

Biocontrol Potential of Actinobacterial Strains Against *Rhizoctonia solani* in Cowpea

Nelamangala Lalithesh Niveditha*¹, Devaki Girija¹, Kulkarni Surendra Gopal¹, Bobby Vattekkattu Unnikrishnan¹ and Reshmy Vijayaraghavan²

¹ Department of Agricultural Microbiology, College of Agriculture, Kerala Agricultural University, Vellanikkara, Thrissur - 680 656, Kerala, India

² Department of Plant Pathology, Kerala Agricultural University, Vellanikkara, Thrissur - 680 656, Kerala, India

Received: 28 Nov 2024; Revised accepted: 04 Jan 2025

Abstract

The management of soil-borne pathogens like *Rhizoctonia solani* is challenging due to their complex biology and limited availability of effective active ingredients. Eco-friendly methods, such as biological control, are essential for disease prevention and maintaining soil health. This study evaluated the antagonistic activity of actinobacteria for the management of collar rot and web blight in cowpea. Among 50 actinobacterial isolates, 12 produced ammonia, nine produced siderophores, and none produced HCN *in vitro*. Ten isolates exhibited antagonistic activity against *Rhizoctonia solani* under dual culture method. Five actinobacterial consortia were tested *in planta* against *Rhizoctonia solani* in cowpea, with comparisons to KAU PGPR Mix-2 and Carbendazim (0.1%). Treatment with consortium 3 (CR-3 and S2-2) recorded significantly higher yield and lower incidence of collar rot, which also exhibited higher per cent inhibition of *R. solani* (63.7% and 70.3%) *in vitro*. Per cent Disease Index (PDI) of web blight was significantly lower (19.4%) in the same treatment and this was comparable to chemical control (Carbendazim). Identification by 16S rRNA gene sequencing revealed that the isolates CR-3 and S2-2 were closely related to *Streptomyces pratensis* and *Streptomyces cinereus*, respectively. These results suggest that actinobacteria could be exploited as a potential alternative to chemical pesticides.

Key words: Actinobacteria, Consortia, Collar rot, Web blight, *Rhizoctonia solani*

Actinobacteria are a group of Gram-positive bacteria, which can be terrestrial or aquatic, having high guanine and cytosine content in their DNA. Most of the actinobacteria are saprophytes, ubiquitous and are one of the most diverse groups of bacteria in nature. Their nature varies from anaerobic unicellular organisms to aerobic, filamentous, and spore-forming lineages [1]. In addition to the plant growth-promoting abilities of actinobacteria, they possess the ability to combat pathogens and effectively manage various plant diseases. Actinobacteria are well-recognized for their ability to synthesize secondary metabolites, which play a crucial role in controlling diverse pathogens, particularly those affecting plants. Actinobacteria are regarded as potential natural biocontrol agents in the soil [2]. Among actinobacteria, the *Streptomyces* genus is a major source of bioactive compounds [3].

Cowpea (*Vigna unguiculata* subsp. *unguiculata* (L.) Verdcourt), an important legume crop in Kerala, is cultivated in various regions across the state. However, its cultivation faces significant production constraints due to soil-borne diseases. It is an excellent source of plant-based protein, vital for improving the diet of many people. However, collar rot and web blight disease, caused by *Rhizoctonia solani*, significantly affect cowpea productivity, resulting in substantial yield losses. These

diseases severely impact plant health and productivity, leading to significant yield losses. Effective management strategies are crucial to mitigate the effects of these soil-borne diseases and ensure sustainable cowpea production. The increasing concern over chemical fertilizer and pesticide application has driven the need for environmentally sustainable alternatives in agriculture. The growing concern over the adverse effects of chemical fertilizers and pesticides has highlighted the urgent need for environmentally sustainable alternatives in agriculture. Sustainable practices, such as the use of biofertilizers, biopesticides, and integrated disease management strategies, are gaining prominence as they help maintain soil health, reduce environmental impact, and ensure the long-term viability of crop production systems. Adopting such approaches is particularly important for crops like cowpea, which are vulnerable to soil-borne diseases.

There are many reports on the use of microorganisms as biocontrol agents as an alternative to agricultural chemical fungicides [4]. *Streptomyces* spp. have been shown to trigger Induced Systemic Resistance (ISR) *in planta* and inhibit pathogen growth *via* induction of plant defense mechanisms [5]. *Streptomyces* spp. produces a novel compound, antifungalmycin N₂, which was found to be effective against *Rhizoctonia solani* [6]. The application of a consortium

*Correspondence to: Nelamangala Lalithesh Niveditha, E-mail: nivedithanln@gmail.com; Tel: +91 6361844299

Citation: Niveditha NL, Girija D, Gopal KS, Unnikrishnan BV, Vijayaraghavan R. 2025. Biocontrol potential of actinobacterial strains against *Rhizoctonia solani* in cowpea. *Res. Jr. Agril. Sci.* 16(1): 16-22.

consisting of *Streptomyces* spp. caused the highest disease control (73%) of *Rhizoctonia solani* in rice, followed by actinobacteria AUdT502 (70%) and validamycin (purified from *Streptomyces hygroscopicus*) (65%) [7]. *Streptomyces griseoviridis* and *Streptomyces lydicus* have shown effectiveness against soil-borne pathogens, including species of *Rhizoctonia*, *Phytophthora*, *Fusarium*, and *Pythium*, in legumes and other crops [8]. In this regard, the present study was an attempt to evaluate the antagonistic ability of actinobacteria against *Rhizoctonia solani*, the fungal pathogen inciting collar rot and web blight in cowpea.

