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 Pakistan Journal of Biotechnology
 (PJB)
 (P-ISSN: 1812-1837 and E-ISSN: 2312-7791)



RHIZOBACTERIA AS ANTAGONIST AGAINST *Fusarium oxysporum* CAUSING TOMATO WILT

Noor Muhammad Shah¹, Syed Zulfiqar Ali¹, Muhammad Waris^{1*}, Atta Ullah², Zobia Jabeen¹,
 Basheer Ahmed¹, Sana Shazia Jiskani³,

¹Department of Plant Pathology, Balochistan Agriculture College Quetta, Pakistan

²Department of Plant Breeding and Genetics, Balochistan Agriculture College Quetta

³Department of Horticulture, Sindh Agriculture University Tandojam, Sindh

*Corresponding email: waris.faqir@gmail.com

Article Received 14-10-2023, Article Revised 02-01-2024, Article Accepted 03-01-2024.

ABSTRACT

Tomatoes (*Lycopersicon esculentum* L.) are the most extensively grown vegetable in the world. It belongs to the Solanaceae family and is commonly planted for its tasty fruits. Pests, weeds, diseases, and parasites are just a few of the numerous variables that significantly affect tomato growth and yield. The most common disease affecting tomatoes is fusarium wilt. Fifteen rhizobacterial strains were identified by morphological and biochemical analyses in this work, and they were employed as an antagonist against *Fusarium oxysporum* f.sp. *lycopersici*. Fifteen isolates were investigated for their antagonistic properties against *Fusarium oxysporum* f.sp. *lycopersici* using an invitro dual culture approach. The growth of *Fusarium oxysporum* f.sp. *lycopersici* was suppressed by each isolate. Out of the 15 rhizobacteria isolates, isolate RBS-5 exhibited the highest level of growth inhibition and strongly suppressed the development of *Fusarium oxysporum* f.sp. *lycopersici*, resulting in a 57.28 percent reduction in pathogen growth as compared to the control. The development of *Fusarium oxysporum* f.sp. *lycopersici* was suppressed by isolates RBS-12, RBS-6, and RBS-15, in decreasing order of merit, compared to the control by 53.7, 51.91, and 51.73 percent. Isolate RBS-13 showed the least amount of pathogen growth inhibition 20.83 percent. The data was statistically analyzed.

Key words. Tomato, Fusarium wilt, Rhizobacteria, Antagonist, Dual culture

INTRODUCTION

Tomatoes are members of the nightshade family (Solanaceae). It is an extremely important and well-known vegetable that is grown all over the world. It provides enough nutritional benefits and is competitively priced when compared to other vegetables. It is a fantastic source of vitamins A and C. Together with pigments like lycopene and β -carotene, it also contains minerals like phosphorus and iron. Tomatoes get their red colour from lycopene, which is essential for the production of β -carotene. (Chohan & Ahmad, 2008; Kumar *et al.*, 2012). The tomato is a notable genetically identified diploid (2n) plant species with a relatively short life cycle, high fertility, and the capacity to procreate, which makes it an excellent subject for practical research. (Moyers *et al.*, 2018). Tomatoes are a heavily farmed and eaten vegetable worldwide. It is a major profitable crop for small farmers in tropical Asia. (Saavedra *et al.*, 2016). China is the global leader in tomato production. The tomato originated in America, and its original planting method was developed in Mexico. Tomatoes were brought to the subcontinent by British soldiers at the beginning of the 1800s. The first documented history of tomato

cultivation in the Indo-Pak Subcontinent is recorded in William Roxburgh's 1832 book "Flora Indica." (Saavedra *et al.*, 2016). Tomatoes are highly valued as a crop in Pakistan. It is the main ingredient in salads and is cooked with other vegetables to enhance their taste. Tomato cultivation has been greatly impacted by the surge in demand for tomato-based products like purees and ketchup brought on by the consumption of fast food. Tomato demand is growing, and this trend is likely to continue. Tomatoes provide 20% of the daily needed amount of vitamin A based on a 2000 calorie diet. A tomato supplies 26% of the recommended daily intake of vitamin C. (Ali *et al.*, 2017). Among other limitations, diseases are the main factor affecting tomato yield and quality. (Pritesh & Subramanian, 2011). Anthracnose, bacterial canker, blights, fungal and bacterial wilts, and tomato spotted wilt are the most common tomato ailments. (Jones *et al.*, 2014). One of the most common causes of losses in greenhouses and fields worldwide is Fusarium wilt. (Sheu *et al.*, 2006; Abdel-Monaim *et al.*, 2011). Crop losses from the disease can vary from 10% to 80% and perhaps 100% when it strikes tomato varieties that have not been modified. (Bharat & Sharma, 2014; Worku & Sahe, 2018). Tomato crops are susceptible

