FACULTY OF AGRICULTURE

Minia J. of Agric. Res. & Develop.
Vol. (44), No. 3 , pp.337-354, 2024

ISOLATION AND CHARACTERIZATION OF *PAENIBACILLUS POLYMYXA* BACTERIUM ASSOCIATED WITH SUGAR BEET ROTTED ROOTS

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Received : 27 May 2024 Accepted: 10 June 2024

ABSTRACT

Sugar beet plants are attacked by several diseases. The most serious diseases that causing post-harvest decay of sugar beet storage roots. Root rot affect sugar beet yield and quality. Various fungi and bacteria are reacted as root rotting incidents. Isolation trails resulted 8 infective bacterial isolates. Three bacterial isolates (B5, B6, B10) were the most virulent as root rotting. Morphological and biochemical properties of the most virulent bacterial isolates showed that they are typically reacted as *Paenibacillus polymyxa*. In addition, molecular identification of the most virulent isolate B5 confirmed it's identification as *P. polymyxa*. The obtained isolate B5 was rotted potato tuber slices. Response of sugar beet genotypes was various to *P. polymyxa* Infection. Sugar beet genotype BTS185 reacted as the most susceptible, while genotype Husam was the least susceptible to *P. polymyxa* infection. Furthermore, the tested isolate of *P. polymyxa* was able to infect storage roots of turnip, carrot and sweet potato beside garlic claves.

Keywords: *Beta vulgaris*, *Paenibacillus polymyxa*, potato, soft rotting bacterium.

INTRODUCTION

The sugar beet (*Beta vulgaris* L, *Chenopodiaceae*) is an industrial crop commercially grown as a hybrid. From it's root considered as the plant compound of interest. It is grown in temperate regions for sugar and biofuel production **(Glenn *et. al.*,1999)**. Recently, global sugar beet production reached more than 4,610,000 million hectares grown through 56 different countries **(Anonymous, 2017)**. It occupies the second after sugar cane production, annually yielded about 40%

of total sugar production all over the world **(El-Hag *et al*., 2015).** In Egypt sugar beet yields about 2.305 million ton of sugar (1.100, 1.050, and 0.155 million ton from sugar cane, sugar beet and sweet sorghum, respectively) and consumes about 3.100 million ton (74.35%), that means about 0.795-million-ton sugar (25.65%) is important annually from foreign countries **(C.C.S.C., 2017).**

Sugar beet is susceptible to many fugal (over 20) and bacterial (over 10) pathogens often caused foliar symptoms **(Harveson *et al*., 2009)**, that caused substantial yield losses. The genus *Paenibacillus* has been recognized from the genus *Bacillus* based on molecular identification of rRNA group 3 bacilli **(Ash *et al*., 1993)**. *P. polymyxa* is a synonym of *Bacillus polymyxa* **(Ash *et al*., 1994)**. Several researches of plant infections by *Bacillus* (*Paenibacillus*) species are rather unusual (**Mikicinski *et al*., 2010)** studies bacterial isolates 15M and 16M from calla lily tubers showing soft rot in commercial farms in Poland. However, various *Bacillus* species have been reported as phytopathogens to several hosts such as garlic cloves rot **(Galal *et al*., 2002)**, potato soft rot **(Bathily *et al*., 2010)**, peach blotch **(Saleh *et al*., 1997)**, Scot wet wood disorder **(Kovaleva *et al*., 2015)**, ginger rhizome rot **(Peng *et al*., 2013)**, mango leaf blight **(Galal *et al*., 2006)**, muskmelon fruit rot **(Song *et al*., 2018),** rubber trunk bulges **(Mazlan *et al*., 2019). Yahyaoui *et al*. (2023)** reported that *Bacillus pumilus* and *Paenibacillus amyloliticus* are potato soft rotting bacteria in Tunisia.

This study aimed to (1) isolate and identify the agent pathogen(s) from sugar beet rotted roots in El-Minya governorate. (2) test their pathogenicity, (3) test the response of some cultivars of sugar beet to the pathogen and (4) study their host range.