MATERIALS AND METHODS

The experiment was conducted at the Department of Agricultural Microbiology, College of Agriculture, Vellanikkara, Kerala from October 2023 to September 2024. PGPR Mix 2 (KAU biofertilizer) is a consortium of *Bacillus subtilis* and *Pseudomonas fluorescence* having a broad spectrum of inhibitory properties with different mechanisms. *Rhizoctonia solani*, a fungal pathogen of cowpea was obtained from the Department of Plant Pathology, Kerala Agricultural University (KAU), College of Agriculture, Vellanikkara. The culture was sub-cultured and maintained on Potato Dextrose Agar (PDA) for further studies.

Isolation of actinobacteria from soil and compost samples

Samples were collected from seven different sources including cowpea rhizosphere soil (CR) from Thrissur district, ginger rhizosphere soils (S) from Wayanad district, mangrove forest soil (MS) from Kadappuram (Chettuva) of Thrissur district, uncultivated soil (US) from Kerala Agricultural University (KAU) campus, Vellanikkara, Thrissur, compost (C), coir pith compost (Cc) and vermicompost (VC) from Thrissur district. Actinobacteria were isolated and enumerated from various samples using serial dilution and plating methods [9] on starch casein agar (HiMedia Laboratories, Mumbai, India). The Petri plates were incubated at 28 ± 2 °C for a period of five to 12 days. Pure cultures of a total of 50 morphotypes were maintained, including isolates from the repository at the Department of Agricultural Microbiology, KAU, Vellanikkara which were originally isolated from the rhizosphere of rice and black pepper for further studies.

In vitro screening of actinobacteria for indirect plant growth promoting (PGP) activities

All the 50 isolates were subjected to indirect PGP activities such as production of Hydrogen cyanide (HCN), ammonia, and siderophores under *in vitro* conditions.

Ammonia production

Freshly grown actinobacterial isolates were inoculated to sterile 4% peptone water. They were incubated at 28 ± 2 °C for three to four days. After incubation, 0.5 ml of Nessler's reagent was added to each tube. The development of orange to brown colour indicated ammonia production [10].

Siderophore production

All the actinobacterial isolates were assessed for siderophore production on Chrome Azurol Sulfonate (CAS) agar [11]. The isolates were spot-inoculated onto the media and incubated for seven days at 30 ± 2 °C. The yellow to orange halo surrounding the colony indicated the presence of siderophore production by actinobacteria.

HCN production

All the actinobacterial isolates were assessed for hydrogen cyanide production [12]. Luria-Bertani (LB) agar was supplemented with glycine at a concentration of 4.4 glycine/L, cooled, and poured into Petri plates to solidify. Whatman filter paper No. 1 dipped in 2% sodium carbonate in 0.5% picric acid for a minute was placed in the Petri plate lids. The plates were sealed with parafilm and incubated at 28 ± 2 °C for seven to 12 days. After incubation, the discoloration of the filter paper to an orange-brown color indicated cyanide production by the actinobacteria.

Secondary screening of actinobacterial isolates for antagonistic activity against *Rhizoctonia solani*

Isolates exhibiting indirect mechanisms of plant growth promotion were further assessed for their antagonistic potential against *Rhizoctonia solani*, the fungal pathogen inciting collar rot and web blight in cowpea, by dual culture method [13]. The antagonistic effect was evaluated by measuring the inhibition zone formed on the plates after incubation at 25 °C for 14 days. The inhibition percentage of *R. solani* was calculated using a formula:

$$I = \frac{C - T}{C} \times 100$$

Where;

I = Percent inhibition

C = Radial growth of the pathogen in control

T = Radial growth of the pathogen in treatment

Compatibility of potential actinobacteria with antagonistic activity

The compatibility of the selected promising actinobacterial isolates was assessed using the *in vitro* cross-streak method. A merger of the actinobacterial growth at the junction of two isolates indicated compatibility [14]. Compatible isolates were used for the preparation of consortia.

In planta evaluation of the efficacy of actinobacterial consortia for management of collar rot and web blight in cowpea

The Five most promising and efficient consortia were evaluated under sterile conditions for the management of collar rot and web blight in cowpea. The experiment was conducted from June to September 2024 using a completely randomized design (CRD) in the net house of the Department of Agricultural Microbiology, Vellanikkara. Seeds of Bhagyalakshmy variety were treated with the KAU culture of *Rhizobium* sp. (strain Rh4), with three to four seeds sown in each pot. 75% of the recommended fertilizer dose as per the Package of Practices recommendations was uniformly applied across all treatments. The talc-based consortia and PGPR Mix-2 were applied as soil treatments, and Bavistin (0.1% Carbendazim) was applied as foliar spray one week after sowing, followed by a second application one month later. The talc-based consortium was used at a rate of 10 g per 5 kg of potting mixture (10 g per polybag). The population of actinobacteria in one g of the talc-based formulation was 10^8 CFU ml⁻¹. *Rhizoctonia solani* was mass multiplied using paddy seeds and incubated at 28 ± 2 °C for two weeks. After 15 days of application of consortia, plants were challenge-inoculated with *R. solani*. Then upon symptom appearance, the plants were drenched with the consortia again at 10-day intervals thrice.

