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Characterization of actinophages specific to *Streptomyces deserti* isolated from Sohag Governorate reclaimed soils

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Abstract

The purpose of this study was to isolate and characterize actinophages specific to *Streptomyces deserti*. First, the antifungal activity of *Streptomyces deserti* against *Rhizoctonia solani* was studied. Results indicated that *Streptomyces deserti* inhibited the fungal growth with 62%. Five actinophage isolates specific to *Streptomyces deserti* were successfully isolated from two different locations of reclaimed soil in Sohag Governorate. Electron micrographs of the isolated phages indicated that all the phage isolates appeared to be head-and-tail types. The five phage isolates were designated Φ SD1, Φ SD2, Φ SD3, Φ SD4 and Φ SD5. Phage isolates were characterized using optimum multiplicity and found to be 1.0 PFU/CFU for phage isolates Φ SD1, Φ SD2 and Φ SD3 and 0.5 PFU/CFU for Φ SD4 and Φ SD5. One-step growth curves were studied; all phage isolates had the same infection cycle period; the differences were in the latent period, rise period, and burst size. The thermal inactivation point of each phage isolate was studied. All phage isolates exhibited tolerance to high temperature, since they kept their activity at 100 °C for 10 minutes. The host range of all phage isolates was examined using fourteen *Streptomyces spp.* isolates; the optimum pH for phage infection was found to be pH 7, and the phage isolates are tolerant of alkalinity and sensitive to acidity.

Keywords:

Streptomyces deserti; Actinophage; TEM; *Rhizoctonia solani*.

INTRODUCTION

The widely used broad-spectrum soil fumigant methyl bromide was banned by the Montreal Protocol in 1987 and phased out in most countries by 2005 due to its depleting effect on the ozone layer and the risk of climate change. There is now an urgent need for ecologically compatible and efficient soilborne pathogen suppression techniques in both organic and traditional desert reclaiming (Köberl *et al.*, 2013). Since desertification is universally acknowledged as a severe threat to biodiversity, converting desert soil into fertile, green ecosystems is a global goal. (Köberl *et al.*, 2013) Deserts, being an unknown extreme ecosystem, are known to host various actinobacteria with biotechnology promises (Xie *et al.*, 2021). These harsh environments are a potentially rich source of bacteria that produce new compounds with biological characteristics. Actinobacteria was the most identified bacterial phylum that occurred in reclaimed soils in Egypt and accounted for 28% of all soil microorganisms (Korkar *et al.*, 2022). In addition, *Streptomyces* have a great potential for secondary metabolite production, including antibiotics (MCIntyre, 2002), anthelmintic enzymes, herbicides (Kariminik and Baniyadi, 2010), anti-cancer medicines (Berdy, 2005), growth factors such as vitamin B12 (Bibb, 2005), and immunological modulators (Mann, 2001). *Streptomyces* species create a plethora of specialized metabolites with activities ranging from nutrient availability (e.g., siderophores for iron uptake) to antagonistic activity with other organisms (e.g., antibiotics and antifungal compounds) (Jones *et al.*, 2017). Actinophages are one of the most common organisms that can reduce the effectiveness of *Streptomyces* spp. for plant protection against pathogenic bacteria and fungi because viruses are usually not affected by antibiotics produced from *Streptomyces* strains. (Li *et al.*, 2019). Therefore, isolating and characterizing actinophages will lead to a better knowledge of the nature of this virus, its prevalence, spread, and potential solutions. This study aimed to isolate and characterize actinophages specific to *Streptomyces deserti*. Moreover, the antagonistic activity of *Streptomyces deserti* against plant pathogenic fungus (*Rhizoctonia solani*) that cause many plant

diseases in reclaimed soils in Sohag Governorate was also studied.

MATERIALS AND METHODS

Microorganisms used

Phage indicator bacterium *Streptomyces deserti* isolate was supplied by the Plant Pathology Research Institute, Agriculture Research Center, Giza, Egypt. The source of the plant pathogenic fungus, *Rhizoctonia solani*, used in this study was the Dept. of Plant and Microbiology, Faculty of Science, Sohag University, Sohag, Egypt. Fourteen unidentified *Streptomyces* spp. isolates were obtained from bacterial laboratory stock at the Dept. of Microbiology, Faculty of Agriculture, Sohag University, Sohag, Egypt, and used to determine the host range of phages.