MATERIALS AND METHODS

1-Collecting infected sugar beet root samples:

Natural infected sugar beet (*Beta vulgaris* L. cv. PTS185) roots were collected from private commercial farms in Qasr Hour (Mallawey district) and Jarris (Abou Qurqas district), El-Minya governorate, during spring 2020. Roots were brought to the laboratory of Department of Plant Pathology, Faculty of Agriculture, Minia University. The collected roots showed dark black streaks running up the petioles in the centers of crowns, and wilt if roots become severely affected. Root symptoms vary from soft rot to dry rot and vascular bundles in roots become dark brown and necrotic (Fig. 1: A1, A2).

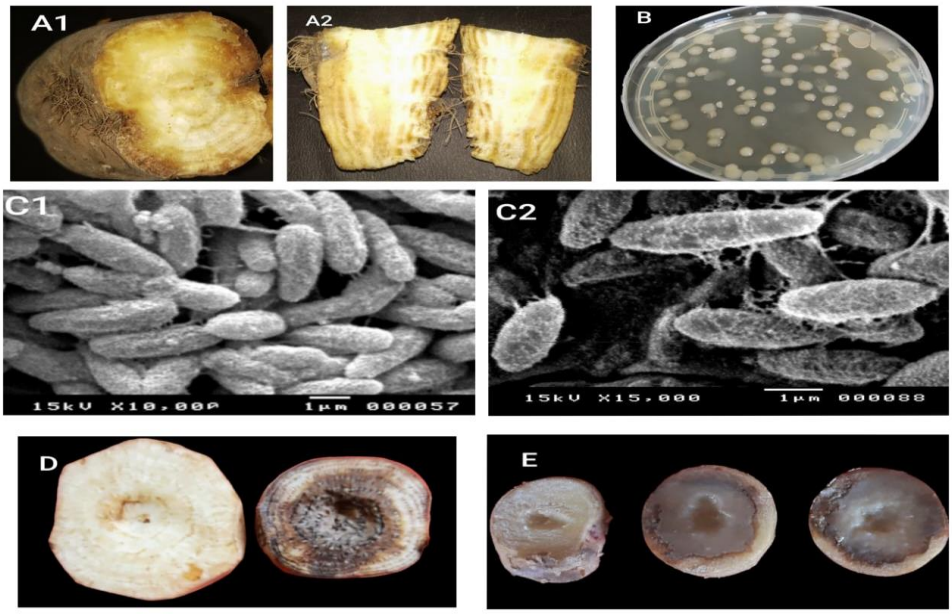

Fig. 1. Natural infection of A1, A2= Sugar beet root that showed symptoms, B= bacterial culture 48h old, C1 and C2= Scanning electron microscope (SEM) of isolate B5 (Rod shaped, 3.5–5.7µm in length and 0.61–1.1µm in width) and D= artificial infection on sugar beet root slices, E= Potato tuber slices).

2-Isolation and purification of bacteria

Sugar beet diseased roots after they have been washed with tap water, surface sterilized with 0.3% sodium hypochlorite for 2 minutes and thoroughly washed three times with sterile distilled water (SDW), were cut aseptically, to pieces (3-mm) and macerated for 90 seconds in 3ml SDW using a sterile mortar and pestle. The macerated tissue was left about 20 minutes then one bacterial loopful was streaked onto nutrient glucose agar (NGA) medium in Petri plates (9cm diameter). To obtain pure bacterial isolates (Fig. 1: B), single colonies of bacteria were chosen and re-subcultures at least 2 times on NGA medium **(García-González *et al*., 2018)**.

3- Pathogenicity tests

Fourteen isolates, *e. g*., B1 to B14, were isolated from sugar beet roots, and subjected for pathogenicity.

3-1- Preparation of bacterial inoculum and inoculation

Cultures of isolated bacteria, were grown on (NGA) medium, incubated at 25^{o}C for 48 hours. Healthy apparently sugar beet (cv. PTS185) roots were washed under tap water, air dried, dipped in sodium hypochlorite (0.3%) for 2 minutes, washed three times using SDW then air dried. Each root was sliced 8-10 mm thick slices. Subsequently, hole (3.6 mm diameter, 5 mm depth) for each slice

were made using a sterile cork borer, then placed on moist, sterile filter paper in Petri plates. For inoculation, 50 µl of bacterial suspension of each isolate (10^8 CFU ml^{-1}) was added to each hole in the sugar beet slice. Six slices / isolate were tested as replicates. The check slices were treated with SDW, then the slices were incubated at 25°C for 5 days. Appearance of soft rot symptoms was a sign of infection. The bacterium was reisolated from the rotted slices using NGA medium. A second inoculation was carried out using the reisolated bacteria to confirm pathogenicity (Fig. 1: D).

