





Exploring the potential of saline-tolerant *Streptomyces* for managing blast disease in rice (*Oryza sativa* L.)

V K Satya¹, S S J Roseleen¹, S Malathi², M Deivamani³, K Sethuraman⁴ & A Nithya Devi⁵

¹Department of Plant Protection, Horticultural College and Research Institute for Women, Tamil Nadu Agricultural University, Tiruchirapalli 620 027, India

²Information and Training Centre, Tamil Nadu Agricultural University, Chennai 600 032, India

³Indian Council of Agricultural Research - Krishi Vigyan Kendra, Tamil Nadu Agricultural University, Dharmapuri 636 809, India

⁴Department of Plant Pathology, Anbil Dharmalingam Agricultural College and Research Institute, Tamil Nadu Agricultural University, Tiruchirapalli 620 027, India

⁵Dr. MSS Agricultural College and Research Institute, Tamil Nadu Agricultural University, Thanjavur 614 902, India

*Correspondence email - sheeba@tnau.ac.in, malathi.s@tnau.ac.in

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Abstract

Rice (*Oryza sativa L.*) is recognized as one of the world's most important staple food crops. Yield loss due to diseases is one of the major constraints in rice cultivation. Among them, the blast disease caused by *Magnaporthe oryzae* ranks first in affecting the rice crop. Hence, the present study proposed to study the efficacy of antagonistic activities of *Streptomyces* spp. against blast disease in rice. Twenty isolates of the antagonistic *Streptomyces* sp. were isolated from rhizosphere soil. From the sequence analysis of 16s rRNA of isolate TRYS 10 was identified as *Streptomyces caelestis*. Antagonistic activity assay was conducted using a dual culture technique for blast pathogens. The isolates *S. caelestis* (TRYS 10) and TRYS 16 were effective against blast and recorded the inhibition zone of 19 and 15.2 mm, respectively followed by TRYS 5 (9.0 mm). An isolate *S. caelestis* (TRYS 10) induced the growth promotion of rice viz, root length (16.72 cm), shoot length (22.13 cm) and vigour index (3807). *S. caelestis* (TRYS 10) was tested for its efficacy against blasts under greenhouse conditions. Disease severity of blast in pathogen-inoculated control plants was 8.89 %, while in *Streptomyces* + pathogen-inoculated, it was 2.22 %. A foliar spray of *S. caelestis* (TRYS 10) at the time of symptom appearance and repeated 15 days later was found to be effective in reducing blast in rice up to 42.64 % under field conditions. Two sprays of *S. caelestis* at the time of initial appearance of blast disease and 15 days later reduced the disease incidence as well as improved the plant growth and yield in rice.

Keywords: blast; Magnaporthe; plant growth; pot culture; rice; Streptomyces; salinity

Introduction

Rice (Oryza sativa L.) is recognized as one of the most important staple food crops in the world and it provides the main source of energy for more than half of the world's population. It is the major food crop in India. Rice is life for most people living in Asia. Rice cultivation is affected by many factors, such as biotic and abiotic factors. Yield loss due to diseases is one of the major constraints in rice cultivation. The diseases infecting rice are blast (Magnapothe oryzae), sheath blight (Rhizoctonia solani), sheath rot (Sarocladium oryzae), false smut (Ustilaginoidea virens), bacterial leaf blight (Xanthomonas oryzae pv. oryzae) and rice tungro virus. Among them, the blast disease is caused by Magnaporthe oryzae, which poses a significant threat to food security, damaging as much as 30 % of the global rice harvest, with yield losses of 11.9 kg/ha in South and Southeast Asia. Yield loss caused by the blast disease ranged 10-30 % and if the disease outbreak, yield loss increased up to 50 % (1).

Rice blast disease produces various polycyclic symptoms, *viz.*, leaf blast, nodal blast, neck blast and grain discolouration. Symptoms produced are spindle-shaped spots with a grey centre and dark brown margin. During a severe case, it leads to drying of leaves, breaking of the nodal region and grain discolouration and chaffy grains. It is a polycyclic disease that spreads through pyriform conidia (2).