to several diseases, including fungus, which can result in substantial damage, substantial financial losses, and low fruit production. (Lichtenzveig *et al.*, 2006). *F. oxysporum* f. sp. *lycopersici* is an extremely dangerous soil-borne pathogen that may persist and proliferate in the soil for a long time. Fusarium wilt is a tomato disease that is widely dispersed and has a substantial commercial impact worldwide. (Abdesselem *et al.*, 2016). Secondary metabolite production frequently happens concurrently with antagonistic PGPR action (Sliva *et al.*, 2001). The most common antagonistic action method is direct physical contact with phytopathogens and the biocontrol agent (Mukerji & Chincholkar, 2007).

MATERIALS AND METHODS

Collection of Samples for *Fusarium oxysporum*

Isolation: The diseased tomato root, stem and soil sample was collected from chashma achozai, Quetta, Balochistan. These samples were brought into Plant Pathology laboratory of Balochistan Agriculture College and stored at room temperature for observation of different pathological aspects.

Collection of samples for Rhizobacteria Isolation:

For isolation of Rhizobacteria soil samples were collected from maize field in chashma achozai, Quetta, Balochistan. The samples were brought into Plant Pathology laboratory of Balochistan Agriculture College and stored at room temperature for observation of different Rhizobacteria isolates.

Preparation of Media

Preparation of PDA from commercial powder: For preparation of 1 liter of media added 39g commercial powder in 600ml of water and mix it on hot starrer plate, add 400ml of water to make 1000ml/ 1 liter of media. For sterilization of media, autoclave was used for 15 minutes at 121°C.

Preparation of NA Media: For 1 liter of media add 28g commercial powder in 600ml of water and mix it on hot starrer plate, add 400ml of water to make 1000ml/ 1 liter of media. For sterilization of media, autoclave was used for 15 minutes at 121°C.

Isolation and Purification of *Fusarium oxysporum*:

The wilt disease-causing *Fusarium oxysporum* f.sp. *lycopersici* was isolated from tomato plants that were afflicted and exhibiting wilt symptoms. Samples that were infected were gathered from Quetta's tomato-growing fields. After chopping the infected roots into tiny pieces and surface sterilising them for 20 to 30 seconds with a 1% sodium hypochlorite (NaOCl) solution, the infected roots were thoroughly cleaned with sterilised distilled water. After plating three pieces on Potato Dextrose Agar (PDA) medium, they were cultured for five to seven days at 28 °C ± 2 °C. The fungal material was isolated using the single hyphal tip method (Rangaswami, 1972), stored at 25 °C, and utilised for subsequent experimental investigations and identification. Refrigerated at 4°C were the pure culture tubes. To identify the culture,

employed morphological and microscopic investigation.

Morphological characterization of *Fusarium*:

oxysporum: The initial step in the morphological characterisation method was to use a microscope and human eye to examine the characteristics of culture plates. Using PDA plates, the morphological properties of fungal colonies were investigated. The characteristics of the colony were studied using a stereoscope. The morphological features of chlamydo spores, macroconidia, and microconidia were investigated under a compound microscope. The fungal isolate's morphological identity was determined by using the procedure outlined by Leslie *et al.* (2006). The microscopic features that were looked at in order to identify the fungal pathogen were the colony's size, color, and appearance; the forms of the macro- and microconidia; and the chlamydo spores.

Isolation and Purification of Rhizobacteria:

Rhizospheric soil that was taken from the root zone was used to isolate rhizobacteria. In this investigation, a soil sample was obtained from Quetta's field regions where maize is grown, and rhizobacteria were identified using the serial dilution method.

Serial Dilution Method: The rhizobacteria were recovered from rhizospheric soil by serial dilution method (Wollum, 1982). To remove the rhizosphere soil from the root zone, the roots were gently shaken and then submerged in sterile water in the laboratory. Soil samples were put in a test tube with 9 mL of distilled water, and the mixture was vortexed to homogenize it. Take 1 mL of the first dilution (10⁻¹) and transfer it to a fresh clean tube along with 9 mL of diluent to create a second dilution. As a result, the solution was diluted up to 10⁻⁸. Each time, the solution was thoroughly mixed using a vortex mixer. After a thorough shaking, 10⁻⁶, 10⁻⁷, and 10⁻⁸ dilutions were evenly distributed on a Petri plate filled with solidified nutritional agar (NA) medium. Petri plates that were taped were incubated at 26 ± 2 °C.

Biochemical Characterization of Rhizobacteria:

The biochemical characterization of each isolate was mostly finished in compliance with the procedure outlined in (Joseph, Patra, & Lawrence, 2007).