3-2- Disease assessment:

Disease severity (DS, %) was determined, 5 days after inoculation, by measuring the weight (g) of the infected tissues according to the following equation:

$$\text{Disease severity (DS, \%)} = \frac{\text{Average weight (g) of rotted tissues}}{\text{The whole weight of the slice}} \times 100$$

4-Pectolytic activity on potato tuber slices:

Three bacterial isolates i.e., B5, B6 and B10 which gave the most virulent were selected to test their ability to secret pectolytic enzymes**.** Potato tubers (cv. Diamond) after the have been washed with tap water, air dried, then dipped into 96% ethanol, flamed and cut into 10 mm thick slices. Subsequently, hole (3.6 mm diameter, 5 mm depth) per slice were made in the center of potato slice using a sterile cork borer. Potato slices were then transferred on moist, sterile filter paper in Petri plaits for singly inoculation similar as described in the pathogenic isolates, of each isolate in each hole in the potato slices. The plates were incubated at 25^{o}C for 5 days after inoculation, they were examined daily being getting soft rot symptoms (Fig. 1: E). The weight of the rotted area was determined, and disease severity was calculated as above mentioned.

5- Identification of the bacterial pathogenic isolates:

5-1- Morphological characterization:

The morphology characters were studied using nutrient glucose (1%) agar (NGA), nutrient sucrose (5%) agar (NSA), potato sucrose (5%) agar, and bouillon glucose, and King B media, spore formation and cell morphology (**Bradbury, 1988 and Schaad *et al*., 2001**).

5-2- Biochemical and physiological characterization:

The keys of **Bradbury (1988), Buchanan and Gibbons (1974), Holt *et al* (1994), Schaad *et al*. (2001)** were helped to identify the three bacterial isolates (B5, B6 and B10) depending on their cultural, morphological, and physiological characters according to the following tests:

Gram staining was determined according to **Bergey and Holt (1994),** production of fluorescent pigment **(King *et al*., 1954),** oxidase reaction **(Kovacs, 1956)**, catalase activity (**Schaad *et al*., 2001**), oxidative/fermentative metabolism of glucose **(Hugh and Leifson, 1953**), Levan test using sucrose, nitrate reduction, arginine dihydrolase activity, ability to produce acid from glucose and lactose **(Lelliott and Stead, 1987),** gelatine, soluble starch hydrolysis and acetoin production from glucose (Voges

Proskauer test) **(Fahy and Persley, 1983).**

5-3- Molecular techniques:

The most pathogenic isolate B5 was subjected to PCR through phylogenic analysis. The PCR technique was done as follows: **DNA extraction amplification and sequencing of 16S-rDNA**:

Extraction of total DNA was conducted according to **Llop *et al*., (1999).** The amplification of 16S rRNA fragments from bacterial strains was carried out using the 27F (5′ AGAGTTTGATCMTGGCTCAG3′) and 1492R (5′ GGTTACCTTGTTACGACTT3′) primers **(Lane, 1991).** The PCR program was as follows: an initial denaturation step of 95°C for 15 min, 30 cycles of denaturation step at 95°C for 20s, annealing step at 50°C for 40s, and extension step at 72°C for 90s and a final step of 72°C for 5 min. The PCR outputs were watched using UV light after electrophoresis in a 1% (w/v) agarose gels stained with ethidium bromide. The PCR outputs were cleaned and sequenced at the sequencing service unite of Macrogen Inc. (Macrogen Inc., Seoul, Korea). The cleaned DNA was edited, and aligned with BioEdit software (http://www.mbio.ncsu.edu/Bioedit/bioedit). The obtained cleaned DNA sequences were compared with bacterial species previously deposited in the GenBank database.

6- Response of some sugar beet genotypes to the bacterial pathogen:

Fourteen genotypes of sugar beet, *viz.* Oscar poly, Clavius, Pleno, BTS 185, Husam, Tarbelli, BTS 435, Karima, Farida, BTS 105, Sahar, Euklid, Perikles, and Dreeman, were inoculated with isolate B5 of the isolated pathogen to test its response to the pathogen. Inoculum preparation, inoculation and disease assessment were carried out as described before in the pathogenicity test.

