).

Common scab caused by pathogenic *Streptomyces* spp. plays a decisive role in the qualitative and quantitative production of potatoes worldwide (Bora *et al.*, 2023). Potato common scab (PCS) disease has long been recognized as one of the most recalcitrant diseases afflicting potatoes worldwide (Bouček-Mechiche *et al.*, 2000, Kumar *et al.*, 2024 and Zhao *et al.*, 2008). Scab disease harms a broad range of root crops, including potato, sweet potato, radish, carrot, sugar beet, and burdock, with potato scab disease especially causing large economic losses (Kumar *et al.*, 2024). The typical scab symptoms on potato tubers are superficial, raised and deep-pitted corky lesions that effect tuber quality and marketability in fresh markets or processing operation. (PCS) can be caused by a gram-positive bacterium from the *Streptomyces* genus, at least 13 different *Streptomyces* spp. have been found to cause PCS on potato worldwide. *S. scabies* is the oldest characterized pathogen. The causative agent of potato scab disease multiple species of the genus *Streptomyces*. *S. scabies*, *S. acidiscabies*, and *S. turgidiscabies* are the most well-known studied causal agents (Lambert and Loria 1989, and Kers *et al.*, 2005). Scab symptoms are mainly caused by secreted toxins from *S. scabies*, such as thaxtomins, concanamycin, borrelidin, or FD-891 (Loria *et al.*, 2006, Bignell *et al.*, 2010 and 2014)

In Japan there are only three species of potato scab pathogens reported. Different *Streptomyces* spp., (more than 14) are able to cause scab diseases on potato worldwide and differ from each other and from *S. scabies* in their morphological and physiological cultural either in the laboratory or in the field (Hao *et al.*, 2009). The purpose of this study was to identify major species of *Streptomyces* that are able to cause common scab pathogens in Egypt by 16S rDNA gene sequencing, and to distinguish between the new pathogenic species by morphological and biochemical characters.

## Materials and Methods

### Isolation of the causal pathogens from the infected potato tubers and infected soil

Infected potato tuber samples were collected from the infected potato fields from three localities, in El-Minia, Assiut, and Sohag. The infected potato tubers were washed thoroughly in running tap water for 5 to 7 minutes to remove adhered soil to their surface and then were air-dried. The small tuber pieces containing scab lesions were cut out using a sterilized knife and surface disinfected for one minute in 70% ethanol. After several rinses in sterile distilled water, the tissues were ground in a pestle mortar in 5 ml of sterilized distilled water. One loop of the

suspension that resulted after grinding the tissues was taken and streaked on nutrient sucrose agar (NSA) Consisting of 5 g/L Sucrose, 5 g/L Beef extract, 10 g/L Peptone, and 20 g/L Agar and pH 7.0 (sterilized at 120°C for 15 min). Inoculated plates were incubated at 30°C and monitored after 3, 5, and 7 days, essentially as described previously by Loria, (2001).

For isolation of plant pathogenic *Streptomyces* spp. from the soil, samples were collected from the rhizosphere of the infected potato tubers from the three localities mentioned before. The collected soil samples were mixed well by handshaking, and then three grams were suspended in 100 ml of physiological saline (NaCl 9 g/L) then incubated under orbital shaker incubation at 30°C with shaking at 200 rpm for 30 min. The mixture was allowed to settle, and serial dilutions up to 10<sup>-5</sup> were prepared. An aliquot of 0.1 ml of each dilution was taken and spread evenly over the surface of NSA (5 g/L Sucrose, 5 g/L Beef extract, 10 g/L Peptone, and 20 g/L agar, pH 7.00, sterilization at 120°C for 15 min). Plates were incubated at 30°C and monitored after 3, 5, and 7 days, essentially as described previously (El-Hadi *et al.*, 2019, and Rahman *et al.*, 2011). The actinomycete colonies were picked up and streaked multiple times on NSA plates, and then incubated at 30°C for 7 days to check their purity. The pure actinomycete isolates were maintained on NSA slants at 4°C.

