

Biocontrol and growth-promoting potential of *Bacillus cereus* TuR6 against rhizome rot disease in turmeric (*Curcuma longa* L.)

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Abstract: Rhizome rot disease caused by *Fusarium solani* (Mart.) Sacc. severely limits turmeric (*Curcuma longa* L.) productivity. This study aimed to isolate endophytic bacterial strains from turmeric roots and screen them for plant growth-promoting (PGP) traits and antifungal activity. Among 10 isolates, strain TuR6, identified as a member of the *Bacillus cereus* Frankland & Frankland 1887 group (98.68 % similarity, GenBank accession: PV565573), showed the highest IAA production (75.41 mg l⁻¹), strong phosphate solubilization (PSI 5.32), and positive siderophore, protease, and cellulase activities. TuR6 inhibited *F. solani* growth by 78.12 % *in vitro*. In greenhouse trials, TuR6 significantly increased plant height (80.57 cm), rhizome yield (371 g/plant), and reduced disease incidence (17.7 %). Field experiments confirmed its efficacy, with notable improvements in growth and disease suppression under natural conditions. These results highlight *B. cereus* TuR6 as a potential bioinoculant for enhancing turmeric yield and managing rhizome rot sustainably.

Key words: endophytic bacteria, plant growth-promoting bacteria, biocontrol, turmeric roots, biofertilizer.

Biokontrola in potencial spodbujanja rasti bakterije *Bacillus cereus* TuR6 proti gnilobi korenike pri kurkumi (*Curcuma longa* L.)

Izvleček: Gniloba korenike, ki jo povzroča gliva *Fusarium solani* (Mart.) Sacc., močno omejuje produktivnost kurkume (*Curcuma longa* L.). Namen te raziskave je bil izolirati seve endofitnih bakterij iz korenin kurkume in jih pregledati za lastnosti, ki spodbujajo rast rastlin (PGP) in protiglivično delovanje. Med 10 izolati je sev TuR6, identificiran kot član skupine *Bacillus cereus* Frankland & Frankland 1887 (98,68 % podobnost, dostop GenBank: PV565573) pokazal največjo produkcijo IAA (75,41 mg l⁻¹), močno topnost fosfata (PSI 5,32) in pozitivne sideroforne, proteazne in celulazne aktivnosti. Sev TuR6 je zaviral raste glive *F. solani* za 78,12 % *in vitro*. V poskusih v rastlinjaku je sev TuR6 znatno povečal višino rastlin (80,57 cm), donos korenin (371 g/rastlino) in zmanjšal pojavnost bolezni (17,7 %). Terenski poskusi so potrdili njegovo učinkovitost z opaznimi izboljšavami v rasti in zatiranju bolezni v naravnih razmerah. Ti rezultati poudarjajo uporabo seva *B. cereus* TuR6 kot potencialnega bioinokulanta za povečanje donosa kurkume in trajnostno obvladovanje gnilobe korenin.

Ključne besede: endofitne bakterije, bakterije, ki pospešujejo rast rastlin, biokontrola, korenine kurkume, biognojilo

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1 INTRODUCTION

Turmeric (*Curcuma longa* L.), a perennial herbaceous plant belonging to the Zingiberaceae family, is widely cultivated in tropical regions for its economic and medicinal value (Sharma *et al.*, 2024). The rhizome of turmeric is renowned not only as a culinary spice and natural dye but also for its pharmacological properties (Sun *et al.*, 2024; Sharma *et al.*, 2024). Among its numerous bioactive compounds, curcumin - the primary curcuminoid - exhibits potent antioxidant, anti-inflammatory, antimicrobial, and cardioprotective effects (Sontsa-Donhoung *et al.*, 2022; Sharma *et al.*, 2024).

Despite its broad utility, soil-borne pathogens, particularly *Fusarium solani* (Mart.) Sacc, hinder turmeric cultivation, causing rhizome rot (Vinayarani & Prakash, 2018; Sontsa-Donhoung *et al.*, 2022). This disease significantly reduces yield and commercial quality. Although chemical fungicides such as Carbendazim and Mancozeb have traditionally been used for disease management, their long-term application raises concerns regarding environmental and human health and may induce resistance in target pathogens (Vinayarani & Prakash, 2018; Xavier *et al.*, 2021; Sagarika *et al.*, 2025). Therefore, there is an urgent need for sustainable alternatives, such as biological control strategies.