Biometric and yield parameters were assessed at regular intervals up to harvest. Observations on collar rot disease incidence were recorded from 60 DAS (after five days of application of actinobacterial consortia, until harvest at an interval of 10 days. Disease incidence was calculated using the formula:

$$\text{Disease incidence (\%)} = \frac{\text{Number of infected plants}}{\text{Total number of plants observed}} \times 100$$

The severity of web blight was observed before treatment application and again at 10 and 20 days after the application of treatments. The infection level was assessed by examining the affected plant parts, considering the size of lesions, along with the yellowing and drying of infected leaves. A 0-9 scale [15] was used for scoring the disease.

Percentage of Disease Index (PDI) of web blight was calculated as follows:

$$\text{PDI} = \frac{\text{Sum of grades of each leaf}}{\text{Number of leaves assessed}} \times \frac{100}{\text{Maximum grade used}}$$

Statistical analysis

Analysis of variance (ANOVA) suitable to CRD was performed on the collected data using GRAPES version 1.0.0 [16] statistical software.

RESULTS AND DISCUSSION

Actinobacteria are recognized for their metabolic diversity and production of bioactive compounds. They release natural products which can inhibit the growth of pathogenic bacteria and fungi. Several *Streptomyces* species have been reported to produce enzymes that degrade components of fungal cell walls, including chitinases, hemicellulases, cellulases, and glucanases [17-18]. The role of these enzymes in contributing to the antifungal activity and biocontrol capabilities of *Streptomyces* was investigated [19].

Cowpea is an important pulse crop but faces several operational constraints, including pests and diseases that hinder its production and yield from seedling through harvest. Collar rot and web blight caused by *Rhizoctonia solani* Kuhn is a major soil-borne disease significantly hindering the cultivation of this high-value crop. The prevalence of high temperature and humidity aggravates the situation and results in severe yield losses [20]. Although the pathogen can be controlled with extensive and repeated fungicide applications [21], the adverse effects of these chemicals on human health and the environment, along with the high costs, highlight the need for more affordable and environmentally sustainable disease management strategies. Therefore, cowpea was selected as a test crop for testing the efficiency of actinobacteria for plant growth promotion and disease management. In this context, the present study focused on the isolation of actinobacteria from different soil and compost samples, followed by their characterization and evaluation of the antagonistic potential of actinobacteria in managing collar rot and web blight in cowpea.

Isolation of actinobacteria from soil and compost samples

Actinobacteria were isolated using starch casein agar, following serial dilution plate method. Among different soil and compost samples analyzed, cowpea rhizosphere soil (CR) recorded a significantly superior population of actinobacteria with a population of 42.9×10^6 cfu g⁻¹. This was followed by coir pith compost, which recorded a population of 19.0×10^6 cfu g⁻¹. The lowest population was recorded in mangrove forest soil and ginger rhizosphere soil with a population of 1.1×10^6 cfu g⁻¹ and 1.2×10^6 cfu g⁻¹ respectively. Reports indicate that rhizosphere soils are good sources of microorganisms, due to the presence of root exudates. For instance, Sreevidya *et al.* [22] isolated actinobacteria from the rhizosphere of chickpea with plant growth promotion. Gopalakrishnan *et al.* [23] used herbal vermicompost to isolate actinobacteria, which were then

employed for the biological control of the *Fusarium* wilt of chickpea.

Predominant colonies were purified by repeated sub-culturing on starch casein agar medium. A total of 50 actinobacteria (including 21 isolates from the Department of Agricultural Microbiology repository) with different morphological characters were selected for further studies.

In vitro screening of actinobacterial isolates for indirect PGP traits

Fifty isolates of actinobacteria were screened *in vitro* for various indirect PGP activities such as the production of HCN, ammonia, and siderophores. Microbial production of HCN has been suggested as an important antifungal activity to control root and soil-borne pathogens. Hydrogen cyanide is a secondary metabolite that hinders the growth and development of competing microorganisms by inhibiting several metal enzymes, especially copper-containing cytochrome c oxidases [24]. In the present investigation, none of the isolates exhibited the ability for HCN production under *in vitro* conditions. This is contradictory to a report by Chaiham *et al.* [25] where four *Streptomyces* species isolated from rice rhizosphere produced HCN and exhibited significantly higher growth inhibition of *Pyricularia* sp. (87.3%, 80.0%, 82.2%, and 80.5) in a dual culture plate.

Another mechanism for the inhibition of plant pathogens by antagonistic microbes is the production of ammonia. This is a form of nitrogen, which can affect plant-pathogen interactions by altering the defensive capabilities of plants and the virulence of the pathogen. In the present investigation, among 50 isolates, 12 isolates (CS-4, Cc-6, CS-6, WA-7, C-2, S4-5, VC-5, DPS-5, CR-3, Cc-4, CR-4, and DPS-6) exhibited ammonia production. Kaur *et al.* [26] made similar observations, by detecting ammonia production in 13 out of 62 actinobacterial isolates.