Bacterial strains isolated from infected potato tubers and infected soil was investigated morphologically. Aerial mycelium and spores were sampled from the surface of developing colonies by lightly pressing a coverslip on the colony surface, mounting it on agarose-coated slide (1% agarose in PBS), and examining the bacteria by phase-contrast microscopy. The isolates were identified as genus *Streptomyces* by comparing the morphology of spore chains and spore-bearing hyphae suggested that the isolates were of genus *Streptomyces* (Holt *et al.*, 1994). Finally, representative colonies were selected and streaked on new plates of *Streptomyces* isolation agar medium. Agar plates were then inoculated with the strains and incubated at 30°C until good growth was observed. The isolated strains were conserved at 4°C for further lab work.

### **Pathogenicity tests**

Pathogenicity assay of the isolates was conducted using the slice technique, and then confirmed by the mini tuber method in the greenhouse (data not shown) (Loria *et al.*, 1995).

Slice technique was performed on the isolated bacteria of *Streptomyces* spp. Potato tubers free from the surface disease were washed and sterilized for 1 min with 0.5 % NaOCl. A tissue disk (2.0 cm in diameter and 1.0 cm in height) was bored from the tuber and placed on moist filter paper in a Petri plate. The pathogenicity of the isolated strains was examined and compared with control and photographed. *Streptomyces* spp. was grown on NSA medium for 7 days, agar plug from *Streptomyces* agar culture was placed at the center of the potato tuber slice. The plates were incubated in a moist closed container for 5 to 7 days at 25-28°C in the dark. Necrosis of the tuber slices was evaluated as the area of necrosis minus

the area of inoculation, essentially as described previously (MARTI *et al.*, 1993 and McKenna *et al.*, 2001).

## **Molecular identification of the pathogenic bacterial isolates using 16S rDNA gene sequencing**

### **DNA extraction**

Total genomic DNA was extracted from *Streptomyces* tested isolates using a downscaled version of the CTAB procedure (Kieser *et al.*, 2000). The isolates were grown on MYM broth medium at 30°C for 72 h with shaking. The cultures were harvested by centrifugation at 3000 g for 10 min, the mycelia pellets were resuspended in 567 µl of 2 mg/ml lysozyme solution then the Eppendorf tubes were incubated at 37°C for 0.5-1 h., 30 µl of 10% SDS was added and mixed, 3 µl of 20 mg/ml Proteinase K was added, mixed and incubated at 37°C for 1 h., 100 µl of 5M NaCl was mixed thoroughly, a prewarmed solution of 10% hexadecyltrimethyl ammonium bromide (CTAB) in 0.7M NaCl was added, mixed, and incubated on 65°C for 10 min or more. The DNA was extracted by adding an equal volume of Chisam (chloroform isoamyl alcohol 24:1) and mixed thoroughly and was left on the bench for a few minutes. Then a centrifugation for 10 min at max speed was applied, and the supernatants were carefully pipetted off, and transferred into a new tube. 0.6 volumes of isopropanol were added and mixed by inverting the tubes then centrifuged at the max speed for 15 min. The supernatants were removed and 500 µl of 70% EtOH were added to wash the DNA pellet, centrifugation for a few minutes, then supernatants were removed. The pellet was kept drying. Then the DNA was dissolved in 50-100 µl TE. The concentrations of the extracted DNA were measured by using the Nanodrop. DNA quality was checked on 1% agarose gel and then kept at -20 C for further work.