Antagonistic activity of *Streptomyces deserti*

The *in vitro* antagonism of *Streptomyces deserti* against *Rhizoctonia solani* was studied according to the method of Crawford *et al.* (1993). A *Streptomyces* spore suspension (20 µl) (10⁶ spores/ml) was spotted on sides of a PDA plate and incubated at 28°C for 5 days. Mycelium fungus discs (6.0 mm in diameter) were placed on PDA plates after incubation and incubated at 28°C for 5 days. For the control, a fungal disc was placed on a PDA plate without *Streptomyces* sp. Five days after incubation, the levels of inhibition on the radial fungal control and dual culture plates were estimated as described by Chaiarn *et al.*, (2018).

Soil Sample collections

Two soil samples were collected from two different locations in the Sohag Governorate. The samples were gathered from reclaimed soil in Alkawthar and New Sohag experimental farm of the Faculty of Agriculture, Sohag University, Sohag, Egypt.

Actinophage isolation

Isolation of actinophage was carried out according to Pringsulaka, *et al.*, (2004) method with some modifications as follows: 10 ml of fresh liquid culture of *Streptomyces* host and 75 gm of soil samples were placed into 250-ml Erlenmeyer flasks containing 150 ml of Tryptone-yeast-extract (TYE) (5g Tryptone, 3g Yeast Extract) (Shirling and Gottlieb 1966) broth medium and incubated in a rotary shaker at 170 rpm for 72 hours at 35.5 °C. After incubation, the suspension was clarified by

centrifugation at 6000 r.p.m. for 15 min. A fresh *Streptomyces* culture was added to the supernatant and incubated for 72 h at 35°C. Chloroform was added to the supernatant with vigorous shaking, and phages were obtained from the upper layer. The presence of phages was detected using the double-agar layer technique with Tryptone-yeast extract solid medium at pH 7.0. The plates were incubated at 35 °C until a lysed clear zone was formed. The lysed clear zones were picked and placed into separately Eppendorf tubes with 1 ml of Trypton buffer (10g tryptone, 4.3g MgCl₂, 1g gelatin) reported by Fathy (2004) and Maniatis *et al.*, (1982). 200 µl of chloroform were placed in each tube, which was then stored at 4 °C.

Phage purification

Single phage particles were purified using a soft top agar method that described by Bertani and Weigle (1953) with minor modifications. 5 mL of soft top agar (NB with 7 g/L agar) was prewarmed at 48 °C, mixed with 100 µL of 10¹⁰ cfu/mL *streptomyces deserti* culture and 100 µl of supernatant produced after actinophage enrichment, and then poured onto a solidified TYE plates. The plates were incubated at 28°C for 24-48 hours after the medium had solidified. To obtain pure actinophage isolates, individual plaques were collected from agar plates using a sterile inoculation loop and the technique was repeated four times. Plaques were collected after the fourth purification, reconstituted in 2 mL of sterile tryptone buffer, and vigorously shaken for 30 minutes at 200 rpm to release the attached viral particles, centrifugated at 4000 rpm, and kept at 4 °C for further analysis. For each bacteriophage, phage concentrations (plaque-forming units /mL) were estimated using method of Toyama *et al.* (1983) and Czajkowski *et al.* (2014).

Characterization of Actinophage

For the characterization of five actinophage isolates specific for *S. deserti*, different studies were conducted.

Electron microscopy

Phage particle morphology was examined according to El-Tarabily *et al.* (1995) and Caruso, *et al.*, (2019) with following modifications: 8 ml of high-titre phage (2.5 ×10⁹ pfu/ml) was concentrated at ultracentrifuge at (25,000 rpm for 2 hour in Beckman Type 30 rotor), and the pellet was resuspended in 0.5 ml of tryptone buffer .

After concentration, each phage isolate was placed on a TEM farmavar-coated copper grid and stained with 5% w/v uranyl acetate at pH 4.5. A TEM (transmission electron microscope; Joel 100 CCXL) was used to examine the samples at Assiut University in Assiut, Egypt.