Actinobacteria can produce siderophores that facilitate iron uptake as well as the production of antibacterial and antifungal compounds that protect plants from pathogen infection [27]. In the present study, nine isolates (US-3, US-4, Cc-1, S2-2, WA-26, WA-22, CS-9, S4-2, and VC-5) exhibited yellow to orange zones around the colonies. Siderophores chelate the available iron and prevent the iron nutrition of respective phytopathogens [28]. For example, *Streptomyces* PC 12 has been described as an effective biocontrol agent against *Pyricularia* sp infection as well as to boost rice growth under iron deficiency conditions [25].

Secondary screening for antagonistic traits against *Rhizoctonia solani* under *in vitro* conditions

Twenty isolates exhibiting indirect mechanisms of PGP traits in primary screening were tested for antagonistic activity against the fungal pathogen *Rhizoctonia solani* under *in vitro* conditions by the dual culture method. Ten isolates (CS-4, S2-2, US-3, US-4, CR-3, CR-4, DPS-5, Cc-6, Cc-4, and Cc-1) exhibited inhibition of *Rhizoctonia solani* (Fig 1). Among the 10 isolates, significantly higher inhibition percentages were observed in CS-4, S2-2, US-3, US-4, CR-3, and DPS-5, with inhibition rates of 74.0%, 70.3%, 65.1%, 64.8%, 63.7%, and 62.9%, respectively. The 10 promising isolates exhibiting antagonistic activity *in vitro* against the fungal pathogen *R. solani*, were further tested for compatibility.

Compatibility of potential actinobacteria exhibiting antagonistic activity

The 10 selected promising isolates were tested for compatibility by cross streak method. Among all the possible combinations of 10 isolates containing two in each

combination, only five combinations (CR-3 x US-4, US-4 x DPS-5, CR-3 x S2-2, DPS-5 x CS-4 and CR-3 x DPS-5) exhibited compatibility and others were non-compatible. These five combinations of isolates were selected for the preparation of five different talc-based consortia. Inconsistent field

performance of bioinoculant applied individually could be overcome by using combinations of several different types of microbial strains and therefore consortial formulations are of great importance in sustainable agriculture.

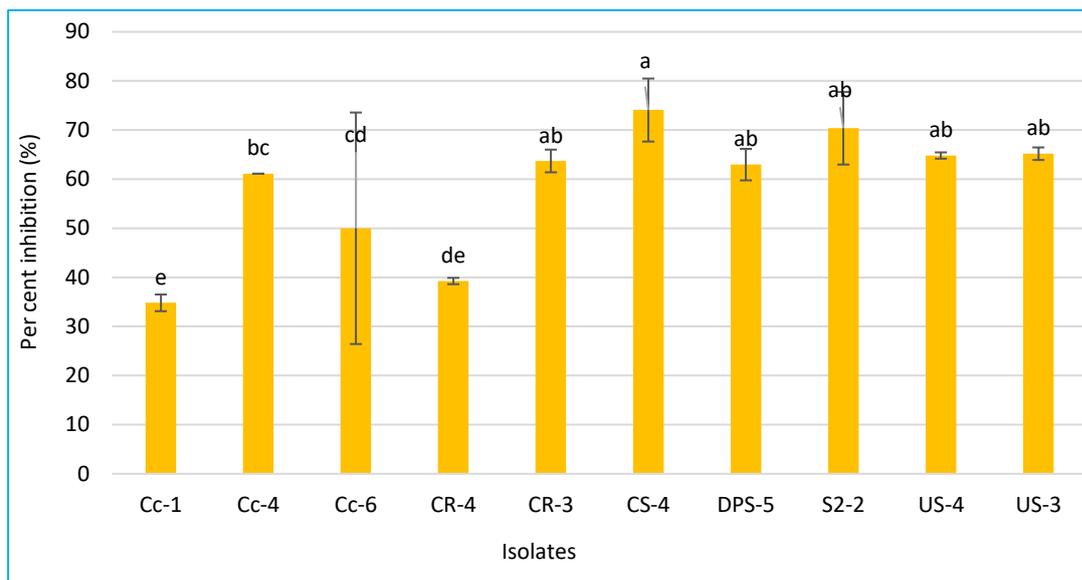


Fig 1 Antagonistic activity of actinobacterial isolates against *Rhizoctonia solani* in secondary screening

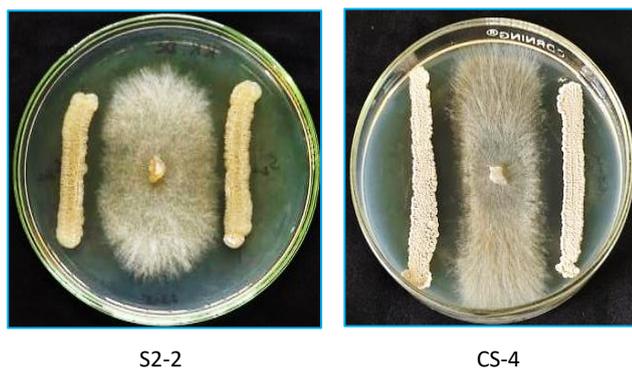


Fig 2 Secondary screening for antagonistic traits against *Rhizoctonia solani* under *in vitro* condition