### **PCR and sequencing**

To identify the isolates by sequencing, the isolates were subjected to PCR amplification of the 16S rDNA gene using 16S rDNA universal primers (Weisburg *et al.*, 1991):

- F-Primer (5-AGAGTTTGATCCTGGCTCAG-3)
- R-Primer (5-CGGTTACCTTGTTACGACTT-3)

Each PCR reaction contained; 100 ng of DNA template, 1X of ThermoPol Reaction Buffer, 2.5 U of Taq DNA polymerase (New England Biolabs), 200 µM dNTPs, 0.5 µM F-Primer, 0.5 µM R-Primer, and H<sub>2</sub>O added to afford a total volume of 50 µl. The PCR reaction was carried out with an initial denaturation at 95°C for 3 min followed by 30 cycles of denaturation at 95°C for 30 s, annealing at 50°C for 1 min, and extension at 68°C for 1 min 30 s. Following the cycling, a final extension at 68°C for 5 min was performed. Amplification of PCR products were tested on 1% agarose gel, and then the tested PCR products were purified using the purification kit (QIAquick PCR Purification Kit, Qiagen). The purified PCR products were sequenced at the sequencing service offered by MWG-Biotech AG (Germany).

## **Morphological and biochemical characteristics**

The obtained isolates that proved to be pathogenic and cause common scab on potato tubers were characterized morphologically and physiologically to distinguish the new reported species of *Streptomyces* as recommended by Schaad (1992), Bergey's Manual of systematic Bacteriology Krieg and Holt (1984), and Bergey's Manual of Determinative Bacteriology 9<sup>th</sup> edition Holt *et al.*, (1994). The following morphological and physiological characteristics were tested: Colony color, Sporulation, Motility, Gram staining, Gelatin liquefaction, Starch hydrolysis, Catalase, Esculin hydrolysis, H<sub>2</sub>S production, Levan production, M. R. Test, V. P. Test, Phenylalanine deaminase, Casein hydrolysis, Urea test, Nitrate reduction, Potassium Hydroxide, Melanin production on Peptone yeast extract iron Agar and Tyrosine Agar and Fermentation of some carbon compound, Mannose, Glucose, Mannitol, Cellulose, Lactose, Fructose, Dextrose, Maltose, Sorbitol, and Sucrose.

## **Results and Discussion**

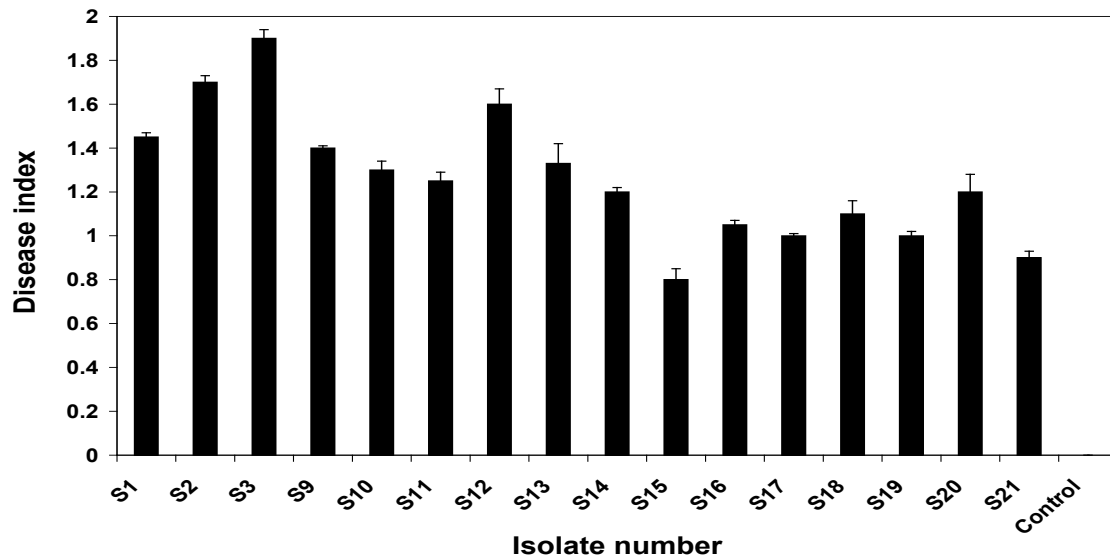
### **Isolation of the common scab disease causal pathogen and the pathogenicity test**