Current strategies used to control rice diseases include the use of disease-resistant varieties and synthetic chemical pesticides. So many fungicides used to manage the blast disease in rice are azoxystrobin, carbendazim, edifenphos, tricyclazole, pyroquilon, probenazole, iprobenfos, metominostrobin and propiconazole (3, 4). However, the use of chemical pesticides causes environmental hazards, the development of pesticide resistance, residual problems and a reduction in soil microflora and soil quality. In recent days, natural products which have low toxicity to living organisms and are safe for the environment have attracted more interest in the development of management strategies for plant

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diseases. Many biocontrol agents, viz., Pseudomonas, Bacillus, Enterobacter, Trichoderma, Gliocladium, Streptomyces, etc., are being used to control plant diseases in many agricultural and horticultural crops. Among these, Streptomyces spp. received more attention due to its excellent capacity to produce a variety of bioactive compounds such as antibacterial, antifungal, antiviral, anticancer and antioxidant properties (5). Streptomyces are gram-positive soil-dwelling bacteria belonging to the order Actinomycetales and the family Streptomycetaceae that have been largely used as biocontrol agents. It can survive under unfavourable environmental conditions due to its filamentous and sporulating nature and is able to compete more efficiently against many pathogens present in the soil (6).

Some studies reported the efficiency of Streptomyces spp. against blast and bacterial panicle blight in paddy. For example, Streptomyces UPMR54 reduced the rice blast disease by 67.9 % and promoted growth and yield (7). S. Vinaceusdrappus from the sediment of Loktak-Lake in India inhibited the growth of *Pyricularia oryzae* by 53.3 % (8). S. Philanthi RM-1-138 isolated from chilli pepper rhizosphere soil of southern Thailand inhibited the growth of P. oryzae PTRRC-18 in vitro (9). Similarly, foliar spray of S. Hygroscopicus OsiSh-2 culture filtrate recorded 23.5 % and 28.3 % disease reduction of blast in rice seedlings under greenhouse and field conditions, respectively (10). S. palmae PC12 reduced the mycelia growth of blast fungus by 87.3 % and also recorded maximum plant height and root length (11). Streptomyces strains A20 and 5.1 inhibited the growth of Burkholderia glumae, the incitant of bacterial panicle blight of rice. Hence, the present study proposed to study the efficacy of antagonistic activities of Streptomyces spp. against blast disease in rice and plant growth-promoting activity.

Materials and Methods

Isolation of blast pathogen Magnaporthe grisea

The blast pathogen was isolated from blast-infected rice leaves (variety TRY 3) using a potato dextrose agar (PDA) medium. The infected leaves were cut into small bits and sterilized with 0.1% mercuric chloride for 30 s and washed four times with sterile distilled water to remove excess mercuric chloride. Then, the sterilized leaf bits were placed in the PDA medium containing streptomycin. The plates were incubated at room temperature for 7 days. The mycelial growth and morphology and spore character were observed and recorded.

Isolation of actinobacteria *Streptomyces*

The isolation of actinomycetes was performed by the conventional serial dilution spread plate technique with rhizosphere soil of paddy. The suspension from an appropriate dilution was inoculated on *Actinomycetes* isolation Agar medium (Hi-Media Laboratories, Mumbai, Maharashtra, India) and incubated at 30 °C for 7 days. The colony character and morphology were observed and recorded.

Testing the efficacy of *Streptomyces* spp. against the blast pathogen

The antagonistic activity of *Streptomyces* sp. against *M. grisea* was tested by the dual culture technique. The mycelial disc of

9 mm from 7-day-old culture of *M. grisea* was placed at one edge of the Petri plate containing PDA media. After 3 days, the *Streptomyces* sp. isolates were streaked on the opposite edge of the Petri dish, 1 cm away from the edge. The plates were incubated at room temperature for 5 to 7 days. The control plate is also maintained by inoculating the fungal discs alone. The width of the inhibition zone was measured and the inhibition zone was found by measuring the distance between the antagonist actinobacteria and the mycelial growth present in the plates. The radial growth of the mycelium of *M. grisea* was also recorded to calculate the per cent inhibition of mycelial growth. Three replications were maintained for each isolate. The per cent inhibition of mycelia growth of pathogen *M. grisea* is calculated using the formula given (12).