An eco-friendly alternative gaining traction is using plant-associated beneficial microorganisms, particularly endophytic bacteria (Degrassi & Carpentieri-Pipolo, 2020; Sagarika *et al.*, 2025). These bacteria colonize internal plant tissues asymptotically and have shown promise as both biostimulants and biocontrol agents (Do, 2025; Sagarika *et al.*, 2025). Endophytes can enhance plant health via direct mechanisms—such as the synthesis of phytohormones (IAA, cytokinins, gibberellins), phosphate solubilization, and nitrogen fixation—and indirect mechanisms, including siderophore production, secretion of lytic enzymes (e.g., chitinases, proteases), and induction of systemic resistance (ISR) (Saryanah *et al.*, 2021; Tiwari *et al.*, 2023; Sagarika *et al.*, 2025).

Recent research underscores the potential of turmeric-associated endophytes, such as *Bacillus cereus* Frankland & Frankland 1887 and *Pseudomonas aeruginosa* (Schröter 1872) Migula 1900, to inhibit key pathogens and promote plant growth under greenhouse and field conditions (Vinayarani & Prakash, 2018; Virk *et al.*, 2023; Sharma *et al.*, 2024; Do & Nguyen, 2025). Their application has resulted in enhanced plant biomass and reduced disease severity. However, in Vietnam, data

on the diversity and biocontrol potential of endophytic bacteria from local turmeric cultivars remains sparse (Tuyet & Giang, 2017; Yen *et al.*, 2018; Do *et al.*, 2023).

This study aims to fill this gap by isolating and characterizing endophytic bacterial strains from turmeric rhizomes cultivated in Vietnam. The objectives were to (i) evaluate their plant growth-promoting (PGP) traits, (ii) assess antifungal activity against *F. solani*, and (iii) determine their efficacy in promoting growth and suppressing rhizome rot under greenhouse conditions. The ultimate goal is to identify potent bacterial bioinoculants for sustainable and environment-friendly turmeric production systems.

2 MATERIAL AND METHODS

2.1 ISOLATION OF ENDOPHYTIC BACTERIA

Turmeric root samples were collected from fields in Quat Lam commune, Giao Thuy district, Nam Dinh province, Vietnam. The isolation procedure followed the protocol of Do *et al.* (2023). Root samples were washed thoroughly under running water to remove soil and then cut into small segments. These segments were surface sterilized by soaking in 75 % ethanol for 2.5 minutes, rinsed once with distilled water, and then treated with 3 % sodium hypochlorite (NaClO) for 2 minutes. Afterward, the samples were rinsed again with sterile distilled water and soaked in 75 % ethanol for 30 seconds, followed by a final rinse with sterile distilled water.

To confirm surface sterility, 0.1 ml of the final rinse water was plated onto nutrient agar medium (NA, Pepton 5 g l⁻¹; NaCl 5 g l⁻¹; beef extract 2 g l⁻¹; yeast extract 3 g l⁻¹; agar 18 g l⁻¹) in Petri dishes and incubated at 30 °C. After 2 days, if no microbial growth appeared, surface sterilization was deemed effective. The sterilized root segments were then placed on NA medium in Petri dishes and incubated at 30 °C to observe endophytic microbial growth.

2.2 EVALUATION OF PLANT GROWTH-PROMOTING (PGP) TRAITS OF ENDOPHYTIC BACTERIAL ISOLATES

Bacterial strains were inoculated into 20 ml of NA medium supplemented with L-tryptophan, shaken at 200 rpm, and incubated at 28 °C. After 48 hours, the culture broth was centrifuged at 3,000 rpm

for 15 minutes, and the supernatant was collected. One milliliter of supernatant was mixed with 2 ml of Salkowski reagent and incubated in the dark for 30 minutes. The appearance of a red color indicated indole-3-acetic acid (IAA) production. The control tubes contained only bacteria-free medium (Do et al., 2023). IAA concentration was measured according to the National Standard for Microorganisms-Determination of IAA synthesis capability (TCVN 10784:2015, 2015).