7- Response of some different plant species to the bacterial pathogen (host range):

Onion (bulbs cv. Giza 6), garlic (cloves cv. sids 40), stored roots of sweet potato (cv. O'Henry), carrot (cv. Chantenay), turnip (cv. Balady) and radish (cv. Balady) were tested for their response to *P. polymyxa* infection under laboratory conditions. For inoculation, 50 µl of 10^8 CFU ml^{-1} of bacterial suspension of isolate (B5) was injected in plant organs and disease assessment were conducted as described before.

8- Statistical analysis:

Standard error for means (SEM) was used to test the differences between treatments **(Gomez and Gomez, 1984).**

RESULTS:

1-Pathogenicity tests

Among 14 bacterial isolates, 8 bacterial isolates were infective to cause rot sugar beet root tissues (Table 1). Isolate B5 gave the most rot severity (88.39%), followed by isolates B10 and B6 which induced 77.57% and 70.39%, respectively. The moderate infection was explored by isolate B2 (40.91%) and B11 (30.16%). The least rot severity was provided by isolate B12 (3.40%), B8 (4.97%) and B1 (13.05%). Six isolates B3, B4, B7, B9, B13 and B14 were failed to incite any symptoms rot to sugar beet root tissues.

Table 1: Infectability of bacterial isolates associated with sugar beet rotted roots to root slices of sugar beet cv. BTS185. Disease severity was evaluated 5 days after inculcation and incubation at 25◦C.

isolate	Disease severity, %		Mean ± SEM
	Exp I	Exp II	
B1	16.80	9.30	13.05 ± 3.75*
B2	45.36	36.45	40.91 ± 4.46
B3	0.00	0.00	0.00 ± 0.00
B4	0.00	0.00	0.00 ± 0.00
B5	85.16	91.62	88.39 ± 3.23
B6	74.83	65.95	70.39 ± 4.44
B7	0.00	0.00	0.00 ± 0.00
B8	6.71	3.22	4.97 ± 1.75
B9	0.00	0.00	0.00 ± 0.00
B10	74.61	80.53	77.57 ± 2.96
B11	24.88	35.43	30.16 ± 5.10
B12	2.30	4.50	3.40 ± 1.10
B13	0.00	0.00	0.00 ± 0.00
B14	0.00	0.00	0.00 ± 0.00

*****Data are means of tow experiments ± standard error for means (SEM).

2-Pectolytic activity:

Data (Table 2) showed that all isolates tested were able to incite soft rot on the potato slices. However, isolates differed as regards the severity of soft rot symptoms. Isolate B5 was the most aggressive one caused 86.27% disease severity; it caused the severest symptoms upon potato slices inoculation. Isolates B6 and B10 could be regarded as moderately pathogenic ones, which caused between 76.1 and 79.6% disease severity.

Table 2: Rot severity on potato slices (cv. Diamond) caused by singly inoculated with bacterial isolates B5, B6 and B10. Disease severity was evaluated 5 days after inculcation and incubation at 25◦C.

Isolates	Disease severity, %		Mean ± SEM	
	Exp I	Exp II		
B5	84.24	88.30	86.27 ± 2.03*	A
B6	73.70	78.50	76.10 ± 2.40	B
B10	81.55	77.65	79.60 ± 1.95	AB

*****Data are means of two experiments ± standard error for means (SEM).

3- Morphological characterization:

The colonies cultured on NGA plate characterized as small, milky white, opaque colonies with irregular edges (Fig. 1: B). Colonies surface was smooth and mucoid, viscose, semi- transparent. While it was white-yellow color produced pulvinate, mucoid, viscous, semi-translucent, circular colonies with a regular edge on NSA medium.

4- Physiological and biochemical characteristics:

Results (Table 3)revealed the three isolates were identical in the reported morphological, biochemical and physiological characters, were positive in (Gram reaction, rod shape, spore forming, flagellation, airobiosis, tolerance of NaCl 3%, growth on potato slices, Levan test, gelatin liquification, starch hydrolysis, catalase, VP test, casein hydrolysis, galactose, d-fructose, d-xylose, nitrate reduction), were weakly positive (utilization of carbon source, glucose, lactose, d-maltose, l- arabinose, glycerol), were negative in (hypersensitive reaction, citrate utilization, indole production, fluorescent pigment, oxidase, arginine dihydrolase). Were that there are slight differences between the three tested isolates, except of their pathogenicity, which showed that the isolate B5 was the most aggressive one than the others.