Determination of optimal multiplicity of infection (MOI) and phage propagation

With slight modifications, the multiplicity of infection (MOI) of phage isolates was determined according to the method by Abdelrhim *et al.*, (2021) *S.deserti* was cultivated to a concentration of 110 CFU/ml in TYE agar media at 35 °C, as determined by dilution plating on TYE broth medium and counting the resulting CFUs (Czajkowski *et al.* 2014). *S. deserti* (110 CFU/ml) was infected for 24 hours at 35 °C with five different phage isolates MOI s (about 0.1, 0.5, 1.0, 2.0, and 4.0 PFU/CFU). The experiment was done three times to find the best MOI of every phage isolate for achieving the maximum phage titer (PFU/ml). The phage isolates were propagated by adding it at a MOI of 1.0 to 200 ml of 112 *S. deserti* culture with CFU/ml. The mixture was shaken at 200 rpm for 24 hours at 35 °C. After clearing the phage suspension by centrifugation at 4000 rpm for 20 minutes, the supernatant was preserved at 4 °C.

One-step growth determination

One-step growth curve

One-step growth curve experiments were carried out as described by Czajkowski (2014) with minor modification to determine the latent time and phage burst size. One ml of phage suspension (10¹⁰ pfu/ml) was mixed with 1 ml of exponential phase culture of *S. deserti* (10⁸ cfu/ml) and incubated at 35°C for 5 min for phage adsorption. The mixture was centrifuged at 10000 r.p.m. for 10 min to remove free phage particles (unadsorped particles). The pellet was resuspended in 60 ml of TYE broth medium, and the culture was continuously incubated at 35°C. Samples were taken at 5 min intervals up to 100 min., and phage titer was determined. Burst sizes of phages were calculated by dividing the phage titers at plateau phase by the initial phage titers (Jiang *et al.*, 1998).

Determined Host range of phage isolates

Plates of double agar layers were prepared as above described, with fourteen isolates of *Streptomyces spp.* bacteria (S1, S2, S3,... to

S14), which were isolated from various regions of the Sohag Governorate and used as the indicator bacteria. Drops of every single phage isolate were applied to the surface of every plate. Plates were checked for clear zones at the locations where the drops had been placed after 24 to 72 hours of incubation. (Sadik, *et al.*, 2014).

Determination of optimum pH values

100 ul of a suspension of the test phages was added to Eppendorf tubes each containing 1 ml of tryptone buffer with various values of pH from 4 up to 10 were prepared, pH 6.5 was used as control (phage /tube). Tubes were incubated at room temperature for 3 hours. A plaque assay was used to estimate the effect of pH on phage isolates. Czajkowski *et al.*, (2012).

Determination of thermal inactivation point

Eppendorf tubes were prepared, each containing 1 ml of MOI-propagated phage from a single phage isolate. temperatures of 40, 50, 60, 70, 80, 90, and 100 °C were applied to each tube in the water bath for 10 minutes, then fast cooled with running tap water. A spot test was used to determine the infectivity of phage at each heat treatment as described by Hammad *et al.* (2018), Othman and Nasr-Eldin (2020)

RESULTS

Streptomyces deserti antagonistic activity

The antifungal activity of *S. deserti* against *Rhizoctonia solani* was studied using the dual culture method, and the inhibition percentage was found to be 62%. *Streptomyces deserti* isolate inhibited the growth of tested fungi, demonstrating antagonistic ability.

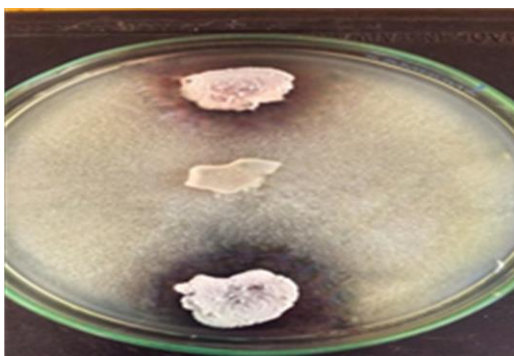


Figure 1: Antifungal activity of *Streptomyces deserti* against *Rhizoctonia solani* using a dual

Soil Sample locations

Two soil samples were collected from two different locations in the Sohag Governorate's reclaimed soil. As shown in table 1, the samples were collected from the east and the west of Sohag Governorate.

Table 1: GPS locations of soil samples:

sample	GPS Location	Location description
1	26°35'27.4"N 31°48'01.9"E	New Sohag experimental farm of the faculty of agriculture, Sohag University, Sohag, Egypt.
2	26°28'07.5"N 31°40'10.7"E	Alkawthar experimental farm of the faculty of agriculture, Sohag University, Sohag, Egypt.