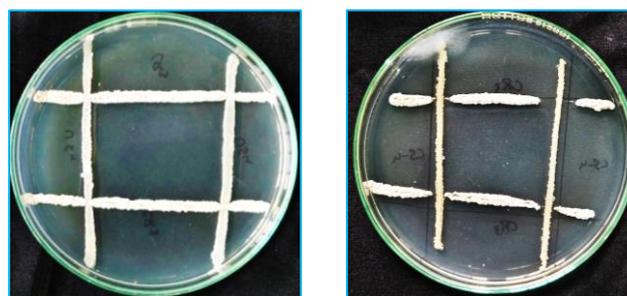


Fig 3 Compatibility of potential actinobacteria with antagonistic activity

Evaluation of selected actinobacterial consortia for management of collar rot and web blight in cowpea

The antagonistic activity of actinobacterial consortia was evaluated *in planta* under greenhouse conditions using a sterile potting mixture. Seven treatments, each with four replications, were maintained. The first five treatments were the selected antagonistic consortia, T₁: consortium 1 (CR-3 + US-4), T₂: consortium 2 (DPS-5 + US-4), T₃: consortium 3 (CR-3 + S2-2), T₄: consortium 4 (DPS-5 + CS-4), and T₅: consortium 5 (DPS-5 + CR-3). T₆ consisted of PGPR Mix-2 from KAU, and chemical treatment with Carbendazim at 0.1% was included as the seventh treatment (T₇) to compare the efficacy of actinobacteria with the fungicide application.

The biometric parameters of cowpea are recorded and the number of effective nodules was significantly higher in all treatments except for T₇, chemical control. This could be due to the synergistic effect of actinobacteria and *Rhizobium* in actinobacterial treatments. Earlier reports indicated that actinomycete co-inoculation with *Bradyrhizobium japonicum* further stimulated soybean growth through increasing nitrogenase activity of root nodules, nutrients uptake, and seed weight at harvest compared to plants

inoculated only with *B. japonicum* [29]. Treatment with consortium 3 (CR-3 and S2-2) significantly increased yield attributes including the number of pods per plant (29.3), seeds per pod (10.5), test weight (20.0 g), fresh weight of pods (171.0 g plant⁻¹), and dry weight of pods (19.9 g plant⁻¹), compared to all the other treatments. This indicated that by enhancing plant growth and inducing early flowering, these actinobacterial isolates improved the yield also in cowpea.

Observations on disease incidence of collar rot from 60 DAS (after five days of application of actinobacterial consortia, PGPR Mix-2 and carbendazim at 0.1%) until harvest at an interval of 10 days are presented in (Table 2). At 60 DAS, the per cent disease incidence (%) of collar rot in cowpea was significantly lower in treatments T₃, T₄, T₆, and T₇ (16.6%, 24.9%, 22.2% and 13.8% respectively), as compared to the other treatments (Fig 3). At 70 DAS, T₃ and T₇ recorded significantly lower incidence of collar rot (16.6% and 11.1% respectively). At 80 DAS and harvest, T₃ (consortium 3 (CR-3 + S2-2)), T₆ (PGPR Mix-2 of KAU) and T₇ (Carbendazim at 0.1%) recorded significantly lower per cent disease incidence of 11.1%, 19.42% and 11.1%. The actinobacterial isolates CR-3 and S2-2 contributed to reducing the per cent disease

incidence as they showed higher per cent inhibition of 63.7% and 70.3% under in vitro conditions against *Rhizoctonia solani*. An investigation by Singh *et al.* [30] also revealed that

actinobacteria from the *Streptomyces* genus decreased the disease incidence caused by *Rhizoctonia solani* in tomato plants by 47-63%.