Per cent inhibition (I) = $[(C - T)/C] \times 100$

Where,

C = mycelial growth of pathogen M. grisea in control

T = mycelial growth of pathogen *M. grisea* in dual culture plate

Molecular characterization of Streptomyces isolates

The total genomic DNA of the superior Streptomyces isolate, TRYS 10 was extracted using the Cetyl Trimethyl Ammonium Bromide (CTAB) method (13). The 16S rRNA gene of Streptomyces isolate, TRYS 10 was amplified with a primer pair of 27F (5'- AGAGTTTGATCCTGGCTCAG-3') and 1492R (5'-GGTTACCTTGTTACGACTT-3') through Polymerase Chain reaction (PCR). The PCR was performed with a 20 µL reaction mixture containing 50 ng of genomic DNA (1 μL), 10 μL of 2 X genei master mix, (readymade mix of Taq polymerase, dNTPs and PCR Buffer, Genei Laboratories Pvt. Ltd., cat no # 0667700041730), 20 pmol of each primer and volume was made to 20 µL by adding sterile distilled water. The PCR reaction was programmed with an initial denaturation at 94° C for 5 min, followed by 35 cycles of 94 °C for 1 min, 55 °C for 1 min, 72 °C for 40 s and final extension at 72 °C for 10 min. PCR amplicons were visualized by electrophoresis in 1 % agarose gel in TAE buffer and documented with an Alpha Imager 2000 (Alpha Innotech, San Leandro, CA). The sizes of the PCR amplicons were analyzed by comparison with a standard 1 kb molecular marker (Bangalore Genei Pvt. Ltd., Bangalore, India). PCR amplicons were sequenced by utilizing the service of PAR Life Science, Tiruchirapalli. The sequence was analyzed by BLAST analysis available at NCBI database (https://www.ncbi.nlm.nih.gov).

Assessment of plant growth promotion

The seedling vigour index was used to assess the plant growth-promoting activity of actinobacteria. The germination paper was soaked in water for nearly 2 hr to remove toxic substances present in it. Paddy seeds were soaked with talc formulation of superior isolate *S. caelestis* (TYRS 10) at 10 g/kg of seeds. The seeds were soaked in water alone and served as a control. The paddy seeds (25 nos.) were placed in each germination paper at an equal distance and it was rolled carefully. The prepared roll of towel was allowed to germinate for 15 days. After 15 days, the shoot length, root length and germination (%) were measured and calculated

seedling vigour index by following the formula. Six replications were maintained for each treatment and 25 seeds for each replication.

Seedling vigour index (VI) =

(Mean root length (cm) + Mean shoot length (cm))

× Germination (%)

Efficacy of *S. caelestis* (TYRS 10) against blast disease under pot culture

The experiment was carried out in a glasshouse at the Department of Plant Protection, Anbil Dharmalingam Agricultural College and Research Institute, Tiruchirapalli. Surface sterilized rice seeds were sown in pots. The spore suspension of the antagonistic *S. caelestis* (TYRS 10) containing 0.2 % (v/v) Tween 20 was sprayed on 10-day-old rice seedlings using a spray bottle. The conidial suspension of the pathogen was sprayed 7 days later in the same way and *vice versa*. The experiment was carried out in pot culture with ten treatments in three replications following a completely randomized design (CRD). The treatment details are mentioned below.

- T_1 -Rice seedlings inoculated with antagonistic *S. caelestis* (TRYS 10) at $10^6\,\text{CFU/mL}$ alone
- T_2 -Rice seedlings inoculate with Magnaporthe oryzae at 10^5 spores/mL alone
- T_3 -Rice seedlings inoculate with antagonistic S. caelestis (TRYS 10) at 10^6CFU/mL first + then Magnaporthe oryzae at 10^5spores/mL
- T_4 -Rice seedlings inoculate with Magnaporthe oryzaeat 10^5 spores/mL first + then antagonistic S. caelestis (TRYS 10) at 10^6 CFU/mL
- T_5 -Rice seedlings inoculate with antagonistic *Bacillus subtilis* Bbv57 at $10^6\,\text{CFU/mL}$ first + then *Magnaporthe oryzae* at $10^5\,\text{spores/mL}$
- $\rm T_6$ -Rice seedlings inoculate with Magnaporthe oryzae at $\rm 10^5$ spores/mL first + then antagonistic B. subtilis Bbv57 at $\rm 10^6$ CFU/mL
- $\rm T_7$ -Rice seedlings inoculate with antagonistic $\it B. \, subtilis \, Bbv57$ alone at $\rm 10^6 \, CFU/mL$
- T₈ -Rice seedlings inoculate with fungicide Tricyclozole 75 WP at500g/ha first + then *Magnaporthe oryzae* at 10⁵ spores/mL
- T_9 -Rice seedlings inoculate with Magnaporthe oryzae at 10^5 spores/mL first + then Tricyclozole 75 WP at 500 g/ha