Gram staining: Procedure performed as prescribed by Vincent & Humphrey (1970).

Catalase Test: Catalase is an enzyme that breaks down hydrogen peroxide (H₂O₂) into oxygen (O₂) and water (H₂O) was performed as Ninama *et al.* (2012).

Starch Hydrolysis Test: Starch agar medium is used in this experiment. Pure bacterial cultures were streaked on Petri plates filled with solidified starch agar medium, and the plates were incubated for 24 hours as instructed by Cappuccino & Sherman (1983).

Potassium Hydroxide (KOH) Solubility Test: A loopful of bacterial culture was collected on a clear, dry glass slide, and then mixed with a drop of 3% potassium hydroxide until an even solution was

formed for the KOH test, as per Kirsop & Doyle (1991).

Ammonia production: Ammonia was measured in rhizobacterial strains using peptone water, as per Cappuccino & Sherman's (1992) methodology. Fresh rhizobacterial cultures were placed in test tubes with 10 mL of peptone water, and they were incubated for two days at 28°C. The test tubes turned brown to orange, indicating that the appropriate rhizobacteria had started to create ammonia when 0.5 ml of Nessler's reagent was added to each one.

Salt and PH Tolerance : The ability of the isolated rhizobacterial isolates to grow in different concentration of salt was tested by streaking them on NA medium containing 1.0%, 3.0% and 5% (wt/v) NaCl. Differences in pH tolerance was tested by adjusting the pH to 6.5, 7.5, and 8.0. All the plates incubated at 28°C for 72 hours and NA medium plates was used as controls.

Dual Culture Method (Dennis & Webster.,1971): A nine-millimeter culture disc was removed from the periphery of a seven-day-old *F. oxysporum* f.sp. *lycopersici* culture and positioned about 75 millimetres away from the edge of the Petri dish containing 15 millilitres of sterile, solidified PDA media. The two-day-old Rhizobacterial strains and the

pathogenic culture were evenly spaced apart on the medium and faintly streaked with it. The zone of inhibition (mm) and the mycelial growth of *F. oxysporum* f.sp. *lycopersici* were measured. The pathogen's growth was impeded during the process of choosing the most potent antagonists. The percentage of mycelia growth inhibition was computed according to Vincent (1947).

$$\text{By using Formula } I = \frac{C-T}{C} \times 100$$

Where, I = Percent inhibition over control
C = Radial growth (mm) in Control
T = Radial growth (mm) in Treatment

RESULTS AND DISCUSSION

Morphological Identification of *Fusarium oxysporum* f.sp. *lycopersici*: Microscopically, *Fusarium oxysporum* f.sp. *lycopersici* has been identified based on characteristics of the colony and spore shape as described by Leslie *et al.* (2006). The mycelium that developed on PDA plates had cottony growth and was hyaline. Microscopic examination revealed septate and branching hyphae. Branched conidiophores hosted microconidia that ranged in shape from straight to bent.

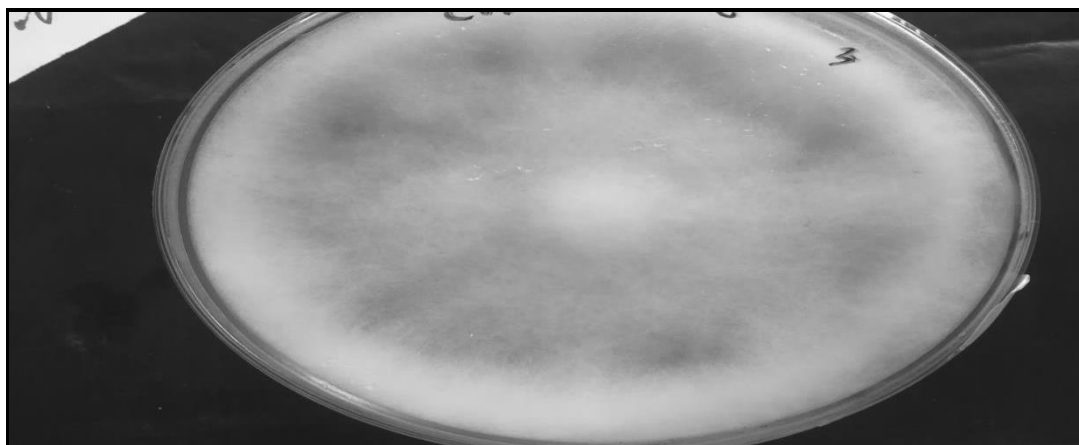


Figure 1 Colony morphology of *Fusarium oxysporum* f.sp. *lycopersici*

Characterization of Rhizobacteria: Through serial dilution method 15 rhizobacterial strains were isolated and characterized on the basis of morphological