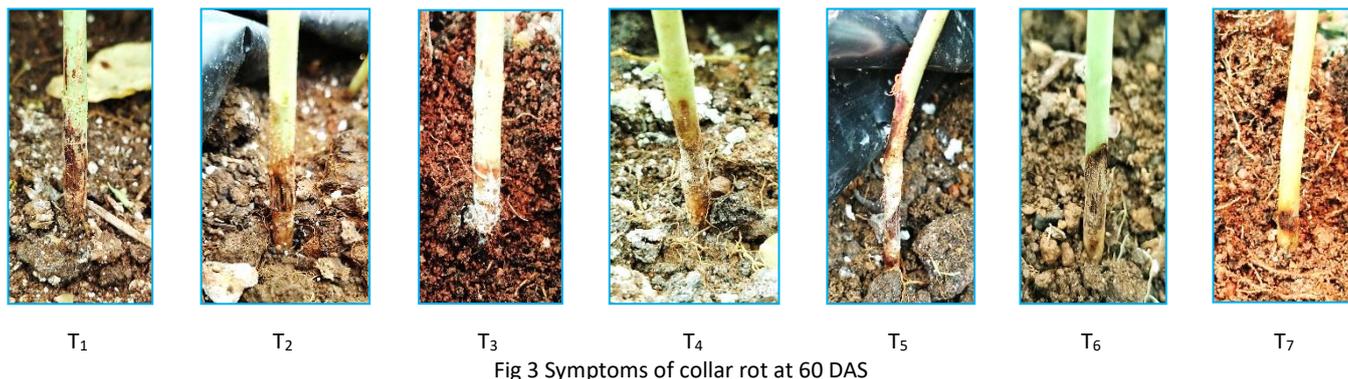


Fig 3 Symptoms of collar rot at 60 DAS

Per cent Disease Index (PDI) was calculated based on the damage caused by *R. solani* to the foliage. Disease scoring was conducted using a 0-9 scale [15] (Fig 4). Initial observations before imposing the treatments, PDI of web blight of cowpea was significantly higher in five treatments: T₁ (41.0%), T₂ (42.85%), T₄ (35.9%), T₅ (44.7%) and T₆ (37.30%). After 10 days of treatment application (actinobacteria/PGPR/Carbendazim), PDI was significantly reduced in T₇ (23.4%) and T₃ (25.6%) when compared to other treatments (Table 3). Similarly, after 20 days of treatment application, PDI

was significantly reduced to 19.4% in T₇ (Carbendazim at 0.1%) and 21.6% in T₃ (consortium 3 (CR-3 + S2-2)). This indicated that T₃ consortium containing CR-3 and S2-2 isolates of actinobacteria could be a possible alternative to the chemical for the management of web blight and collar rot in cowpea. This is in agreement with the findings of Diaz *et al.* [31], who reported that the combination of two *Streptomyces* strains (CBQ-EA-2 and CBQ-B-8) significantly reduced the disease severity caused by *R. solani* compared to the untreated and inoculated control.

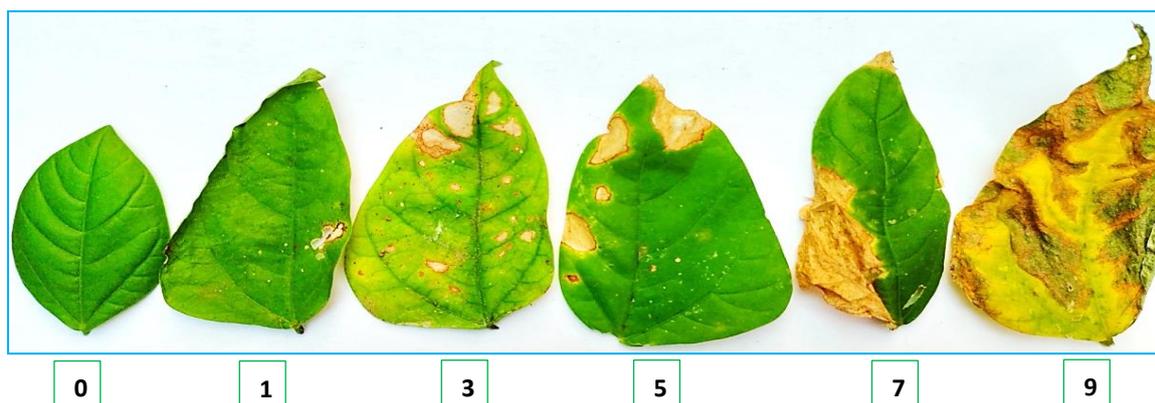


Fig 4 Grades for scoring web blight of cowpea

In planta evaluation, treatment with consortium 3 (CR-3 + S2-2) was equally effective as chemical control (carbendazim) in significantly reducing the incidence of collar rot and severity of web blight in cowpea. The same treatment also significantly increased the yield attributes. Later identification by 16S rRNA gene sequencing revealed that the isolates CR-3 and S2-2 were closely related to *Streptomyces*

pratensis and *Streptomyces ramulosus*, respectively. The antagonistic mechanisms of these isolates were confirmed by *in vitro* results, where CR-3 exhibited a positive reaction for ammonia production, and S2-2 showed a positive reaction for siderophore production. Thus, consortium 3 (CR-3 and S2-2) could be used as a potential alternative to the chemical Carbendazim.

Table 2 Effect of different treatments on per cent (%) disease incidence of collar rot in cowpea at various intervals

Treatments	Per cent disease incidence of collar rot in cowpea (%)			
	60 DAS	70 DAS	80 DAS	At harvest
T ₁ : Consortium 1 (CR-3 + US-4)	33.3 (35.1) ^{ab}	30.5 (33.3) ^{ab}	27.7 (31.1) ^{abc}	27.7 (31.1) ^{abc}
T ₂ : Consortium 2 (DPS-5 + US-4)	27.7 (31.1) ^{ab}	24.9 (29.9) ^{ab}	30.5 (33.4) ^{ab}	30.5 (33.4) ^{ab}
T ₃ : Consortium 3 (CR-3 + S2-2)	16.6 (23.8) ^{bc}	16.6 (23.8) ^{bc}	11.1 (21.6) ^{cd}	11.1 (21.6) ^{cd}
T ₄ : Consortium 4 (DPS-5 + CS-4)	24.9 (29.5) ^{abc}	27.7 (31.5) ^{ab}	27.7 (31.5) ^{abc}	27.7 (31.5) ^{abc}
T ₅ : Consortium 5 (DPS-5 + CR-3)	41.6 (40.1) ^a	41.6 (40.1) ^a	41.6 (40.1) ^a	38.8 (38.5) ^a
T ₆ : PGPR Mix-2 of KAU	22.2 (27.7) ^{bc}	22.2 (27.7) ^b	19.4 (25.5) ^{bcd}	19.4 (25.5) ^{bcd}
T ₇ : Carbendazim at 0.1%	13.8 (19.1) ^c	11.1 (16.7) ^c	11.1 (16.7) ^d	11.1 (16.9) ^d
CD (0.05)	11.92	10.38	11.22	11.27