T₁₀ -Healthy control

The disease incidence level was recorded 45 days after inoculation, following a standard evaluation system for rice given by the International Rice Research Institute (IRRI), Philippines (14). The disease score ranged from 0 to 9 as follows

- 0 no lesions observed
- 1 small brown specks of pinpoint size without a sporulating centre
- 2 larger brown specks but less than 1 mm in diameter
- 3 small roundish to slightly elongated, necrotic grey spots 1-2 mm
- 4 typical susceptible blast lesions (spindle-shaped) 3 mm or

longer, infecting less than 4.0 % of the leaf area

- 5 typical blast lesions infecting 4.1-10.0 % of the leaf area
- 6 typical blast lesions infecting 10.1-25.0 % of the leaf area
- 7 typical blast lesions infecting 25.1-50.0 % of the leaf area
- 8 typical blast lesions infecting 50.1-75.0 % of the leaf area
- 9- typical blast lesions infecting more than 75.1 % of the leaf area

The per cent disease index (PDI) was calculated using the following formula

Per cent disease index (PDI) =

The sum of individual ratings × 100

Total no. of leaves observed × Maximum grade

Efficacy of *S. caelestis* (TYRS 10) against blast under saline soil conditions

A field trial was conducted at Anbil Dharmalingam Agricultural College and Research Institute, Tiruchirapalli. The experiment was conducted with six treatments as detailed below and four replications under Randomized block design (RBD) using rice variety BPT 5204. The spraying was done at the time of the first appearance of disease symptoms and 15 days later.

- T_1 Foliar spray of antagonistic *S. caelestis* (TRYS 10) at 2.5 kg/ha
- T_2 Foliar spray of antagonistic *Streptomyces* sp. (TRYS 16) at 2.5 kg/ha
- T₃- Foliar spray of antagonistic B. subtilis Bbv57 at 2.5 kg/ha
- T₄- Foliar spray of Tricyclazole 75 WP at 500 g/ha
- T₅- Untreated control (No spray)

The disease severity was calculated at 65 DAP. The yield and benefit-cost ratio were estimated to find out the performance of actinobacteria.

Results and Discussion

Isolation of blast pathogen and pathogenicity

Magnaporthe grisea was isolated from the diseased leaves of rice variety TRY 3. Morphological characterization of the PDA plate showed that isolated fungi developed a light grey to dark-grey mycelium that formed concentric rings on the growth medium. Microscopic observation showed that pyriform conidia consisted of 2-3 septations. A pure culture of isolated Magnaporthe grisea was inoculated onto rice cultivar BPT 5204 to prove its pathogenic nature. The spindle-shaped spots were expressed 14 days after inoculation (DAI) in inoculated plants, whereas disease was not developed in uninoculated plants. The pure culture was subcultured and maintained at 4 °C for further study.

Isolation of Streptomyces

A total of twenty isolates of *Streptomyces* sp. were isolated from rhizosphere soil in rice with a pH of 9.5 and EC of 2.56 ds m⁻¹ at Anbil Dharmalingam Agricultural College and Research Institute, Tiruchirapalli (10.7557 °N, 78.6010 °E). Morphological

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characters were observed for all the isolates. Powdery colonies with filamentous, branched mycelium were observed and colonies appeared as various colours *viz.*, white, green, brown and cream (Table 1). The isolates of *Streptomyces* sp. were named TRYS 1 to TRYS 20 for easy communication.

Efficacy of Streptomyces sp. against blast

Antagonistic activity assay was conducted using a dual culture technique for blast pathogens. The diameter of the inhibition zone was measured after the control plate achieved 100 % mycelial growth and mycelial growth reduction was also calculated using the formula. The isolates TRYS 10 and TRYS 16 were effective against blast and recorded the inhibition zone of 19 and 15.2 mm respectively, followed by TRYS 5 (9.0 mm) (Fig. 1 & Table 2). Isolate TRYS 10 recorded a maximum reduction in mycelial growth of 81.1 %, followed by TRYS 16 (68.89 %).