characters like shape, size, elevation, edges, surface and colour

Table 1 Colony Morphology of Rhizobacteria

Strain No	Colony Morphology						Tentative identification
	Size	Shape	Elevation	Edges	Color	Surface	
RBS-1	Small	Irregular	Raised	Entire	Yellowish	Rough	Bacillus
RBS -2	Medium	Circular	Convex	Undulate	Creamy	Smooth	Bacillus
RBS -3	Small	Irregular	Convex	Entire	Creamy	Smooth	Pseudomonas
RBS -4	Small	Irregular	Flat	Lobate	Off-white	Smooth	Streptomyces
RBS -5	Medium	Irregular	Raised	Entire	Off-white	Smooth	Bacillus
RBS -6	Medium	Mucoid	Raised	Undulate	Light-Pink	Smooth	Pseudomonas
RBS -7	Small	Circular	Flat	Lobate	Creamy	Smooth	Pseudomonas
RBS -8	Small	Circular	Convex	Entire	Off-white	Rough	Bacillus
RBS -9	Medium	Mucoid	Raised	Entire	Yellowish	Smooth	Pseudomonas

RBS-10	Large	Irregular	Raised	Entire	Light-pink	Smooth	Streptomyces
RBS -11	Medium	Irregular	Raised	Lobate	Pink	Rough	Pseudomonas
RBS -12	Large	Circular	Flat	Entire	Off-white	Smooth	Bacillus
RBS -13	Small	Irregular	Convex	Undulate	Yellowish	Smooth	Bacillus
RBS -14	Large	Irregular	Raised	Entire	Green	Smooth	Pseudomonas
RBS -15	Medium	Circular	Convex	Entire	Off-white	Smooth	Bacillus

Morphological characterization of Rhizobacteria: It is one the phenotypic studies of cell to evaluate and identify the colony of various species of bacteria which is help full to identify the physical properties of cell colonies such as the size, the shape, elevation, edges and color. For the colony morphology, the size, the shape, elevation, edges, surface and color were considered for fixing the standard for this parameter. For size, strain RBS-10, RBS-12 and RBS-14 exhibited Large while strain RBS-1, RBS-3, RBS-4, RBS-7, RBS-8, and RBS-13 were observed to be small. Similarly, medium size was shown by the strain RBS-2, RBS-5, RBS-6, RBS-9, RBS-11 and RBS-15 respectively. For shape the strain RBS-2, RBS-7, RBS-8, RBS-12 and RBS-15 were identified as circular while strain RBS-1, RBS-3, RBS-4, RBS-5, RBS-RBS-10, RBS-11, RBS-13 and RBS-14 were recorded as irregular and strain RBS-6, and RBS-9

was Mucoid. As for elevation of strain is concern, RBS-1, RBS-5, RBS-6, RBS-9, RBS-10, RBS-11 and RBS-14 were observed to be Raised, strain RBS-2, RBS-3, RBS-8, RBS-13 and RBS-15 convex, and strain RBS-4, RBS-7 and RBS-12 observed to be Flat. Similarly, for edges of strain RBS-1, RBS-3, RBS-5, RBS-8, RBS-9, RBS-10 RBS-12, RBS-14 and RBS-15 were exhibited as entire while strain RBS-4, RBS-7 and RBS-11 Lobate, strain RBS-2, RBS-6 and RBS-13 Undulate respectively. For surface all the strains were smooth except RBS-1, RBS-8 and RBS-11. In this regard, the color of strain quite different from one another as: strain RBS-1, RBS-9 and RBS-13 were yellowish, RBS-4, RBS-5, RBS-8, RBS-12 and RBS-15 observed off white, RBS-6 and RBS-10 light pink, RBS-11 and RBS-14 green, RBS-2, RBS-3 and RBS-7 creamy.

Table 2 Cell Morphology of rhizobacteria

CELL MORPHOLOGY			
Strain N0	Shape	Motility Y/N	Gram +/-
RBS-1	Rod	Y	+
RBS -2	Rod	Y	+
RBS -3	Rod	Y	-
RBS -4	Filamentous	N	+
RBS -5	Rod	Y	+
RBS -6	Rod	Y	-
RBS -7	Rod	Y	-
RBS -8	Rod	Y	+
RBS -9	Rod	Y	-
RBS-10	Filamentous	N	-
RBS -11	Rod	Y	-
RBS -12	Rod	Y	+
RBS -13	Rod	Y	+
RBS -14	Rod	Y	-
RBS -15	Rod	Y	+