Endophytic strains were evaluated for phosphate solubilization using the method of Jasim et al. (2014), with NBRIP medium replacing Pikovskaya's medium. Strains were spot-inoculated on NBRIP medium (National Botanical Research Institute's phosphate: glucose 10 g l⁻¹; Ca₃(PO₄)₂ 5 g l⁻¹; MgCl₂·6H₂O 5 g l⁻¹; MgSO₄·7H₂O 0.25 g l⁻¹; KCl 0.2 g l⁻¹; (NH₄)₂SO₄ 0.1 g l⁻¹; pH 7.0; agar 18 g l⁻¹; 1000 ml H₂O) and incubated at 30 °C for 3 days. Strains that produced clear halos around colonies were considered phosphate-solubilizing. The phosphate-solubilizing index (PSI) of a bacterial strain was calculated by the ratio of the clear zone and colony diameter (Jasim et al., 2014).

Isolates were tested on Chrome Azurol S (CAS) agar medium. A log-phase bacterial culture was applied onto nutrient agar plates supplemented with CAS solution. These plates were then incubated in the dark at 28 °C for a period of 3 to 5 days. The development of yellow to orange halos around the colonies indicated the production of siderophores. Each assay was performed in triplicate to ensure reproducibility (Jasim et al., 2014). The change from blue to orange indicated iron-chelating activity.

A loopful of the bacterial culture was streaked onto skim milk agar plates (composed of 100 g skim milk, 5 g peptone, 15 g agar, and 1000 ml distilled water) to assess protease activity. After incubating the plates at 28 °C for 48 hours, a clear halo around the bacterial streak indicated positive protease production.

For cellulase activity, carboxymethyl cellulose (CMC) was incorporated into a basal medium containing NaNO₃ (1 g), KCl (1 g), K₂HPO₄ (1 g), MgSO₄ (0.5 g), yeast extract (0.5 g), agar (15 g), and 1000 ml of distilled water. Bacterial strains were streaked onto this medium and incubated at 28 °C for three days. Following incubation, the plates were stained with 0.01 % Congo red for 15 minutes and subsequently washed with 1 % NaCl solution for 5 minutes. The appearance of a clear zone around the colonies confirmed the degradation of CMC, indicating positive cellulase activity (Cappuccino & Sherman, 1992).

2.3 MOLECULAR IDENTIFICATION

Genomic DNA was extracted from bacterial isolates using the One-Tube Bacteria Genomic DNA Isolation Kit (Biobasic, Canada) following the manufacturer's protocol. Amplification of the 16S rRNA gene was carried out using PCR with the primers: 16S rDNA forward primer (27F, 5'-AGAGTTTGATC-CTGGCTC-3') and reverse primer (1429R, 5'-GGC-TACCTTGTTACGACTT-3') (Do, 2025), procured from Phusa Genomics company (Can Tho, Vietnam). PCR products were subsequently purified and subjected to Sanger sequencing at the 1st Base company (Singapore). The obtained nucleotide sequences were analyzed using the BLAST tool available at the National Center for Biotechnology Information (NCBI; <http://www.ncbi.nlm.nih.gov>). The resulting sequences were submitted to GenBank, and accession numbers PV565573 were obtained.

2.4 ANTIFUNGAL ACTIVITY AGAINST *FUSARIUM SOLANI*

A highly virulent strain of *F. solani*, isolated from naturally infected turmeric rhizomes, was sourced from the culture collection of the Faculty of Biotechnology, Dai Nam University, Hanoi, Vietnam. Actively growing fungal hyphae were subcultured onto fresh PDA medium, and the resulting cultures were maintained on slants and stored at 4 °C.

The antifungal capabilities of the bacterial strains were assessed using the well diffusion technique, as outlined by Do et al. (2023). Fungi were cultured on PDA medium. After 72 hours, the plugs of PDA media with actively growing fungi were prepared and placed on other plates containing PDA media. Then, 0.2 ml of bacterial culture (approximately 2 × 10⁷ CFU/ml) was added to the wells created in the agar. The plates were incubated at 30 °C for 5 days, and clear inhibition zones surrounding the wells were noted as indicators of antifungal activity. The growth inhibition zone was calculated by using the formula:

$$\text{The growth inhibition zone (\%)} = [(D-d)/D] \times 100$$

In which D = Diameter of pathogenic mycelia growth in the absence of antagonists (mm), and d = Diameter of pathogenic mycelia growth in the presence of antagonists (mm).