Detection of *Streptomyces deserti* actinophage in soil samples

Streptomyces deserti actinophages were successfully detected in soil samples that were collected from reclaimed soils near the deserts. The clear zones of spot tests for phage detection that were observed in Fig. 2 indicated that the phages of *S. deserti* were found to be common in this reclaimed soil.

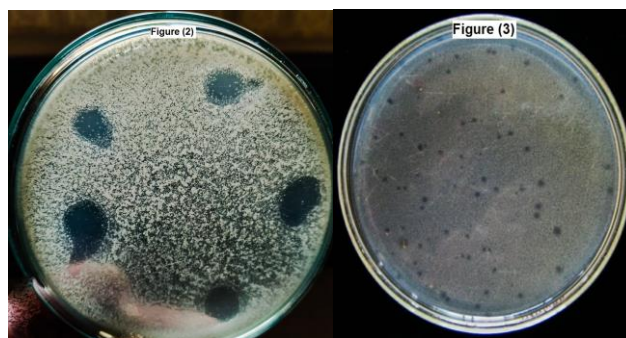


Figure 2: Phage-clear zones that lyse the *Streptomyces deserti* bacterial lawn.

Figure 3: The difference in morphology between the single plaques of *S. deserti* actinophage

Phage purification

Five *S. deserti* phage isolates were isolated from five single plaques that varied in size and morphology and were found to have a hazy and/or

clear circular shape of 0.5–1 μm in diameter as shown in Fig. (3). Phage numbers 1, 2, and 3 were isolated from soil samples collected from the first location; phage numbers 4 and 5 were isolated from the second one (table 1).

Electron microscopy

The morphology of all five *S. deserti* specific-phage isolates was examined by transmission electron microscopy (TEM), and negative staining was used to show details of phage particles. According to electron microscope photographs (Fig. 4), all phage particles were head-and-tail types, with icosahedral heads that varied in their dimensions and non-contractile tails of different lengths. The morphology of phage isolates number 1,2 and 3 matches the specifications of the family *Siphoviridae*. Phage Isolates number 4 and 5 matches the specifications of the family *Podoviridae*. Table (2) shows the variation in dimension measurements of the head and tail of every phage.

Table 2: Dimensions* of *Streptomyces deserti*-specific phage particles.

phage	Head Diameter SD (nm)	Tail	
		Width SD (nm)	Length SD (nm)
1	75 \pm 3	11 \pm 3	223 \pm 1
2	98 \pm 2	17 \pm 3	178 \pm 2
3	83 \pm 1	13 \pm 3	83 \pm 2
4	66 \pm 2	15 \pm 3	71 \pm 3
5	177 \pm 2	46 \pm 2	104 \pm 2

SD= standard deviation.

According to the difference in dimensions the five phage Isolates of *S. deserti* were designated Φ SD1, Φ SD2, Φ SD3, Φ SD4 and Φ SD5 for phage numbers 1, 2, 3, 4, and 5, respectively.

Optimal multiplicity of infection (MOI)

The MOI for phage isolates Φ SD1, Φ SD2 and Φ SD3 were found to be 1.0 PFU/CFU among the five tested ratios phage isolates. Φ SD4 and Φ SD5 phage isolates MOI was 0.5 PFU/CFU, resulting in the highest phage titer of 2.3×10^9 PFU/ml for the phage Φ SD2, followed by Φ SD1 with an MOI of 1.6×10^9 PFU/ml, then Φ SD4 with 1.3×10^9 PFU/ml, then Φ SD5 with 1.2×10^8 PFU/ml, and Φ SD3 with 2.1×10^8 PFU/ml. High titre phage suspensions were prepared and stored at 4°C with chloroform to be used at further studied.

Table 3: Multiplicity of infection for *S. deserti* specific actinophages.

Phage Isolates	Multiplicity of infection (PFU/CFU)				
	0.1	0.5	1.0	2.0	4.0
	Plaque forming unit /ml				
Φ SD1	1.3×10^7	2.1×10^7	1.6×10^9	0.2×10^8	3.1×10^7
Φ SD2	0.1×10^7	1.0×10^7	2.3×10^9	1.2×10^8	2.2×10^7
Φ SD3	1.1×10^7	1.2×10^7	2.1×10^8	2.4×10^7	1.7×10^7
Φ SD4	1.1×10^7	1.3×10^9	1.6×10^7	2.3×10^8	2.6×10^6
Φ SD5	1.1×10^7	1.2×10^9	1.1×10^7	2.7×10^7	3.5×10^6