Cell Morphology: Cell morphology is one of the essential tests of identification of bacterial cell genotypic studies which comprise the cells shape, motility, gram positivity and negativity. For cell morphology, the PGPR isolates were scrutinized for shape, their motility and gram positivity and negativity. For shape parameter, all the isolate recorded as Rod shape, except Strain RBS-4 and RBS-10, which are filamentous. For motility all the strains were recorded as motile except RBS-4 and RBS-10. For gram test, the strain RBS-1, RBS-2, RBS-4, RBS-5, RBS-8, RBS-10, RBS-12, RBS-13

and RBS-15 were gram positive while strain NRBS-3, RBS-6, RBS-7, RBS-9, RBS-11 and RBS-14 were identified as gram negative

Biochemical characterization of rhizobacteria: The biochemical characterization of all the isolates was essentially done as per the procedure outlined in (Joseph, Patra, & Lawrence, 2007). The test conducted is detailed below.

Catalase test: Catalase test is the essential test to check the enzyme activity of the bacterial isolates. For catalase activity test all the selected rhizobacterial strain positive result

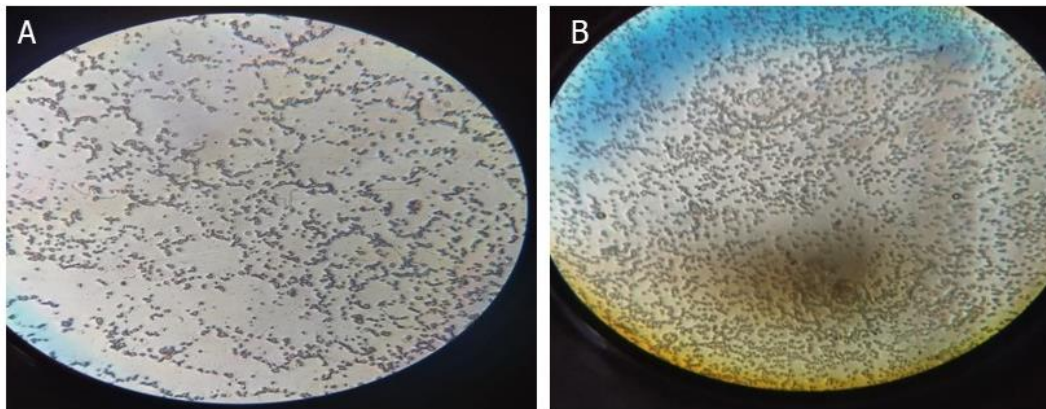


Figure 2 Microscopic picture of Rhizobacteria

Table 3 Biochemical characterization of rhizobacteria

Biochemical characterization of rhizobacteria				
Strain No	Catalase test +/-	Starch hydrolysis test +/-	KOH Test +/-	Ammonia production test +/-
RBS-1	+	+	+	+
RBS-2	+	+	+	+
RBS-3	+	+	-	+
RBS-4	+	+	-	-
RBS-5	+	+	+	+
RBS-6	+	+	-	+
RBS-7	+	+	-	+
RBS-8	+	+	+	+
RBS-9	+	+	-	+
RBS-10	+	-	-	+
RBS-11	+	-	-	+
RBS-12	+	-	+	+
RBS-13	+	-	+	+
RBS-14	+	-	-	+
RBS-15	+	+	+	+

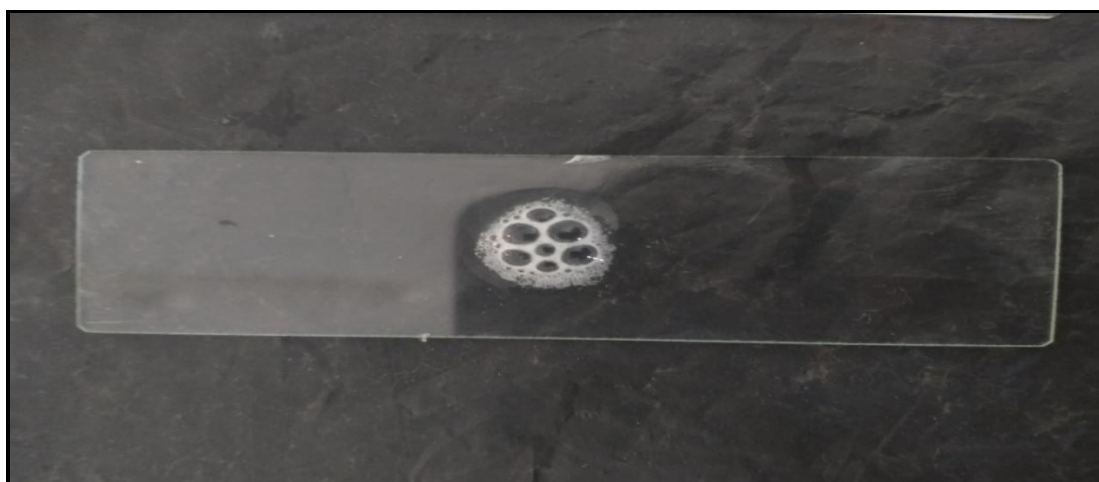


Figure 3 Catalase activity test

Starch Hydrolysis test: One important test to assess a bacteria's capacity to produce starch is the hydrolysis of starch test. For starch hydrolysis test the strain RBS-1, RBS-2, RBS-4, RBS-5, RBS-8 RBS-12, RBS-13 and RBS-15 show positive result while strain