Molecular characterization of superior isolate TRYS 10

PCR amplicons of the 16s rRNA gene of isolate TRYS 10 were sequenced and sequence analysis was done with the BLAST search program and it showed a maximum 90 % identity with *S. caelestis* (MT760574). Hence, the isolate was identified as *S. caelestis*.

Table 1. Morphological characterization of isolates of Streptomyces

S.No.	Native isolates	Colony appearance
1	TRYS 1	Dark brown
2	TRYS 2	Cream colour
3	TRYS 3	Pure white
4	TRYS 4	Dull white
5	TRYS 5	Grey and creamy
6	TRYS 6	White cottony
7	TRYS 7	Golden yellow
8	TRYS 8	Cream colour
9	TRYS 9	Cream colour
10	TRYS 10	Light green
11	TRYS 11	Cream colour
12	TRYS 12	Cream colour
13	TRYS 13	Cream colour
14	TRYS 14	Golden yellow
15	TRYS 15	Circular grey
16	TRYS 16	White cottony
17	TRYS 17	Dark brown
18	TRYS 18	Circular white
19	TRYS 19	Cream colour
20	TRYS 20	Light green

An isolate of *S. caelestis* (TRYS 10) was tested for its plant growth promotion activity using the roll towel method with the variety TRY 3. The plant growth parameters, root length, shoot length and vigour index in *S. caelestis* (TRYS 10) sprayed plants were 16.72 cm, 22.13 cm and 3807, respectively, which was higher than untreated plants (Table 3). Whereas, in the case of control plants, root length, shoot length and vigour index were 12.56 cm, 18.26 cm and 2897, respectively. The results revealed that it induced the growth promotion of rice.

Efficacy of *Streptomyces* sp against blast pathogen under pot culture

S. caelestis (TRYS 10) was selected based on their performance in *in vitro* studies and tested their efficacy against blast pathogens under greenhouse conditions with different combinations of treatment. The inoculated plants were maintained in a greenhouse and observed for their symptom expression. The initial disease symptoms appeared six days post inoculation (dpi) as brown spots. Then, the lesion size increased and became spindle-shaped spots with a grey centre and dark brown margin. Percent disease index was calculated at 45 dpi. Disease severity in pathogen-inoculated control plants was 8.89 %, while in pre-inoculation of *S. caelestis* (TRYS 10) + pathogen inoculated, it was 2.22 %.

Table 2. Efficacy of Streptomyces sp. against blast pathogen

S.No.	Name of the isolates	Inhibition zone (mm)*	Reduction in mycelial growth (%)*
1	TRYS 1	0.1	21.11
2	TRYS 2	0.5	46.67
3	TRYS 3	0.5	48.89
4	TRYS 4	0.2	17.78
5	TRYS 5	9.0	62.22
6	TRYS 6	0.1	15.56
7	TRYS 7	0.1	18.89
8	TRYS 8	0.2	16.67
9	TRYS 9	0.1	22.22
10	TRYS 10	19.0	81.11
11	TRYS 11	0.2	23.33
12	TRYS 12	1.0	8.89
13	TRYS 13	2.0	6.67
14	TRYS 14	1.0	12.22
15	TRYS 15	0.0	0.0
16	TRYS 16	15.2	68.89
17	TRYS 17	1.1	13.33
18	TRYS 18	0.5	15.56
19	TRYS 19	0.4	10.00
20	TRYS 20	2.0	20.00

^{*} Mean of three replications



Control



Streptomyces caelestis (TRYS10)



TRYS 16



TRYS 5

Fig. 1. Efficacy of Streptomyces sp. against blast pathogen.

Plant growth promotion

Table 3. Effect of *Streptomyces caelestis* (TRYS 10) on plant growth promotion

Particulars	Germination (%)	Shoot length (cm)	Root length (cm)	Vigour index
Treated	98	22.13	16.72	3807.30
Control	94	18.26	12.56	2897.08

The disease severity in pathogen inoculated + post-treatment of *S. caelestis* (TRYS 10) plants was 3.7 %. No disease development was observed in the healthy control and *Streptomyces* alone sprayed plants (Fig. 2 & Table 4). The results indicated that the prophylactic spray of *S. caelestis* (TRYS 10) in rice plants was most effective as compared to post-treatment.