2.5 EVALUATION OF ENDOPHYTIC BACTERIA FOR PLANT GROWTH PROMOTION AND DISEASE CONTROL UNDER GREENHOUSE CONDITIONS

Based on *in vitro* antagonistic activity and multiple plant growth-promoting (PGP) attributes, the bacterial isolate TuR6 was selected for greenhouse evaluation to determine its efficacy in suppressing rhizome rot in turmeric (*C. longa* 'N8'), a cultivar known for its susceptibility to such infections. The experimental procedure for disease suppression and plant growth assessment followed the methodology described by Vinayarani & Prakash (2018) and Prabhukarthikeyan *et al.* (2018).

Greenhouse trials were conducted from May 2022 to June 2023 at the College of Forestry Biotechnology, Vietnam National University of Forestry, Hanoi, Vietnam. The experimental design followed a completely randomized design (CRD), with three replicates per treatment, and each replicate consisting of five pots (20 cm diameter), each pot grown with one 5–6 cm long turmeric rhizome (approximately weighing 20–25 g). To ensure reproducibility, the entire experiment was performed twice.

A talc-based formulation of the selected bacterial isolate was prepared, standardized to a final concentration of approximately 3×10^8 CFU per gram of carrier. Before planting, turmeric rhizomes were surface-sterilized using 2 % sodium hypochlorite, followed by immersion in a bacterial suspension (20 g l⁻¹ of formulation) for 12 hours. The treated rhizomes were air-dried overnight before being planted in pots filled with 5 kg of sterilized soil.

The pathogen challenge was initiated 30 days after planting by inoculating *Fusarium solani* via a sand-maize medium (1:19 ratio of inoculum to soil). Each treatment received three soil applications of the bacterial formulation (8 g per application) at 15-day intervals over 90 days. For comparative purposes, a chemical control treatment using a combination of Carbendazim (0.1 %) and Mancozeb (0.25 %) was applied through both rhizome treatment and soil drenching.

Control treatments included: (1) uninoculated and untreated plants, (2) plants inoculated with the pathogen only, and (3) plants treated with fungicides. Disease incidence was assessed by calculating the Percent Disease Index (PDI) based on the number of infected plants per treatment. In a parallel experimental setup, growth-promoting effects were evaluated by measuring plant height and fresh rhizome mass at harvest.

2.6 EVALUATION OF ENDOPHYTIC BACTERIA FOR PLANT GROWTH PROMOTION AND DISEASE CONTROL UNDER FIELD CONDITIONS

Field experiments were conducted in Hoang Trung village, Hong Duong district, Thanh Oai, Hanoi, Vietnam, from June 2023 to June 2024. The study was arranged in a randomized block design (RBD) with three replications per treatment. Each experimental plot measured 3.0 × 1.0 m and contained 40 uniformly sized turmeric rhizomes (fingers), each approximately 5–6 cm long and weighing 20–25 g. Rhizomes were sown in four rows at a depth of 3–4 cm, with 30 × 15 cm spacing both within and between rows. Standard agronomic practices were followed throughout the experimental period. Treatment protocols were consistent with those employed in the preceding greenhouse trials, and an untreated control was maintained for comparison. Data were collected on rhizome rot incidence, plant height, and fresh rhizome mass using the same methodology as described in the greenhouse evaluation.

2.7 DATA ANALYSIS

All experimental data underwent statistical analysis using SPSS software version 17.0 (SPSS Inc., Chicago, IL, USA) and Microsoft Excel 365 (Microsoft Corp., Redmond, WA, USA). For statistical analysis, percentage data were subjected to an arcsine square root transformation, expressed as $\arcsin(\sqrt{x/100})$, where x denotes the disease index. Treatment means were compared using Tukey's Honest Significant Difference (HSD) test, and differences were considered statistically significant at $p \leq 0.05$.