RBS-3, RBS-6, RBS-7, RBS-9, RBS-11 and RBS-14 show negative result.

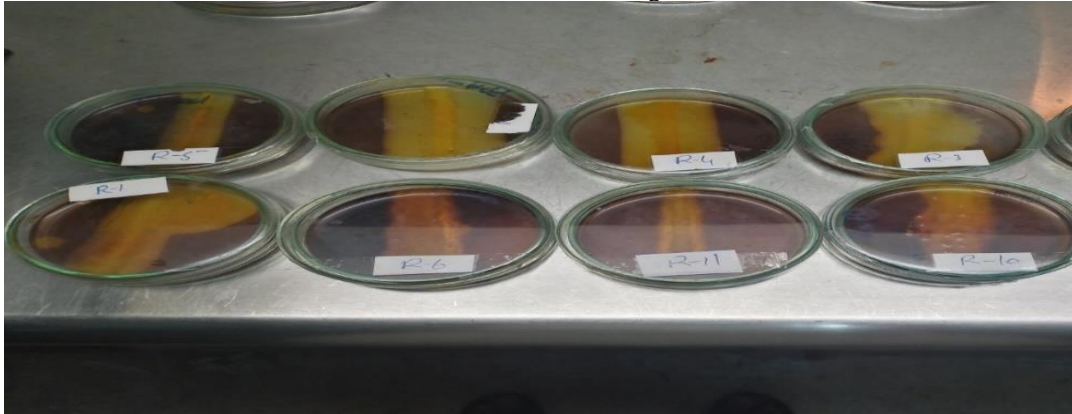


Figure 4 Starch Hydrolysis test

Potassium Hydroxide (KOH) Solubility Test: For potassium hydroxide solubility test the strain RBS-1, RBS-2, RBS-5, RBS-8 RBS-12, RBS-13 and RBS-15

were positive and strain RBS-3, RBS-4, RBS-6, RBS-7, RBS-9, RBS-10, RBS-11 and RBS-14 and 15 show negative result.

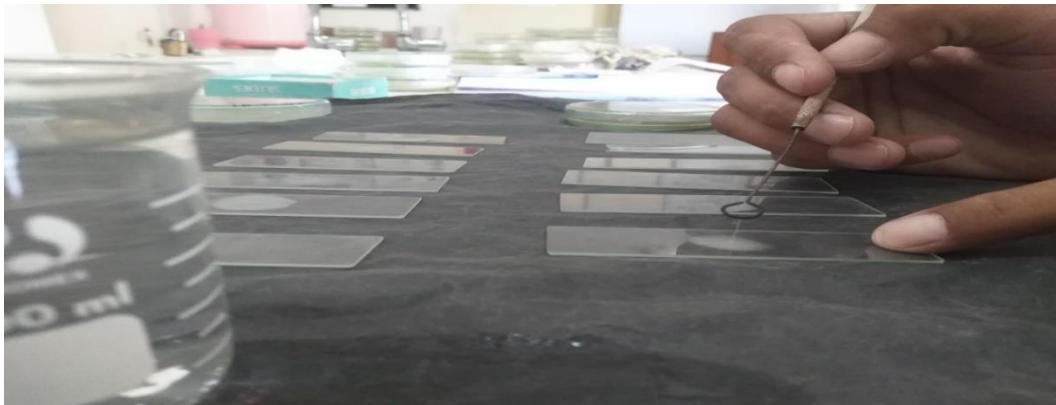


Figure 5 Potassium Hydroxide (KOH) Solubility Test

Ammonia production test: An essential test for determining a bacteria's capacity to produce ammonia is the ammonia production test. In this test strain RBS-1, RBS-2, RBS-3, RBS-5, RBS-6, RBS-7, RBS-

8, RBS-9, RBS-10, RBS-11, RBS-12, RBS-13 and RBS-15 indicated ammonia production and strain RBS-4 shown no ammonia production.