Table 3: The reported morphological, biochemical and physiological characters of the three isolates B5 (*Paenibacillus polymyxa*), with those of the isolated pathogens B6 and B10.

Test	Isolates			*Paenibacillus polymyxa* reported by:	
	B5	B6	B10	Mikicinski *et al.*, 2010	Zhai *et al.*, 2021
Size	0.9-x4.6µm			0.8-1 x 4-10 µm	
Gram reaction	+*	+	+	+	±
Shape	rod shaped	rod shaped	rod shaped	Bacilliform cells	rod-shaped
Spore presence	+	+	+	+	
Motility	motile	motile	motile		
Airobiosis	+	+	+		anaerobic growth
Tolerance of NaCl 3%	+	+	+		+
Growth on potato slices	+	+	+	+	
Levan test	+	+	+	+	
Gelatin liquification	+	+	+	+	
Starch hydrolysis	+	+	+	+	+
Hypersensitive reaction	-	-	-	-	
Catalase	+	+	+	+	
VP (Voges-Proskauer) test	+	+	+	+	
Casein hydrolysis	+	+	+		+
Citrate utilization	-	-	-		-
Indole production	-	-	-		-
Utilization of carbon source: Glucose	±			+ but not produce gas	
Lactose	±				
D-Maltose	±			+	
Galactose,	±	±	±		±
D-fructose,	±	±	±		±
D-xylose	±	±	±		±
L- Arabinose	±				±
Glycerol	±				
Nitrate reduction	+	+	+	+	±
Fluorescent pigment	-	-	-	-	
Oxidase	-	-	-	-	
Arginine dihydrolase	-	-	-	-	

*+ = Positive reaction, - = negative reaction and ± = weak reaction.

Molecular identification:

The 16S rRNA gene was cloned and sequenced. The partial sequences of the isolated bacterium compared to the sequences using the BLAST search program in NCBI's GenBank, that exhibited high correlations with the genes of *Paenibacillus* species. The phylogenetic tree was constructed through MEGA 11 application with the partial sequences of 16S rRNA of B5 isolate and other typical strains of *Paenibacillus* spp. (Fig.1: C1, C2).

Paenibacillus polymyxa (IsB5) was identified as the isolate exhibiting high activity of sugar beet root rot based on partial nucleotide sequencing of the 16S rRNA gene. However, based on DNA sequence analysis (Fig. 2) of the 16S rDNA gene of strain HX-140 (GenBank accession number PP763473, and on the combined morphological and physiological, strain B5 was identified as *P. polymyxa*.

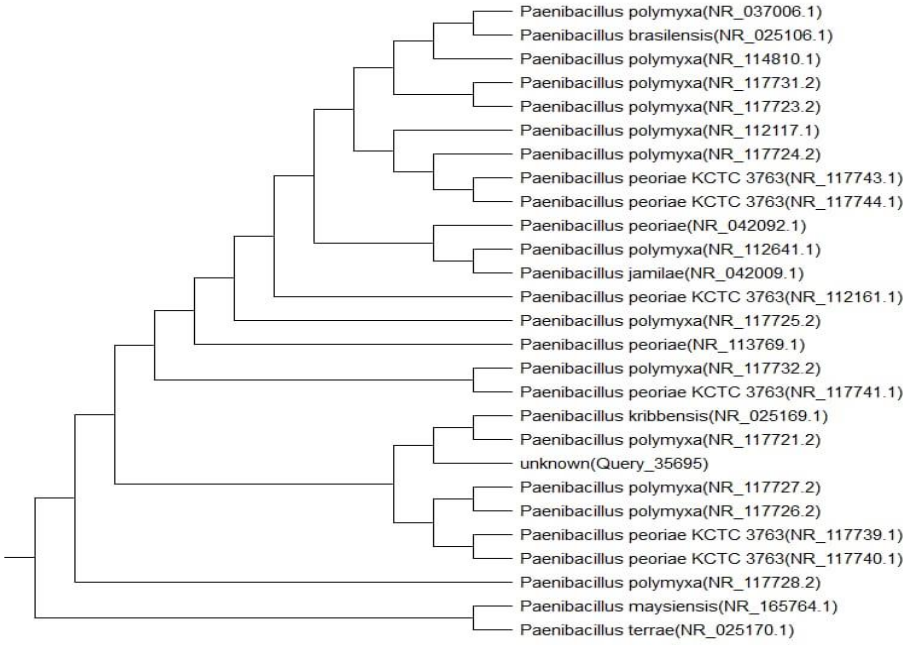

Fig. 2: Phylogenetic analysis based on 16S rRNA sequences of strain B5 and closely related species constructed by the neighbor-joining method. GenBank accession numbers analyzed here are shown in parentheses.