3 RESULTS AND DISCUSSION

3.1 ISOLATION AND CHARACTERIZATION OF PLANT GROWTH-PROMOTING TRAITS

Ten endophytic bacterial strains (TuR1 to TuR10) were isolated from surface-sterilized turmeric roots. All isolates could synthesize indole-3-acetic acid (IAA), with concentrations ranging from 2.25 to 75.41 mg l⁻¹. Among these, TuR6 exhibited the highest IAA production, suggesting strong potential for promoting root elongation and development (Table 1). Similar findings were reported by Saryanah *et al.* (2021), who observed IAA production among *Bacillus* and *Pseudo-*

Table 1: Characterization of endophytic bacterial isolates for plant growth-promoting abilities

Isolates	IAA production (mg l ⁻¹)	PSI	Protease activity	Cellulase activity	Siderophore production
TuR1	2.25 ± 0.12 ^f	1.73 ± 0.11 ^{bc}	-	-	+
TuR2	6.21 ± 0.15 ^e	1.41 ± 0.19 ^c	+	+	+
TuR3	12.23 ± 0.31 ^c	-	+	-	-
TuR4	15.51 ± 0.23 ^b	3.15 ± 0.21 ^b	+	+	-
TuR5	10.52 ± 0.11 ^d	1.72 ± 0.10 ^{bc}	+	-	-
TuR6	75.41 ± 0.25 ^a	5.32 ± 0.15 ^a	+	+	+
TuR7	17.62 ± 0.43 ^b	1.33 ± 0.09 ^{cd}	+	+	-
TuR8	12.14 ± 0.15 ^c	1.12 ± 0.15 ^d	+	+	-
TuR9	10.53 ± 0.09 ^d	-	+	+	+
TuR10	11.15 ± 0.072 ^d	-	+	-	+

Values represent the mean of three replicates ± standard error (SE). Means followed by the same letter within a column are not significantly different at $p \leq 0.05$ according to Tukey's Honest Significant Difference (HSD) test. PSI: phosphate-solubilizing index.

monas isolates from *Curcuma xanthorrhiza* Roxb. that significantly promoted seedling growth.

In addition, phosphate solubilization was observed in seven of the ten isolates, with PSI ranging from 1.12 to 5.32. The highest PSI was recorded for TuR6, indicating its strong ability to convert insoluble phosphates into bioavailable forms. This trait is critical for enhancing plant phosphorus uptake, especially under nutrient-limited conditions (Mukherjee et al., 2021; Sagarika et al., 2025).

Furthermore, siderophore production was detected in four isolates. TuR6 again showed positive activity, as indicated by the formation of orange halos on CAS agar. Siderophores facilitate iron acquisition and play a key role in suppressing phytopathogens through competitive iron sequestration (Ahmed and

Holmström, 2014; Eid et al., 2021; Shalovylo et al., 2025). Ahmed and Holmström (2014) emphasized the significance of microbial siderophores in both plant nutrition and pathogen inhibition.

In the present study, protease and cellulase activities were detected in 9 and 6 bacterial isolates, respectively (Table 1). The enzymatic activities of cellulase facilitate microbial colonization through the degradation of plant cell wall components, thereby enabling penetration into plant tissues. Moreover, protease and cellulase are recognized for their roles in the biological control of phytopathogens, contributing to plant protection through degradation of pathogenic structures (Trung et al., 2023; Sharma et al., 2024; Sagarika et al., 2025).

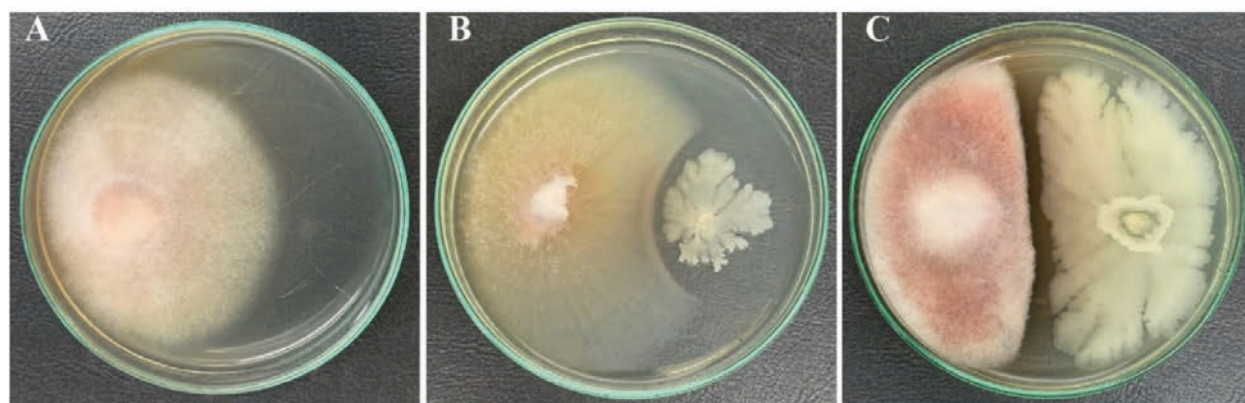


Figure 1: Representative of the antagonistic activity of isolates against the *F. solani* under laboratory conditions. A: Control (*F. solani*); B: isolate TuR9; C: isolate TuR6