Figure 6 Ammonia production test

Table 4 Salt and pH Tolerance Test of Rhizobacterial Strains

Rhizobacteria Strains	Salt (NaCl) Tolerance Test			pH Tolerance Test		
	1%	3%	5%	6	7	8
RBS-1	+	+	+	+	+	+
RBS-2	+	+	+	+	+	+
RBS-3	+	+	+	+	+	-
RBS-4	+	+	-	+	+	-
RBS-5	+	+	+	+	+	+
RBS-6	+	+	+	+	+	+
RBS-7	+	+	+	+	+	-
RBS-8	+	+	+	+	+	+
RBS-9	+	+	+	+	+	+
RBS-10	+	+	-	+	+	-
RBS-11	+	+	+	+	+	-
RBS-12	+	+	+	+	+	+
RBS-13	+	+	+	+	+	+
RBS-14	+	+	+	+	+	-
RBS-15	+	+	+	+	+	+

Salt and pH Tolerance Test: The ability of the isolated rhizobacterial isolates to grow in different concentration of salt was tested by streaking them on NA medium containing 1%, 3% and 5% (wt/v) NaCl. All the rhizobacterial strains show positive result at 1%, 3% and 5% NaCl concentrations, except RBS-4 and RBS-10 at 5% NaCl concentration.

Differences in pH tolerance was tested by adjusting the pH to 6, 7, and 8. All the rhizobacterial isolate shows positive growth at pH 6, 7 and 8 except strain

RBS-4, RBS-7 and RBS-10 which shows negative growth at pH 8.

In Vitro antagonistic activity against FOL: Antagonistic activities of 15 isolates were tested against *Fusarium oxysporum* f.sp. *lycopersici* by dual culture in laboratory. All the isolates were found to inhibit the growth of *Fusarium oxysporum* f.sp. *lycopersici*. The percent inhibition was measured by the formula Percent inhibition (%) = (C-T)/CX100. T

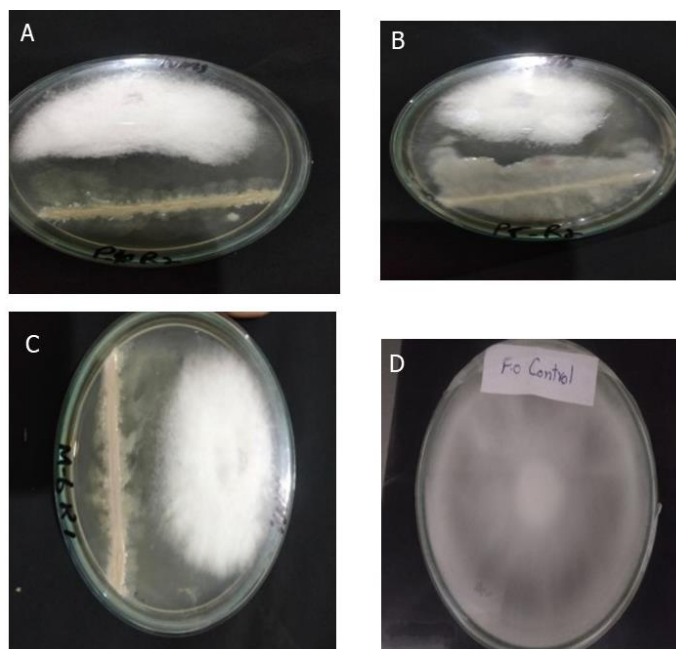


Figure 7 (A) Inhibition by Bascillus, (B) Inhibition by Pseudomonas (C) Inhibition by Streptomyces (D) Mycllium growth in control

Table 5 In vitro efficacy of rhizobacterial isolates against *Fusarium oxysporum* f.sp. *lycopersici* (Dual culture).

Strain No	<i>Fusarium oxysporum</i>	
	Mycelial growth (mm)	Percent inhibition over control(%)
RBS-1	51.48	42.80h
RBS -2	65.41	27.32l
RBS -3	68.38	24.02m
RBS -4	49.72	44.75g
RBS -5	38.44	57.28a
RBS -6	43.28	51.91c
RBS -7	53.54	40.51i
RBS -8	46.46	48.37e
RBS -9	45.35	49.61d
RBS-10	60.58	32.68k
RBS -11	47.19	47.56f
RBS -12	41.67	53.7b
RBS -13	71.25	20.83n
RBS -14	56.34	37.4j
RBS -15	43.44	51.73c
Control	89.09	0.01O

In vitro efficacy of rhizobacterial strains against *Fusarium oxysporum* f.sp. *lycopersici*: Antagonistic activities of 15 isolates were tested against *Fusarium oxysporum* f.sp. *lycopersici* by dual culture in laboratory. All the isolates inhibited the growth of *Fusarium oxysporum* f.sp. *lycopersici*. In 15 isolates of rhizobacteria the isolate RBS-5 show maximum growth inhibition and significantly inhibited the growth of *Fusarium* f.sp. *lycopersici*, which is 57.28 percent reduction on the growth of pathogen when compared to control. This is followed by isolate RBS-12, RBS-6, and RBS-15 in decreasing order of merit, which inhibited the growth of *Fusarium oxysporum* f.sp. *lycopersici* by 53.7, 51.91 and 51.73 percent over control. The least growth inhibition of pathogen was exhibited by isolate RBS-13 which is 20.83 percent.