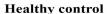
Efficacy of *S. caelestis* (TRYS 10) against blast under saline soil conditions

A field trial was conducted at ADAC & RI, Trichy, under salinity soil conditions with a pH of 9.8. The experiment was conducted with five treatments and five replications under Randomized block design (RBD) using rice variety BPT 5204. The first spraying was done at the time of the first appearance of disease symptoms and 15 days later.

From the results, it is inferred that foliar spray of *S. caelestis* (TRYS 10) at the time of symptom appearance and repeated 15 days later, recorded the disease incidence of 19.44 % blast, which was found to be low when compared to the untreated control (33.89 %) (Fig. 3 & Table 5). It was on par with the treatment, foliar spray of antagonistic *B. subtilis* Bbv57 at 2.5 kg/ha (T_3). A foliar spray of *S. caelestis* (TRYS 10) recorded a high yield of 5301 kg/ha with a BCR of 2.47. While the control plot recorded a yield of 3977 kg/ha with a 1.96 BCR. Yield increase and disease reduction by *S. caelestis* (TRYS 10) was almost on par with the chemical check tricyclazole treated plot (67 % reduction over control).

The common practice followed by the farmers for the management of blast disease in rice is the use of various fungicides. It causes environmental hazards, as well as residue in rice grains leads to health problems for humans. In addition to that, it increased the cost of cultivation. Repeated applications of chemical fungicides developed resistance and it paved the way for the development of new strains/-races of the blast pathogen in rice. Hence, in this study, 20 isolates of *Streptomyces* were isolated from the rhizosphere soil of rice







Streptomyces caelestis (TRYS 10) treated + M. grisea inoculated



M. grisea alone inoculated

Fig. 2. Testing the efficacy of Streptomyces caelestis (TRYS 10) against blast disease under pot culture.

Table 4. Testing the efficacy of Streptomyces caelestis (TRYS 10) against blast under pot culture

	Treatment details	Blast PDI
T ₁	Rice seedlings inoculated with antagonistic <i>Streptomyces caelestis</i> (TRYS 10) at 10 ⁶ CFU/mL alone Rice seedlings inoculated with <i>Magnaporthe oryzae</i> at 10 ⁵ spores/mL alone Rice seedlings inoculated with antagonistic <i>Streptomyces caelestis</i> (TRYS 10) at 10 ⁶ CFU/mL first + then <i>Magnaporthe oryzae</i> at 10 ⁵ spores/mL	
T ₂		
T ₃		
Γ ₄	Rice seedlings inoculated with <i>Magnaporthe oryza</i> eat 10 ⁵ spores/mL first + then antagonistic <i>Streptomyces caelestis</i> (TRYS 10) at 10 ⁶ CFU/mL	(7.21) 3.7 (10.96)
Γ ₅	Rice seedlings inoculated with antagonistic <i>Bacillus subtilis</i> Bbv57 at 10 ⁶ CFU/mL first + then <i>Magnaporthe oryzae</i> at 10 ⁵ spores/mL	2.22 (7.21)
Γ ₆	Rice seedlings inoculated with <i>Magnaporthe oryzae</i> at 10 ⁵ spores/mL first + then antagonistic <i>Bacillus subtilis</i> Bbv57 at 10 ⁶ CFU/mL	2.96 (9.76)
7	Rice seedlings inoculated with antagonistic <i>Bacillus subtilis</i> Bbv57 aloneat 10 ⁶ CFU/mL	
8	Rice seedlings inoculated with fungicide Tricyclozole 75 WP at 500 g/ha first + then $\it Magnaporthe\ oryzae$ at 10 $^{\rm 5}$ spores/mL	1.48 (4.66)
9	Rice seedlings inoculated with $\it Magnapor the\ or yzae$ at 10^5 spores/mL first + then Tricyclozole 75 WP at 500 g/ha	1.48 (4.66)
T ₁₀	Healthy control	0 (0.0)
	CD (0.05) SEd	7.68 3.68

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Field overview Streptomyces caelestis (TRYS 10) treated

Untreated control

Fig. 3. Testing the efficacy of Streptomyces caelestis (TRYS 10) against blast disease under field conditions.