One hundred and twenty-three pure isolates of *Streptomyces* spp. were isolated from different sources and different governorates. All the isolates were applied to the pathogenicity test by the slice technique (*in vitro*) that divided them according to their pathogenicity into high, moderate, and non-pathogenic isolates. 67 isolates were pathogenic, and 56 isolates were non-pathogenic. The 67 pathogenic isolates included 13 highly pathogenic isolates and 54 moderately pathogenic isolates Table (1). The non-pathogenic isolates were excluded from further studies. Then, 16 isolates that resemble the two groups (8 highly pathogenic isolates and 8 moderately pathogenic isolates) were selected randomly for further studies (Molecular identification by sequencing, and Morphological and biochemical characterization).

Results in Fig. (1) showed that the selected 16 isolates were pathogenic and produced symptoms of necrosis on potato slices, but they varied in their virulence. Isolate No. S3 gave the highest disease index, while S15 caused the least disease index.

### **Identification of the pathogenic isolates by the 16S rDNA sequencing**

The 16S rDNA sequences of the 16 isolates of *Streptomyces* were determined. The obtained sequences were applied to the BLAST tool available in the GenBank database (NCBI). The molecular identification of the isolates was based on the most significant alignments (highest bits scores; and nucleotide similarity) of this sequence with the sequences available on the GenBank. The isolate identification and the Accession number of each isolate are summarized in Table (1).



**Fig. 1. Pathogenicity test of *Streptomyces* spp. on potato slices. Means and standard deviation for three potato slices as replicates per isolate and a negative control is shown.**

The data of the 16S rDNA sequencing has revealed that the 16 isolates included seven different *Streptomyces* species. Out of these seven species, we found three species (*S. coelicoflavus* (S2), *S. europaeiscabiei* (S3, S16, S19), and *S. acidiscabies* (S9 and S12) were reported in Egypt as a causal pathogen of common scab in potato (Table 1). These old reported isolates were not included in the further experiments.

Out of the 16 sequenced isolates, 10 isolates were identified as *S. rochei* (S1, S10, S14, S17, S18, S20, and S21), *S. rutgersensis* (S11), *S. lateritius* (S13), and *S. bottropensis* (S15). This is to our knowledge the first time it is reported that strains of these species can cause common scab disease in Egypt. Only the 10 new pathogenic isolates were included in the further studies.

The obtained sequences were used to estimate the genetic similarity among the 10 isolates (Table 2). The 16S sequences of all the identified *S. rochei* isolates (S1, S10, S14, S17, S18, S20, and S21) were identical and showed 100% similarity. The lowest genetic similarity was 95.2 % between *S. bottropensis* (S15) and the *S. rochei* group.

A multiple sequence alignment for the obtained sequence of the 10 isolates was performed using the DNAMAN software (Lynnon Biosoft, San Ramon, USA) to construct a phylogenetic tree (Fig. 2A) to show the relationship among the newly reported isolates.

Table 1. The pathogenicity of the total number of the isolated *Streptomyces* isolates and the molecularly identification of the selected isolates