DISCUSSION

The tomato, or *Solanum lycopersicum* L., is a member of the Solanaceae family and is grown worldwide because of its short growing season and excellent yield.

Carotenoids, antioxidants, vitamins C and E, and other nutrients found in tomatoes can assist improve the nutritional state of the general public, especially the under privileged. (Rai et al., 2012).

Healthy plants are essential for the welfare of both humans and animals. Pathogenic bacteria negatively impact the nutritional content, yield, and general well-being of plants (Fletcher et al., 2010). The infamous tomato pathogen *Fusarium oxysporum* f. sp. *Lycopersici* (FOL) is the cause of wilt disease and causes considerable crop losses. (Ahmed, 2011).

In this study, 15 Rhizobacterial isolates were isolated and tentatively identified. The isolates were identified as genus; *Bacillus*, *Pseudomonas* and *Streptomyces*. As an antagonist against *Fusarium oxysporum* f.sp. *lycopersici*, the isolates were used. In dual culture, every bacterial isolate showed reduction of *Fusarium oxysporum* mycelial growth. Numerous

investigators have also seen similar results with a range of fungi, such as *Fusarium* species. (Sivamani & Gnanamanickam, 1988; Khan & Zaidi, 2002).

Seven *Bacillus* isolates, six *Pseudomonas* isolates, and two *Streptomyces* isolates were found among the fifteen rhizobacterial isolates. Isolate RBS-13 was shown to be least efficient against *Fusarium oxysporum* mycelial growth, whereas isolate RBS-5 demonstrated the best growth inhibition at 57.28 percent. The biocontrol capability of seven *Bacillus* isolates obtained from the rhizosphere was assessed in vitro against FOL. The findings showed that every *Bacillus* isolate inhibited *F. oxysporum*'s mycelial growth to a different extent, with isolate RBS-2 suppressing it by 27.32% and isolate RBS-5 suppressing it by 57.28%. Several investigations have used *Bacillus* species as biological control agents and biofertilizers. (Cao et al., 2011; Li et al., 2012; Chen et al., 2013) It has been observed that *B. subtilis* produces a volatile material with antifungal qualities against soil-borne infections. (Fiddaman & Rossales, 1993).

Six isolates of *Pseudomonas* and two isolates of *Streptomyces* were evaluated for their biocontrol potential against FOL in vitro. The result revealed that *Pseudomonas* and *Streptomyces* isolates inhibit the mycelial growth of *Fusarium oxysporum* ranging from 20.83% to 51.91%. A review of earlier research has demonstrated *Pseudomonas species*' potential as biological control agents for harmful plant diseases. (Pastor et al., 2010; Khalimi & Surpata, 2011; Karimi et al., 2012; Saravanan et al., 2013).

The bioefficacy of thirty *P. fluorescens* isolates against *Fusarium* sp. was reported by Shahzaman et al. (2016). Pf3 was shown to be the most effective antagonist, showing an inhibiting percentage of 93.33 percent. Similar findings were reported by Vethavalli et al. (2012) The PV2 isolates exhibited the highest percentage of inhibition, with 77.22 percent suppressing *F. oxysporum* f.sp. *lycopersici* growth

over control. The efficiency of three isolates' in vitro antagonistic interactions was assessed in this work (PV2, BVE1, and SM1).

Pseudomonas fluorescens, *Putida*, *Chloropyrus*, *Bacillus subtilis*, *Streptomyces pulcher*, *S. corchorusii*, and *S. mutabilis* are bacterial bio control organisms that may be able to control *Fusarium oxysporum* f. sp. *lycopersici*, according to Monda (2002). Rhizobacteria can work as direct or indirect biological fertilisers and biostimulants by generating plant growth hormones that include indole acetic acid, gibberelin, cytokinin, ethylene, and dissolved minerals. Because they produce antibiotics and siderophore, they can additionally effectively impede the development of harmful microbes (Sarma et al., 2009).

CONCLUSION

It is concluded that the Rhizobacteria inhibit the mycelial growth of *Fusarium oxysporum* f.sp. *Lycopersici*. Among the Rhizobacteria, *Bacillus* show highest mycelial growth inhibition, followed by *Pseudomonas* and *Streptomyces*

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