Table 5. Efficacy of Streptomyces spp. against blast disease in rice

	Blast				
	Treatment Details	Disease severity (%)	Percent reduction over control	Yield (kg/ha)	BCR
_	Foliar spray of antagonistic Streptomyces caelestis (TRYS 10) at 2.5 kg/ha	19.44	42.64	5301.25	2.47
T₁ Folia		(26.08)			
T ₂ Foliar spray	Foliar spray of antagonistic Streptomyces spp. (TRYS 16) at 2.5 kg/ha	21.39	36.88	5270.75	2.45
	Foliar Spray of antagonistic <i>Streptomyces</i> Spp. (1 KYS 16) at 2.5 kg/fla	(27.47)			
_	Foliar spray of antagonistic TNAU Bacillus subtilis Bbv57 at 2.5 kg/ha	20.56	39.33	5277.25	2.46
T ₃		(26.93)			
T ₄	Folian aggress of Triangle-ole 75 WD at 500 m/ha	11.11	67.22	5926.25	2.78
14	Foliar spray of Tricyclazole 75 WP at 500 g/ha	(19.41)	67.22		
_	Untreated control (No spray)	33.89		3977.50	1.96
T ₅		(35.59)	-		
	CD (0.05)	5.40		184.11	
	SEd	2.53		86.37	

and tested for their efficacy against blast pathogens. The isolate *S. caelestis* (TRYS 10) recorded the highest inhibition zone (19 mm) against blast and arrested maximum mycelial growth (81.1 %). *Streptomyces* have the ability of antifungal, antibacterial, antiviral, anticancer, antioxidant and immunosuppressive due to the production of various bioactive compounds such as volatiles and secondary metabolites, etc. (6).

The present study was supported by many researchers. Mycelial growth of the blast pathogen was effectively arrested by *S. globisporus* JK-1 (15). *Streptomyces* isolates isolated from Iran, *viz.*, *Streptomyces* isolate 339 and *S. sindeneusis* isolate 263 was found effective against the blast pathogen in rice (16). *S. flavotricini* isolated from rice fields in Egypt showed effectiveness against blast disease (17). *S. palmae* PC 12 showed inhibition against *M. grisea* as well as promoted the plant growth activity (18).

Under glasshouse conditions, *S. caelestis* (TRYS 10) as a preventive spray arrested the lesion development (2.22 PDI) when compared to the untreated control (8.89 PDI). Rice plants treated with *S. caelestis* (TRYS 10) after infection of blast pathogen also showed effectiveness in arresting the symptom development. This work was supported by some research (19). When the rice plants were treated with *S. sindeneusis* isolate 263 + blast pathogen, lesion development in rice leaf by blast pathogen was 0.5 % only. Whereas 8 % of lesion development in rice leaf was observed in the case of rice plants only with *M. oryzae*. It confirmed the antagonistic activity of *Streptomyces*. *S. koyanogensis* (Strain TA-47) was found to reduce the blast

disease incidence to 19.66 % over the control plot (44.66 %) as well as improve the root and shoot growth by 16.54 cm and 72.22 cm, respectively (20). Streptomyces isolates U, G and No. 5 were reported to be effective in the reduction of blast fungus growth in laboratory and glasshouse conditions out of thirty isolates in Northern Iran (21).

Conclusion

This study effectively isolated the antagonistic *Streptomyces* from the rhizosphere soil of rice. Antifungal activity of *Streptomyces* isolates was assessed and a significant reduction in mycelial growth was observed. Effective superior isolate, *S. caelestis* (TRYS 10), showed plant growth promotion significantly and reduced the blast disease severity under pot culture as well as in field conditions under saline soil. It can be salinity tolerant as it is isolated from rice in saline soil. This study suggests that the application of *S. caelestis* (TRYS 10) is found effective against rice disease. Future research could concentrate on metabolite profiling of *S. caelestis* (TRYS 10) and identify the compounds responsible for disease resistance and it can be used as a biomarker in resistance breeding.

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Authors' contributions

VKS executed the conceptualization, methodology and research work. VKS, KS, SSJ and SM were involved in writing, reviewing and editing the manuscript. MD did the statistical analysis. AND, VKS and SSJ wrote the draft proof and edited the manuscript. All authors read and agreed to the final version of the manuscript.

Compliance with ethical standards

Conflict of interest: There is no conflict of interest among the authors

Ethical issues: None

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