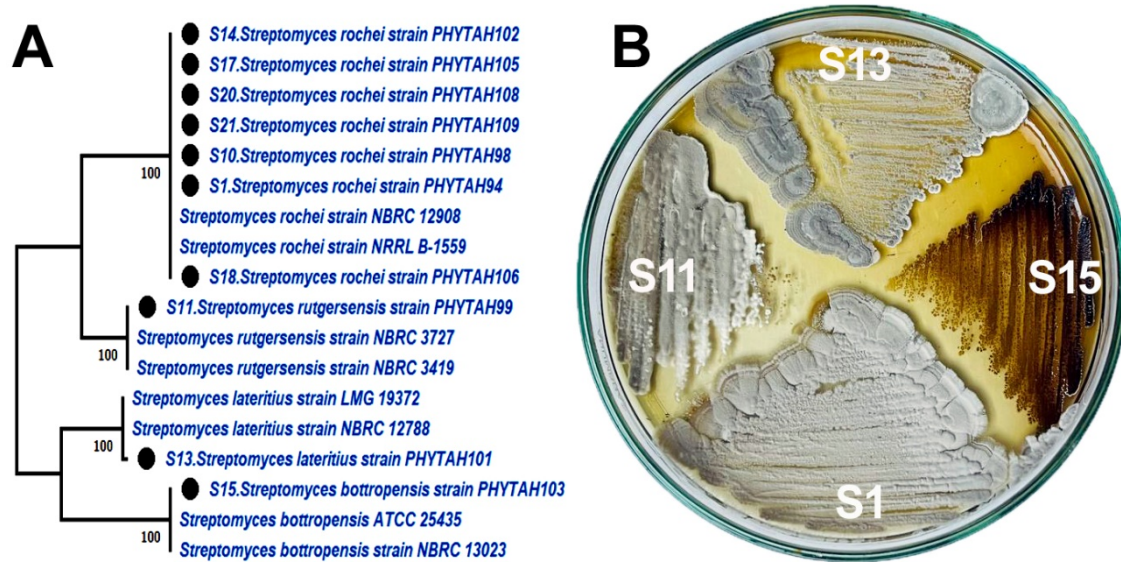
Total No. of isolates	Pathogenicity	Isolation Source	Selected isolates	Isolate identification	Percentage of identity	Length of alignment	Accession number	Reported as a common scab disease pathogen in Egypt	
13	High	Assiut infected tubers	S1	<i>Streptomyces rochei</i>	99.92%	1487	MZ267260	New Reported	
	High		S2	<i>S. coelicoflavus</i>	100.00%	1404	MZ267261	(Hassan <i>et al.</i> , 2016)	
	High		S3	<i>S. europaeiscabiei</i>	99.92%	1441	MZ267262	(Abdel-Rahman <i>et al.</i> , 2012)	
	moderate	Assiut infected soil	S9	<i>S. acidiscabies</i>	99.85%	1526	MZ267263	El-Sayed (2000)	
	moderate		S10	<i>S. rochei</i>	100.00%	1487	MZ267264	New Reported	
	moderate		S11	<i>S. rutgersensis</i>	100.00%	1473	MZ267265	New Reported	
	High		S12	<i>S. acidiscabies</i>	99.85%	1526	MZ267266	El-Sayed (2000)	
	moderate		S13	<i>S. lateritius</i>	99.92%	1426	MZ267267	New Reported	
	moderate		S14	<i>S. rochei</i>	100.00%	1487	MZ267268	New Reported	
	54	moderate	El-Minia infected tubers	S15	<i>S. bottropensis</i>	100.00%	1527	MZ267269	New Reported
		moderate		S16	<i>S. europaeiscabiei</i>	99.92%	1441	MZ267270	(Abdel-Rahman <i>et al.</i> , 2012)
		moderate	Sohag infected tubers	S17	<i>S. rochei</i>	100.00%	1487	MZ267271	New Reported
		moderate		S18	<i>S. rochei</i>	100.00%	1487	MZ267272	New Reported
moderate		Assiut infected tuber	S19	<i>S. europaeiscabiei</i>	100.00%	1441	MZ267273	(Abdel-Rahman <i>et al.</i> , 2012)	
moderate			S20	<i>S. rochei</i>	100.00%	1487	MZ267274	New Reported	
moderate		El-Minia infected soil	S21	<i>S. rochei</i>	100.00%	1487	MZ267275	New Reported	
moderate			Not selected for further studies						
56		Non-pathogenic	Not selected for further studies						

**Table (2) The relative similarity of the sequence of 16S region of then pathogenic *Streptomyces* isolates sequences obtained from the gene data bank ([www.ncbi.nlm.nih.gov](http://www.ncbi.nlm.nih.gov))**