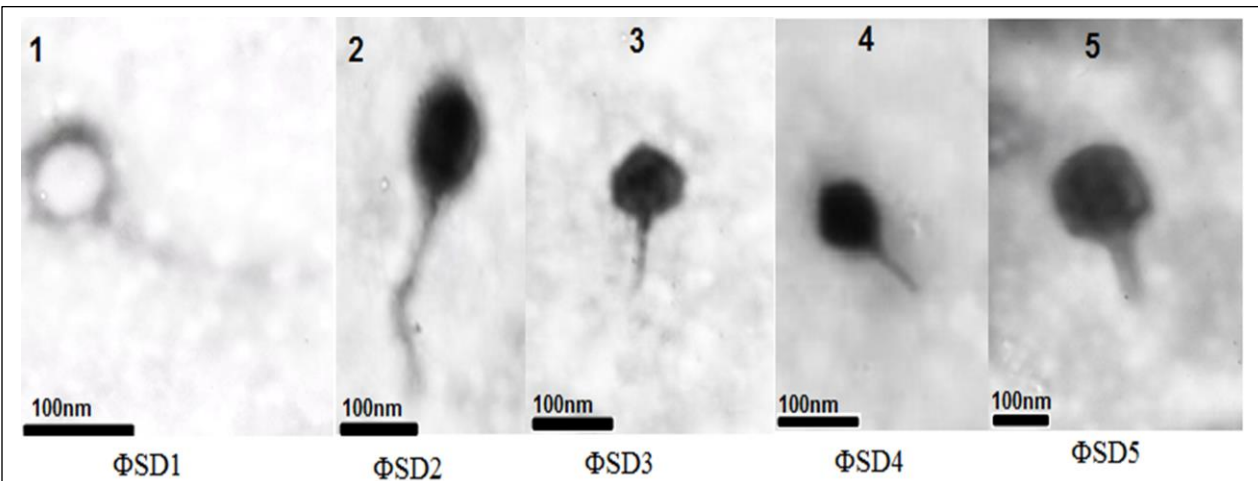


Figure 4: Transmission electron micrographs of *S. deserti* displaying phage particle morphologies.

One step growth curve

To estimate the latent time and burst size, one-step growth curves for isolated actinophages were designated (Fig. 3). The infection cycle of all five phage isolates was determined to be 80 minutes, with a latent period of approximately 40 min. for phage Φ SD1, 30 min. for phages Φ SD2 and Φ SD5, 20 min. for Φ SD3 and Φ SD4 phage isolates. The rise period of all phages ranged from 40 to 60 minutes, with a burst size of around 180 to 200 PFU per infected cell for all phage isolates (Fig. 3).

Thermal activation points of *S. deserti*-specific actinophages

Temperatures of 40, 50, 60, and 100 °C were applied to phage isolates in studies on the effect of temperature on five phage isolates to determine thermal activation points. In this study, we found that all phage isolates were tolerant to all temperature degrees that were applied. The five phage isolates kept their activities at temperatures ranging from 40 to 100 °C for 10 minutes each.