Response of some sugar beet genotypes to *Paenibacillus polymyxa* (Is. B5) infection:

Fourteen of sugar beet genotypes were tested in their response to infection with *P. polymyxa*. Data in (Fig 3, Table 4) pointed to ability of *P. polymyxa*, isolate B**5,** to infect all tested sugar beet genotypes inducing soft rot symptoms and showing different degrees of disease severity. The genotype BTS 185 showed the highest percentage of disease severity (89.4), followed by Oscar poly, Clavius, Perikles, BTS 105, Karima, Tarbelli, BTS 435, Dreeman, and Euklid, of DS% was ranged between 70.4 - 80.0%. Whereas, the genotypes Farida, Pleno, Sahar and Husam were the lowest infected genotypes showing 69.2 - 61.0% of DS%.

Table 4: Ability of *P. polymyxa*, to infect roots of sugar beet genotypes. Disease rating on root slices was evaluated 5 days after inoculation under laboratory conditions.

Genotypes	Disease severity (%)		Mean ± SEM
	Exp I	Exp II	
Oscar poly	68.2	72.6	70.40 ± 2.20*
Clavius	71.0	75.6	73.30 ± 2.30
Pleno	66.5	69.8	68.15 ± 1.65
BTS 185	87.2	91.6	89.40 ± 2.20
Husam	63.3	58.7	61.00 ± 2.30
Tarbelli	70.0	73.2	71.60 ± 1.60
BTS 435	65.9	71.0	68.50 ± 2.60
Karima	69.8	72.3	71.50 ± 1.30
Farida	71.1	67.3	69.20 ± 1.90
BTS 105	68.5	74.1	71.30 ± 2.80
Sahar	66.4	62.6	64.50 ± 1.90
Euklid	76.7	83.3	80.00 ± 3.30
Perikles	80.2	76.0	78.10 ± 2.10
Dreeman	71.7	77.1	74.40 ± 2.70

*****Data are means of two experiments ± standard error for means (SEM).

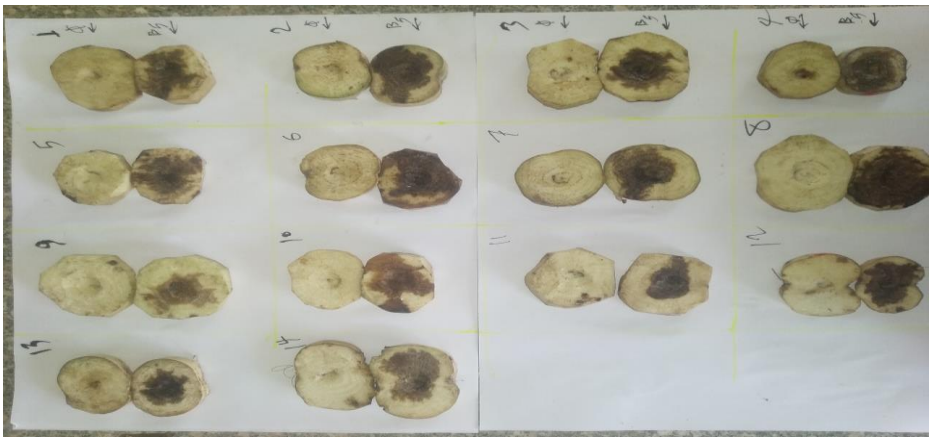

Fig 3: Sugar beet root slices 14 genotypes as influenced by *P. polymyxa* isolate B5 infection.

3 -Response of some different plant species to the bacterial pathogen (host range):

Storage roots were tested for their response to infection with *P. polymyxa* (isolate B5) under laboratory conditions. Data in Table (5) showed that infectivity of *P. polymyxa* was varied with plant species. The bacterial isolate was infected roots of potato, carrot, sweet potato, turnip, and garlic cloves, showing soft rot symptoms. Contrary, roots of radish plus onion bulbs were not infected by the bacterial isolate, showing resistance to the tested pathogen isolate.