	S1	S10	S11	S13	S14	S15	S17	S18	S20	S21
S1 ( <i>S. rochei</i> )	100.0									
S10 ( <i>S. rutgersensis</i> )	100.0	100.0								
S11 ( <i>S. lateritius</i> )	98.0	98.0	100.0							
S13 ( <i>S. rochei</i> )	95.7	95.7	96.5	100.0						
S14 ( <i>S. bottropensis</i> )	100	100.0	98.0	95.2	100.0					
S15 ( <i>S. rochei</i> )	95.2	95.2	95.7	100.0	95.2	100.0				
S17 ( <i>S. rochei</i> )	100.0	100.0	98.0	100.0	95.2	100.0	100.0			
S18 ( <i>S. rochei</i> )	100.0	100.0	98.0	100.0	95.2	100.0	100.0	100.0		
S20 ( <i>S. rochei</i> )	100.0	100.0	98.0	100.0	95.2	100.0	100.0	100.0	100.0	
S21 ( <i>S. rochei</i> )	100.0	100.0	98.0	100.0	95.2	100.0	100.0	100.0	100.0	100.0

The phylogenetic tree (Fig. 2A) showed that the 10 strains were clustered in 2 large groups, each group has 2 clusters.

One of the clustered groups included strains S1, S10, S14, S17, S18, S20, and S21, they were all identified molecularly as *S. rochei*. They have identical 16S sequences; however, they were isolated from different sources and locations (Table 1). The pathogenicity in this group (*S. rochei* group) was variable since only S1 isolate caused high pathogenicity, while the other isolates (S10, S14, S17, S18, S20, and S21) were moderate.



**Fig. 2. (A) The dendrogram demonstrates the relationship among the species (10 marked obtained isolates + 8 species used as a reference) based on the 16S rDNA sequencing data. (B) Colony morphology of representatives of the new four species.**



Since *S. rochei* was found in different localities and sources, it confirms its presence and prevalence in Egypt as a new common scab disease causal pathogen. It has been reported that *S. rochei* was isolated and used as a beneficial microorganism for controlling multiple plant diseases (Awad and El-Shahed 2013; Abd El-hafez and Abd El-rahman 2019 and Gebily *et al.* 2021). However, our study confirms that the *S. rochei* strains that were isolated from infested potato soil and infected potato tubes with the common scab disease of potato and also were able to cause symptoms of necrosis on potato slices and symptoms of scab on potato mini tubers. Loria *et al.* (2002) found that the pathogenicity genes of *Streptomyces* could be transferred through horizontal gene transfer, this could explain the emergence of *S. rochei* as a pathogenic strain.

The second cluster includes *S. rutgersensis*. It has been reported by Jung *et al.* (2013) that *S. rutgersensis* has a biological efficacy against the cereal head blight pathogen *Fusarium graminearum* and was successfully used to suppress the disease. But our results confirm for the first time that the genetically identified *S. rutgersensis* that have been isolated from soil collected from fields infected with potato common scab disease can cause scab disease on potato.

The third cluster has *S. lateritius*, this strain was isolated before from the soil-Taif by Sonya *et al.* 2015. Our study indicates that *S. lateritius* was isolated from infested potato soil of common scab disease and also proved to be pathogenic and can cause the symptoms of necrosis on potato slices.

The fourth cluster includes *S. bottropensis*. This strain was considered to be pathogenic and causes the common scab symptoms on potato in Spain and China (Zhou *et al.*, 2017 and Sarwar *et al.*, 2018). But it has never been mentioned to cause the disease in Egypt and in our study, this strain was isolated from diseased potato showing common scab symptoms and this is the first report of it as a pathogenic strain of potato in Egypt.

In this study, each of *S. rutgersensis*, *S. lateritius*, and *S. bottropensis* strains were only found once among the isolates. They were collected from three different governorates (*S. rutgersensis* was isolated from Assiut, *S. lateritius* was isolated from Sohag, while *S. bottropensis* was isolated from El-Minia). With these findings, we can purpose that these strains are rare and spread less in Egypt compared with the *S. rochei*.