*DAS - Days after sowing

Figures given in parenthesis are transformed values

Means followed by common letter(s) are not significantly different by one-way ANOVA at $P = 0.05$

Table 3 Effect of different treatments on per cent disease index (PDI) of web blight in cowpea

Treatments	Per cent disease index	PDI at 10 days of	PDI
	(PDI*) before treatment application	application of treatments	At 20 days of application of treatments
T ₁ : Consortium 1 (CR-3 + US-4)	41.0 (39.8) ^a	38.0 (38.0) ^a	34.4 (35.6) ^a
T ₂ : Consortium 2 (DPS-5 + US-4)	42.8 (40.8) ^a	39.8 (39.1) ^a	37.4 (36.7) ^a
T ₃ : Consortium 3 (CR-3 + S2-2)	30.8 (33.6) ^b	25.6 (30.3) ^{bc}	21.6 (27.6) ^{bc}
T ₄ : Consortium 4 (DPS-5 + CS-4)	35.9 (36.7) ^{ab}	32.9 (34.9) ^{ab}	30.7 (32.4) ^{ab}
T ₅ : Consortium 5 (DPS-5 + CR-3)	44.7 (41.9) ^a	41.7 (40.2) ^a	41.3 (37.8) ^a
T ₆ : PGPR Mix-2 of KAU	37.3 (37.6) ^{ab}	34.3 (35.8) ^{ab}	30.3 (33.3) ^{ab}
T ₇ : Carbendazim at 0.1%	29.9 (33.0) ^b	23.4 (28.5) ^c	19.4 (25.5) ^c
CD (0.05)	5.85	6.32	6.68

*PDI - Per cent Disease Index

Figures given in parenthesis are transformed values

Means followed by common letter(s) are not significantly different by one-way ANOVA at $P = 0.05$

CONCLUSION

The present investigation revealed that treatment with consortium 3 consisting of *Streptomyces pratensis* strain CR-3 and *Streptomyces ramulosus* strain S2-2 was equally effective as chemical control (carbendazim) in significantly reducing the incidence of collar rot and severity of web blight in cowpea. This suggests that native actinobacteria are promising candidates for the biological control of *R. solani* in cowpea.

Exploration of the secondary metabolites produced by actinobacteria for novel antimicrobial, antifungal, and biostimulant compounds and also the potential of actinobacteria to mitigate abiotic stresses, such as drought, salinity, and nutrient deficiency may be carried out. These isolates may further be evaluated under field conditions before commercialization. Thus, Actinobacteria represent a promising, eco-friendly, and sustainable alternative to pesticides in agriculture.

LITERATURE CITED

- Lewin GR, Carlos C. Chevrette MG, Horn HA, McDonald BR, Stankey RJ, Fox BG, Currie CR. 2016. Evolution and ecology of Actinobacteria and their bioenergy applications *Annu. Rev. Microbiology* 70(1): 235-254.
- Gopalakrishnan S, Srinivas V, Sree Vidya M, Rathore A. 2013. Plant growth-promoting activities of *Streptomyces* spp. in sorghum and rice. *Springer Plus* 2: 1-8.
- Alexander M. 1978. Introduction to soil microbiology. *Soil Science* 125(5): 331.
- Welbaum GE, Sturz AV, Dong Z, Nowak J. 2004. Managing soil microorganisms to improve productivity of agro-ecosystems. *Crit. Rev. Plant Science* 23(2): 175-193.
- Martínez-Hidalgo P, García JM, Pozo MJ. 2015. Induced systemic resistance against *Botrytis cinerea* by *Micromonospora* strains isolated from root nodules. *Front. Microbiology* 6: 922.
- Wu ZM, Yang Y, Li KT. 2019. Antagonistic activity of a novel antifungalmycin N₂ from *Streptomyces* sp. N₂ and its biocontrol efficacy against *Rhizoctonia solani*. *FEMS Microbiology Letters* 366(3): 18.
- Suryawanshi PP, Krishnaraj PU, and Suryawanshi MP. 2020. Evaluation of actinobacteria for biocontrol of sheath blight in rice. *Jr. Pharmacogn. Phytochemistry* 9(3): 371-376.
- Vurukonda SSKP, Giovanadri D, Stefani E. 2021. Growth promotion and biocontrol activity of endophytic *Streptomyces* spp. *Prim. Arch. Mol. Sci.* 2nd Edition 1: 1-55.
- Johnson LF, Curl EA. 1972. Methods for research on the ecology of soil borne plant pathogens, Burgess, Minneapolis. *Jr. Biotechnology* 7(8): 967-972.
- Cappucino JC, Sherman N. 1992. Nitrogen cycle. Microbiology: A laboratory manual. 3rd Edition, Benjamin/Cumming Pub. Co., New York.
- Schwyn B, Neilands J. 1987. Universal chemical assay for the detection and determination of siderophores. *Anal. Biochemistry* 160(1): 47-56.
- Lorck H. 1948. Production of hydrocyanic acid by bacteria. *Physiology Plant* 1(2): 142-146.
- Xu SJ, Kim BS. 2014. Biocontrol of *Fusarium* crown and root rot and promotion of growth of tomato by *Paenibacillus* strains isolated from soil. *Mycobiology* 42(2): 158-166.
- Al-Hussini HS, Al-Rawahi AY, Al-Marhoon AA, Al-Abri SA, Al-Mahmooli IH, Al-Sadi AM, Velazhahan R. 2019. Biological control of damping-off of tomato caused by *Pythium aphanidermatum* by using native antagonistic rhizobacteria isolated from Omani soil. *Jr. Plant Pathology* 101: 315-322.
- Mayee CD, Datar VV. 1986. *Phytopathometry*. Tech. Bull.1. University Press. Marathwada Agriculture University, Parbhani (M.S.). pp 186.
- Gopinath PP, Parsad R, Joseph B, Adarsh VS. 2021. GrapesAgril: collection of shiny apps for data analysis in agriculture. *Jr. Open-Source Software* 6(63): 3437.
- Petrosyan P, Garcia-Varela M, Luz-Madrigal A, Huitron C, Flores ME. 2003. *Streptomyces mexicanus* sp. nov., a xylanolytic micro-organism isolated from soil. *Int. Jr. Syst. Evol. Microbiology* 53(1): 269-273.
- Ding CH, Jiang ZQ, Li XT, Li LT, Kusakabe I. 2004. High activity xylanase production by *Streptomyces olivaceoviridis* E-86. *World Jr. Microbiol. Biotechnology* 20: 7-10.
- Ramesh S, Rajesh M, Mathivanan N. 2009. Characterization of a thermostable alkaline protease produced by marine *Streptomyces fungicidicus* MML1614. *Bioprocess Biosyst. Engineering* 32: 791-800.