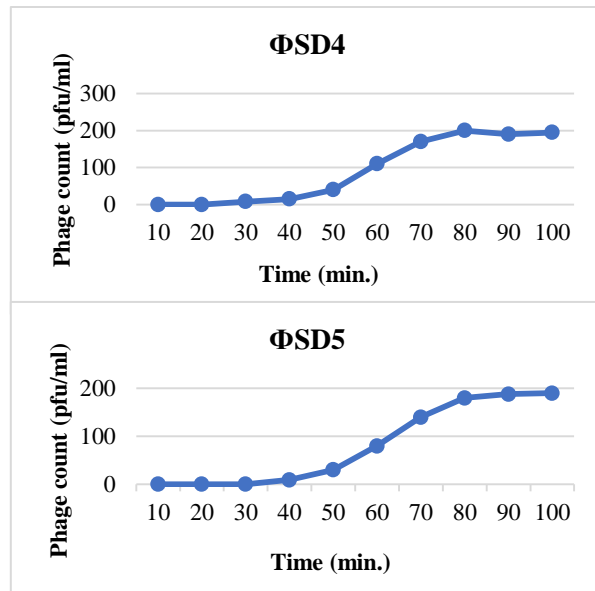
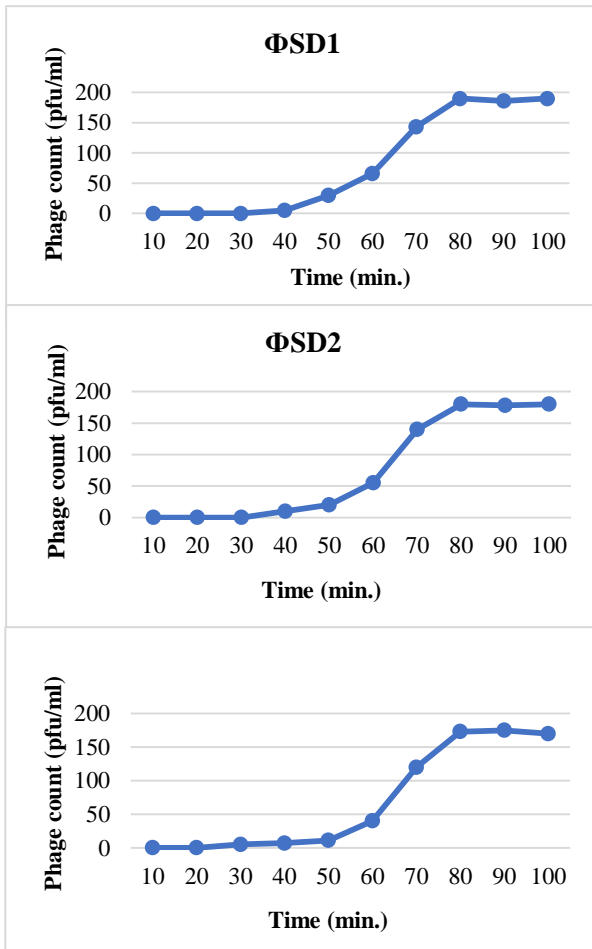


Figure 5: One-step growth curves for *S. deserti*-specific actinophages Φ SD1, Φ SD2, Φ SD3, Φ SD4 and Φ SD5.

Host range of *S. deserti* actinophages

As presented in Table 4, fourteen *Streptomyces spp.* isolates obtained from various regions of the Sohag Governorate were used to determine the host range of phages. None of the phage isolates infected S12 or S14 *Streptomyces* isolates. Phage Φ SD1, Φ SD2, and Φ SD3 formed lysed clear zone on 12 isolates from S1 to S11 and S13. Phage Φ SD4 infected all strains except S1, in addition to S12 and S14. Phage Φ SD5 infected all strains except S8 and S11 in addition to S12 and S14.

Table 4: Host range of actinophage isolates specific for *S. deserti*.

Bacterial hosts	Actinophage Isolates				
	Φ SD1	Φ SD2	Φ SD3	Φ SD4	Φ SD5
S1	+	+	+	-	+
S2	+	+	+	+	+
S3	+	+	+	+	+
S4	+	+	+	+	+
S5	+	+	+	+	+
S6	+	+	+	+	+
S7	+	+	+	+	+
S8	+	+	+	+	-
S9	+	+	+	+	+
S10	+	+	+	+	+
S11	+	+	+	+	-
S12	-	-	-	-	-
S13	+	+	+	+	+
S14	-	-	-	-	-

+ = Lysis; - = No lysis

Effect of pH on actinophage infectivity

The effect of pH and the optimum pH for phage isolate infectivity were investigated using different pH levels. As shown in Table 5, for all five phage isolates, the highest pfu/ml of phage particles was at pH 7, and the lowest active phage particles were at pH 4. At pH 8, phage activity for all isolates was slightly reduced compared to pH 7 but was still very high. The reduction in pfu/mL of all isolates continued at pH 9 and pH 8 but was still very high when compared to pH 4 and pH 5.

Table 5: Effect of pH on actinophages specific for *S. deserti*.

Phage Isolates	pH levels						
	4	5	6	7	8	9	10
	Phage forming unit /ml						
ΦSD1	2.3×10 ³	3.3×10 ⁴	1.8×10 ⁷	2.1×10 ⁸	2.0×10 ⁷	2.1×10 ⁷	1.8×10 ⁵
ΦSD2	2.1×10 ²	2.6×10 ⁴	2.2×10 ⁷	2.4×10 ⁸	1.9×10 ⁷	1.3×10 ⁷	1.6×10 ⁵
ΦSD3	0	1.8×10 ⁴	2.2×10 ⁷	2.8×10 ⁸	2.3×10 ⁷	1.6×10 ⁷	2.1×10 ⁵
ΦSD4	1.5×10 ²	1.1×10 ⁴	1.7×10 ⁷	2.3×10 ⁸	1.9×10 ⁷	1.4×10 ⁷	2.7×10 ⁵
ΦSD5	1.1×10 ²	1.3×10 ⁴	1.1×10 ⁷	2.1×10 ⁸	1.8×10 ⁷	1.6×10 ⁷	1.1×10 ⁵