Fig. 2B shows the differentiation of the colony morphology among the four new strains, where *S. rochei* (S1) was white, *S. rutgersensis* (S11) was whiteish gray, *S. lateritius* (S13) was darkish gray, and *S. bottropensis* (S15) was whiteish brown

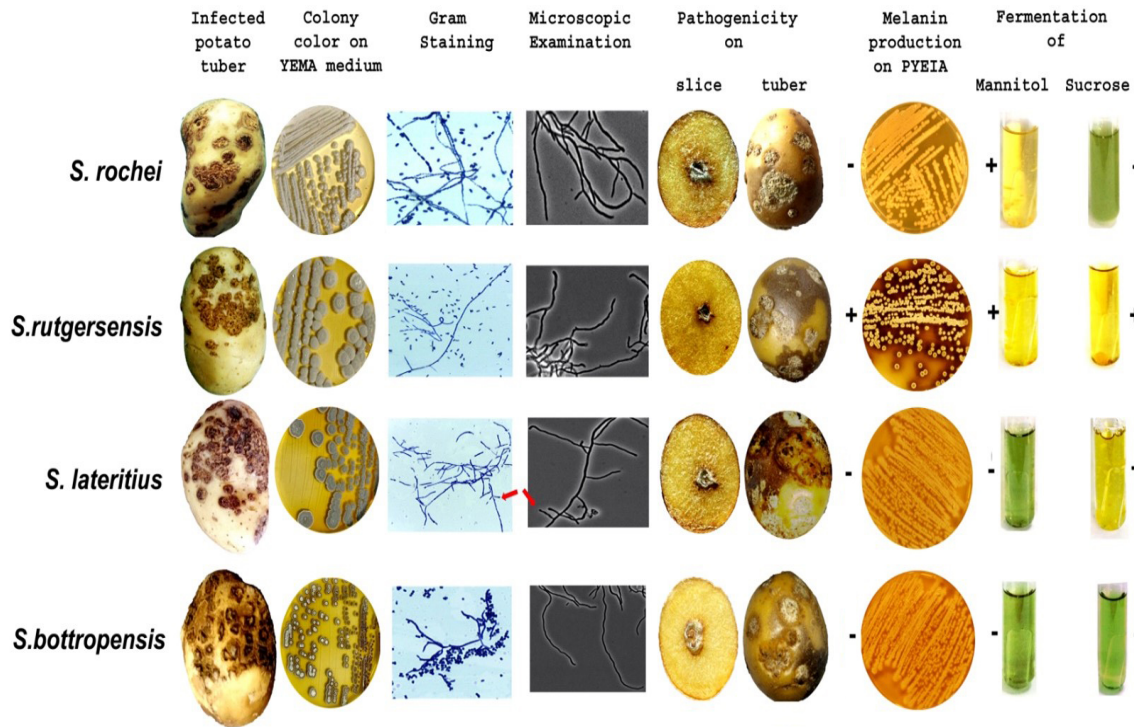
### Morphological and biochemical characteristics of the new reported species

Bergey's Manual of Systematic Bacteriology, Morphological, and Biochemical Characteristics were used to characterize the isolates (Table 3). The results indicated that all tested isolates were sporulating, non-motile, and gram-positive. All tested isolates were positive for; the Gelatin Liquefaction test, Starch Hydrolysis test, Catalase test, H<sub>2</sub>S Production test, Casein Hydrolysis test, and Urea test. All tested isolates were negative for the Levan Production test, M. R. test, V. P. test, Phenylalanine Deiminase test, Nitrate Reduction test, and Potassium Hydroxide test. However, there were variations in certain tests (Fig. 3) showing these variations between the four new species. S1 was chosen to represent the seven strains of *S. rochei*. S1, S11, and S13 were isolated at the beginning from infected tubers showing similar scab symptoms (strain S11 and strain S13 were originally isolated from infected soil of common scab disease and the shown tuber symptoms are the symptoms that were observed in the infected fields that were used for isolation), whereas S15 was isolated from tubers showing different symptoms of scab. The colony color of each species has varied, S1 was white, S11 was whiteish gray, S13 was darkish gray, and S15 was whitish brown, the brown color was related to its ability to produce some kinds of pigments in the media. The microscopic examination and Gram-staining showed that all investigated strains were Gram-positive and showed both mycelial growth with branching hyphae and sporulation in an aerial mycelium, which are typical features of *Streptomyces* spp. S15 had thicker and short mycelium branches comparing to the other three strains. For the Esculin Hydrolysis and Melanin Production on Peptone Yeast Extract Iron Agar, and Tyrosine Agar all the tested isolates were negative except S11, which was positive. For fermentation of the carbon compound, all tested isolates were positive and produced acid from Glucose, Lactose, Fructose, Dextrose, and Maltose. For Mannitol all the tested isolates were positive and produced acid, except S13 and S15 that were negative. For Sucrose all the tested isolates were negative, except S11 and S13 that were positive. These observed variations confirm the differences among the new pathogenic four species of *Streptomyces*.