20. Vavilapalli S, Celine VA, Girija VK. 2014. Collar rot and web blight caused by *Rhizoctonia solani* Kuhn in vegetable cowpea (*Vigna unguiculata* (L) Walp.) and its organic management. *Agrotechnology* 2: 4.
21. Upamanyu S, Gupta SK, Shyam KR. 2002. Innovative approaches for the management of root rot and web blight (*Rhizoctonia solani*) of French bean. *Jr. Mycol. Plant Pathology* 32: 317-331.
22. Sreevidya M, Gopalakrishnan S, Kudapa H, Varshney RK. 2016. Exploring plant growth-promotion actinomycetes from vermicompost and rhizosphere soil for yield enhancement in chickpea. *Braz. Jr. Microbiology* 47: 85-95.
23. Gopalakrishnan S, Pande S, Sharma M, Humayun P, Kiran BK, Sandeep D, Vidya MS, Deepthi K, Rupela O. 2011. Evaluation of actinomycete isolates obtained from herbal vermicompost for the biological control of *Fusarium* wilt of chickpea. *Crop Protection* 30(8): 1070-1078.
24. Hassanein WA, Awny NM, El-Mougith AA, El-Dein SH. 2009. Characterization and antagonistic activities of metabolite produced by *P. aeruginosa* Sha8. *Jr. Appl. Sci. Research* 5(4): 392-403.
25. Chaiham M, Theantana T, Pathom-Aree W. 2020. Evaluation of biocontrol activities of *Streptomyces* spp. against rice blast disease fungi. *Pathogens* 9(2): 126.
26. Kaur T, Sharma D, Kaur A, Manhas RK. 2013. Antagonistic and plant growth promoting activities of endophytic and soil actinomycetes. *Arch. Phytopathol. Plant Protection* 46(14): 1756-1768.
27. Viaene T, Langendries S, Beirinckx S, Maes M, Goormachtig S. 2016. *Streptomyces* as a plant's best friend? *FEMS Microbiology and Ecology* 92: 119.
28. Sadeghi A, Karimi E, Dahaji PA, Javid MG, Dalvand Y, Askari H. 2012. Plant growth promoting activity of an auxin and siderophore producing isolate of *Streptomyces* under saline soil conditions. *World Jr. Microbiology and Biotechnology* 28: 1503-1509.
29. Soe KM, Bhromsiri A, Karladee D. 2010. Effects of selected endophytic actinomycetes (*Streptomyces* sp.) and *Bradyrhizobia* from Myanmar on growth, nodulation, nitrogen fixation and yield of different soybean varieties. *CMU Journal of Nat. Science* 9: 95-109.
30. Singh SP, Gupta R, Gaur R, Srivastava AK. 2017. Antagonistic actinomycetes mediated resistance in *Solanum lycopersicon* Mill. against *Rhizoctonia solani* Kühn. *Proc. Natl. Acad. Sci. India Sect. B Biol. Science* 87: 789-798.
31. Díaz-Díaz M, Bernal-Cabrera A, Trapero A, Medina-Marrero R, Sifontes-Rodríguez S, Cupull-Santana RD, García-Bernal M, Agustí-Brisach C. 2022. Characterization of actinobacterial strains as potential biocontrol agents against *Macrophomina phaseolina* and *Rhizoctonia solani*, the main soil-borne pathogens of *Phaseolus vulgaris* in Cuba. *Plants* 11(5): 645.