DISCUSSION

The harsh desert conditions are a potentially rich source of bacteria that create new metabolites with bioactivity. *S. deserti* is one of the microorganisms that can live in these severe conditions and can be used in the biocontrol of harmful microorganisms such as *Rhizoctonia solani* that cause plant diseases in reclaimed soils (El-Hadidy 2018 and Santhanam *et al.*, 2012). The antifungal activity of *S. deserti* against *R. solani* was studied, and it shows that the bacteria can affect the fungus's growth with about 62% inhibition. Therefore, it can be said that this strain of *S. deserti* can produce bioactive compounds and can be used in biocontrol for plant diseases caused by *R. solani*. Similarly, Köberl *et al.*, (2013) found high antagonistic activity from *Streptomyces* strains against *R. solani* that were isolated from the Egyptian desert and reclaimed soils. Bacteriophage

is an important factor in the soil microorganism society that inhibits the activity of beneficial bacteria such as *streptomyces sp.* In this study, bacteriophages specific for *S. deserti* were successfully isolated from reclaimed soil in two different locations of the Sohag Governorate. Such results showed that phages of *S. deserti* were found to be common in this kind of soil that suffers from harsh desert conditions like high temperature, aridity, salinity, and alkalinity. Abdelrhim *et al.* (2021) isolated, new phages infecting *Streptomyces scabies* from Egyptian soils. In addition, two phages specific to *Streptomyces griseoflavus* were isolated from soil and characterized by Othman *et al.* (2008). Five single plaques were isolated and purified, with some consideration given to differences in plaque morphology and soil original source. The isolated plaques were hazy and/or clear, with a circular shape of 0.5–1 mm in diameter. Plaque morphology is the first criterion that was used in the characterization and differentiation of phage isolates by many researchers, such as Fathy (2008); Farahat (2016) and Hammad (2018). Negative staining was used to examine the *S. deserti*-specific phage isolates under an electron microscope. Transmission electron micrographs showed that all phage isolates (ΦSD1, ΦSD2, ΦSD3, ΦSD4 and ΦSD5) were head-and-tail type with differences in their dimensions. That morphology and dimensions of phages ΦSD1, ΦSD2 and ΦSD3 matches the specifications of the family *Siphoviridae* (McCorquodale 1999). ΦSD4 and ΦSD5 phage isolates matches specifications of the family *podoviridae* (Molineux 1999). That difference in morphology of phage isolates may refer to the difference in locations that the phages were isolated from, as the phages were isolated from two different locations. To characterize the phage growth, Optimal multiplicity of infection (MOI) was determined for all phage isolates. The best MOI for all phage isolates was found to be 1.0 PFU/CFU among the five tested ratios. MOI was studied by Czajkowski *et al.*, (2014) for bacteriophages infecting *Dickeya spp.* and found to be between 1.0 and 0.01 for different phage isolates. The optimal multiplicity of infection for phage isolates was then used to determine the one-step growth curve. The one-step growth curve of phage isolates was studied. All phage isolates had