Table 5: Ability of *P. polymyxa,* to infect of some different plant species:

Common name	***Scientific name***	***Disease severity, %***	***Mean ± S.E.M***	**Mean ± S.E.M**
		Exp I	Exp II	
Radish	*Raphanus sativus* (cv. Balady)	0.0	0.0	0.0 ± 0.0*
Turnip	*Brassica rapa var. rapa* (cv. Balady)	25.2	31.2	28.2 ± 3.0
Carrot	*Daucus carota* subsp. *Sativus* (cv. Chantenay)	63.0	69.4	66.2 ± 3.2
Sweet Potato	*Ipompea batatas* (cv. O'Henry)	64.1	59.3	61.7 ± 2.4
Garlic	*Allium sativum* (cv. sids 40)	38.2	43.6	40.9 ± 2.7
Onion	*Allium cepa* (cv. Giza 6)	0.0	0.0	0.0 ± 0.0

*****Data are means of two experiments ± standard error for means (SEM).

DISCUSSION

Various plant disease control approaches, along with different agronomic practices used in contemporary agriculture, outlets improper consequences, such as misleading biodiversity and other natural resources **(Lucas, 2011; Gonthier *et al.* 2014)**, environmental destruction **(Enserink *et al.* 2013)**, and invoked evolution in pathogens **(Zhan and McDonald, 2013**). The present study concerned sugar beet root rot that affected its root quality and yield.

Fourteen bacterial isolates were isolated from sugar beet rotted roots. There pathogenicity showed 3 bacterial isolates were the most infective i.e., B5, B6 and B10 while 6 bacterial isolates were not pathogen bacteria. Accordingly, the most virulent isolates were selected for further studies, these isolates that caused the most rot severity to sugar beet root tissue were expressed their ability to rot potato slices. Data indicated that isolates B5, B6 and B10 were able to produce pectolytic enzymes, subsequently, morphological, physiological and biochemical activities were examined to identify these bacterial isolates. The obtained data showed that the tested isolates were typically reacted as were positively reacted in (Gram reaction, rod shape, spore forming, motile, airobiosis, tolerance of NaCl 3%, growth on potato slices, Levan test, gelatin liquification, starch hydrolysis, catalase, VP test, casein hydrolysis, galactose, d-fructose, d-xylose, nitrate reduction), were weakly positive (utilization of carbon source, glucose, lactose, d-maltose, l- arabinose, glycerol), were negatively reacted in (hypersensitive reaction, citrate utilization, indole production, fluorescent pigment, oxidase, arginine dihydrolase).

Based on their reaction, isolates could be identified as *Paenibacillus polymyxa* **(Mikicinski *et al*., 2010; Zhai *et al.*, 2021)**. To confirm the identification, DNA profile of isolate B5 was made and it's phylogenic analysis was assured that isolate B5 is *Paenibacillus polymyxa* (GenBank accession number PP763473).

The genus *Paenibacillus* has been derived from the genus *Bacillus* based on molecular identification for rRNA group 3 bacilli **(Ash *et al.*, 1993)**. Studies of plant infections by *Bacillus* (*Paenibacillus*) species are rather rare. **Fahy and Persley (1983)** demonstrated, that bacteria of this genus were known to be weakly pathogenic to potato tubers as soft rotting. Many *Bacillus* spp. is pectolytic and have been caused soft rot. Such species of *Bacillus i.e. polymyxa*, *macerans* (presently *P. polymyxa*, *P. macerans*), *circulans*, *subtilis* and *cereus*. Meanwhile, only *P. polymyxa* is generally recognized as soft rotting **(Lelliott and Stead, 1987)**. Bacteria of this species were isolated from vegetables e.g., onion and cabbage, exhibiting substantial problems under conditions **(Bradbury, 1986)**. However, it has been reported that *P. polymyxa* is the causal agent of mushroom bacterial pit **(Lelliott and Stead, 1987)**. This bacterium causes numerous necrotic spots on stems, cotyledons and rarely on tomato seedlings roots **(Caruso *et al*., 1984)**. Moreover, this bacterium elicits a characteristic rot on ginseng plants **(Jeon and Kim, 2008)**. Plant- associated *Bacillus* spp. is also provided as

saprotrophes and/or endophytes **(Holt *et al.,* 1994)**.