### Conclusion

The common scab disease is one of the major plant bacterial diseases attacking potato and cause production reduction. In this study, the authors aimed to study the disease and identify potentially new species of the disease. Among the selected isolates, four new pathogenic species of *Streptomyces* were detected.





**Fig. 3. Morphological, microscopic characteristics, pathogenicity test symptoms, and fermentation of mannitol and sucrose by the new pathogenic species of *Streptomyces* causing common scab disease on potato tubers (*S. rochei*, *S. rutgersensis*, *S. lateritius*, and *S. bottropensis*).**

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## ظهور أنواع جديدة ممرضة من الجنس البكتيري *Streptomyces* التي تسبب مرض الجرب العادي على درنات البطاطس في مصر

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### الملخص

تهدف هذه الدراسة إلى تحديد الأنواع الرئيسية من الجنس البكتيري *Streptomyces* القادرة على إصابة نباتات البطاطس بمرض الجرب العادي في مصر. تم جمع درنات البطاطس التي تظهر عليها الأعراض النموذجية لمرض الجرب العادي من ثلاث مناطق مختلفة من صعيد مصر. تم عزل البكتيريا المسببة للمرض، وأجريت الاختبارات المورفولوجية والكيميائية والقدرة المرضية في المعمل لتعريف العزلات مبدئياً. علاوة على ذلك تم استخدام تسلسل جينات S 16 RDNA الجزئي لتعريف العزلات وراثياً. تم تعريف أربعة أنواع جديدة ممرضه لجنس *Streptomyces* لم يتم تسجيلها في مصر قبل ذلك. كانت الأنواع الأربعة هي *Streptomyces rochei* و *S. rutgersensis* و *S. lateritius* و *S. bottropensis*. تعريف العزلات S1 و S10 و S14 و S17 و S18 و S20 و S21 على أنها *Streptomyces rochei*، وتم تعريف العزلات S11 على أنها *S. rutgersensis*، وتم تعريف العزلات S13 على أنها *S. Lateritius* والعزلات S15 تم تعريفها على أنها *S. bottropensis*. وفقاً للبيانات التي تم الحصول عليها، هذا هو التقرير الأول الذي يشير إلى أن الانواع التالية لجنس *Streptomyces*:

*S. rutgersensis* ، *S. Lateritius* ، *S. bottropensis* ، *S. rochei* يسببون مرض جرب البطاطس العادي في مصر. تم إيداع مختلف تسلسلات 16S rDNA التي تم الحصول عليها في NCBI GenBank بأرقام الانضمام التالية: (S1) MZ267260 ، (S10) MZ267264 ، (S14) MZ267268 ، (S17) MZ267271 ، (S18) MZ267272 ، (S20) MZ267274 ، (S21) MZ267275 ، (S11) MZ267265 ، (S13) MZ267267 ، (S15) MZ267269. أظهرت العزلات الأربع التي تم تشخيصها و تعريفها حديثاً اختلافاً في الخصائص المرضية والمورفولوجية والكيميائية الحيوية.