the same infection cycle period; the differences were in the latent period, rising period, and burst size. However, the results of the growth curve are still close, which makes us wonder whether the morphological form of viruses is not enough to differentiate between phage isolates. This question is answered by Francki (1973), who noted that because there are a variety of unknown factors that can impact particle size throughout various preparative techniques, reliable comparisons across published morphometric data are challenging. Additional characterizations of the isolated phages were performed to confirm this explanation. Therefore, thermal inactivation points for phage isolates was studied. Surprisingly, all phage isolates are tolerant to high temperature and still active at 100 °C for 10 minutes. This is an unusual result for soilborne phages, but it was predictable because the source of the phage isolate was a hot, dry environment in the Upper Egypt desert and reclaimed soils. Furthermore, the indicator bacteria (*S. deserti*) that lives in hot deserts is highly temperature tolerant (Santhanam *et al* 2012). Jończyk *et al.* (2011) reported the influence of external factors on bacteriophages and found phage isolates stable at high temperatures (up to 97 °C). Fourteen isolates of *Streptomyces spp.* were used to determine the host range of the *S. deserti* phage isolates. ΦSD1, ΦSD2 and ΦSD3 phage isolates infected the same strains of *Streptomyces spp.* ΦSD4 and ΦSD5 phage isolates participated in infecting some isolates of *Streptomyces spp.* and differed in others. This result indicates that the host range of phage isolates cannot be used as a single method to differentiate between phage isolates. Kankila *et al.*, (1994) used 32 Rhizobium strains to investigate the host range of 11 bacteriophages specific to *R. leguminosarum* bv *trifolii*. They concluded that three morphological types of phage isolates were detected. Optimum pH of *S. deserti* specific phage isolates was studied. The optimum pH of all phage isolates was at pH 7. In addition, *S. deserti* specific phage isolates were found to be sensitive to acidity; therefore, the infectivity of all phage isolates was dropped to the lowest level (zero PFU/ml for ΦSD3 phage isolate). At pH 8 and pH 9, the pfu of all phage isolates was still very high and found to be very active against indicator bacteria. The alkaline tolerance of phage isolates refers to the alkalinity of the soil that was

the origin of the phage isolate, and the indicator bacteria are well adapted to the alkaline conditions, which give rapid growth to these bacteria and consequently provide appropriate conditions for phage multiplication. Similarly, Pringsulaka *et al.* (2004) and Sykes *et al.* (1981) studied the effect of pH on actinophages isolated from soil samples.

CONCLUSION

The findings suggest that bacteriophage differentiation based on TEM may be insufficiently sensitive to accurately analyses the systematic relationships of closely related bacteriophages. There is no single method for categorizing and identifying bacteriophages. Isolating and characterizing actinophages will lead to a better knowledge of the nature of these viruses, their prevalence, spread, and potential solutions. Characterization and taxonomy of bacteriophages are expected to be secondary priorities in terms of practical phage for agricultural needs.

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الملخص العربي

توصيف الأكتينوفاجات المتخصصة على بكتريا الأستربتوميسيس ديزيرتاي التي تم عزلها من أراضي مستصلحة في محافظة سوهاج

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الهدف من هذه الدراسة هو عزل وتوصيف الأكتينوفاجات المتخصصة على بكتريا الأستربتوميسيس ديزيرتاي، وفيها تم دراسة التضاد بين بكتريا الأستربتوميسيس ديزيرتاي وفطر الريزوكتونيا سولاني حيث وجد أن البكتريا تثبت نمو الفطر بنسبة 62%. وبنجاح تم عزل خمس عزلات من الأكتينوفاج المتخصص على الأستربتوميسيس ديزيرتاي من موقعين مختلفين لأراضي مستصلحة بمحافظة سوهاج. باستخدام الميكروسكوب الإلكتروني النافذ تم دراسة الشكل المورفولوجي للأكتينوفاجات المعزولة ووجد أن جميع الفاجات الخمسة التي تم عزلها من النوع ذو الرأس والذيل وتم تسمية هذه الفاجات طبقاً للاختلاف في حجم جزيء الفاج والشكل المورفولوجي له إلى (Φ SD1، Φ SD2، Φ SD3، Φ SD4 و Φ SD5). تم توصيف الأكتينوفاجات المعزولة بمعرفة معدل التلقيح المثالي لعملية التضاعف ووجد أنه 1.0 جزيء فاج لكل خلية بكتيرية للفاجات Φ SD1 و Φ SD2، و 0.5 جزيء فاج لكل خلية بكتيرية للفاجات Φ SD4 و Φ SD5. في دراسة منحنى النمو ذو الخطوة الواحدة وجد أن جميع الفاجات لها نفس وقت ذروة الإصابة، وكانت الاختلافات في مرحلة الكمون ومرحلة صعود المنحنى وحجم الانفجار. تم دراسة درجة التثبيط الحراري للخمس فاجات ووجد أنها تتحمل درجات الحرارة المرتفعة حتى 100 درجة مئوية لمدة عشر دقائق. باستخدام 14 عزلة من الأستربتوميسيس تم دراسة المدى العوائلي للخمس فاجات. بعد دراسة تركيز أيون الهيدروجين المناسب لنشاط الفاجات وجد أنه عند تركيز 7، وجميع العزلات كانت مقاومة للقوية وحساسة للحموضة.