As for sugar beet genotypes reaction to *P. polymyxa* infection, two of 14 genotypes are the most susceptible BTS185 and Euklid. Ather sugar beet genotypes were infected by *P. polymyxa and* reacted as moderate infected, meanwhile genotype Husam gave the list rot severity value (61%).

The genus *Paenibacillus* enclosesubiquitous strains more than 200 species of notable suitable to plants, humans, animals or the environment, that have been isolated from highly diverse habitats, e.g., polar regions, tropics, aquatic ecosystems or even deserts. Due to the ability of this bacterium to produced proteases, lipases and phospholipases, and to grow at low temperatures, *Paenibacillus* spp is contribute to the food spoilage development (**Grady *et al.,* 2016**). In the present study, we have explored that concerned *Paenibacillus polymyxa*. strains are able to cause disease symptoms on potato tuber slices and sugar beet root slices and that the vast majority of this isolate exhibits the activity of plant cell wall-degrading enzymes, **(Van der Wolf, J.M *et al.,* 2001; Yahyaoui *et al.,* 2023**).

The verlans potential of *Paenibacillus* spp. is unusually enhanced as, to the best of our knowledge, there are only three articles on the isolation of *Paenibacillus* spp. strains from diseased plants, i.e., wheat and barley, showing leaf deformation, wilting, spotting, strokes and stripes **(Slovareva, 2020)**. In addition, *Solanum lycopersicum*, *Alium cepa*, *Pelargonium* sp., *Lilium* sp., *Begonia obliqua*, *Anthurium* sp., *Dianthus caryophyllus* and *Hypericum perforatum* in Poland in 2013–2019 (**Zenelt, *et al.,* 2001**). **Yahyaoui *et al.* (2023)** reported that potato soft rot symptoms in potato tubers were exhibited In vitro after artificial inoculation of two potato cultivars (‘Lilly’ and ‘Spunta’), the pathogenicity of *Paenibacillus* spp. strains should be taken into consideration and further investigated.

The present work indicated that *P. polymyxa* was able to infect turnip, carrot and sweet potato roots, causing root rot. Moreover *P. polymyxa* reacted as rot incitant to garlic cloves.

In conclusion rod shaped, spore forming and Gram positive bacteria are associated with sugar beet root rot. The biochemical and physiological identification indicted that three bacterial isolates were typically to *paenibacillus polymyxa*. Molecular identification was confirmed that isolate B5 is *P. polymyxa*. Results showed *P. polymyxa* was able to rot potato slices and all sugar beet genotypes tested. Otherwise, *P. polymyxa* has ability to rot roots of turnip, carrot and sweet potato beside garlic claves.

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الملخص العربي

عزل وتوصيف بكتيريا *Paenibacillus polymyxa* المصاحبة لعفن جذور بنجر السكر

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تصاب نباتات بنجر السكر بالعديد من الامراض وأخطر هذه الامراض التي تكون مستمرة بعد الحصاد اعفان الجذور مما يؤثر على المحصول في الإنتاجية والجودة. ويسبب عفن جذور بنجر السكر فطريات وبكتيريا مختلفة. ونتج عن عملية العزل من جذور بنجر مصابة بالعفن ثمانية عزلات بكتيرية ممرضة، وكانت ثلاثة عزلات منها (B5، B6، B10) هي الأكثر خطورة في احداث عفن للجذور المخزنة. أظهرت الاختبارات المورفولوجية والكيموحيوية للعزلات البكتيرية الأكثر قدرة امراضية أنها تتفاعل بصورة متطابقة للبكتيريا *Paenibacillus polymyxa* اكد التعريف الجزيئي للعزلة B5 وكانت الاعلى قدرة امراضية انها بكتيريا *P. Polymyxa* أظهرت النتائج المتحصل عليها ان العزلة B5 ذات قدرة على تعفن شرائح البطاطس، وشرائح جذور جميع التراكيب الوراثية لبنجر السكر المختبرة، وكان التركيب الوراثي BTS185 اكثر التراكيب الوراثية قابلية للاصابة بينما التركيب الوراثي حسام كان اقلهم قابلية للاصابة. علاوة على ذلك أظهرت البكتيريا قدرة على احداث عفن للجذور المخزنة لكل من اللفت والجزر والبطاطا بجانب فصوص الثوم .