3.2 *IN VITRO* ANTAGONISTIC ACTIVITY AGAINST *FUSARIUM SOLANI*

Antifungal assays revealed that TuR6 exhibited the most significant antagonistic activity, with an inhibition zone of 78.12 %. Isolates TuR3, TuR9, and TuR10 also showed moderate inhibition, ranging from 11.0 % to 12.4 % (Table 2, Figure 1). These results align with previous studies by Vinayarani & Prakash (2018), who demonstrated the efficacy of *B. cereus* in inhibiting *Pythium aphanidermatum* (Edson) Fitzp. and *Rhizoctonia solani*, J.G. Kühn pathogens responsible for turmeric rhizome rot and leaf blight.

The experimental results (Table 2) indicate that the bacterial strain TuR6 not only possesses direct plant growth-promoting traits (such as inorganic phosphate solubilization and phytohormone production) but also exhibits indirect growth-promoting effects through antifungal activity against plant pathogens, like siderophore production. Earlier studies have demonstrated that bacterial siderophores can effectively inhibit phytopathogenic fungi's growth, with the antifungal activity level closely associated with siderophore concentration (Calvente *et al.*, 2001). Through iron sequestration, siderophore production restricts iron availability to pathogens, thereby acting as a key biocontrol strategy employed by bacteria to suppress pathogen development (Afzal *et al.*, 2019; Sah *et al.*, 2021). Other reports, in which bacterial strains isolated from the rhizosphere and turmeric rhizome demonstrated their abilities in antagonizing *P. aphanidermatum*, the causal agent of turmeric rhizome rot. These strains significantly reduced the incidence of

turmeric rhizome rot under greenhouse and field conditions (Vinayarani & Prakash, 2018; Sharma *et al.*, 2024; Sagarika *et al.*, 2025). Moreover, the strain TuR6 was molecularly identified as *B. cereus* TuR6, exhibiting 98.68 % sequence similarity to *B. cereus* strains deposited in GenBank, based on BLAST analysis. Accordingly, in the present study, TuR6 has been identified as a promising PGP candidate with high potential for application in agriculture, especially for turmeric cultivation in Vietnam.

3.3 EFFECT OF *B. CEREUS* TUR6 ON PLANT GROWTH AND DISEASE SUPPRESSION UNDER GREENHOUSE CONDITIONS

The greenhouse evaluation of *B. cereus* TuR6 demonstrated significant efficacy in promoting turmeric plant growth and suppressing *F. solani*-induced rhizome rot. Plants treated with TuR6 exhibited markedly higher plant height (80.57 ± 1.43 cm), fresh rhizome mass (371 ± 1.38 g per plant), and significantly reduced disease incidence (17.7 ± 0.31 %) compared to untreated and pathogen-inoculated controls. These results underscore the biocontrol potential of strain TuR6 under controlled environmental conditions.

The biocontrol efficacy observed with TuR6 aligns well with earlier reports on plant growth-promoting rhizobacteria (PGPR) strains. For instance, *Pseudomonas fluorescens* TuR6, when applied as a combined rhizome dip and soil drench, achieved a 19 % disease incidence rate in turmeric under glasshouse conditions, considerably lower than the untreated control (70.83 %) (Prabhukarthikeyan *et al.*, 2018). In addition, TUR6-treated plants showed improved plant height, rhizome yield, and induction of defense-related enzymes such as phenylalanine ammonia lyase, peroxidase, polyphenol oxidase, superoxide dismutase, and β -1,3-glucanase, catalase, indicating enhanced ISR in host plants (Van Loon *et al.*, 2008; Prabhukarthikeyan *et al.*, 2018).

Similarly, *B. cereus* strain RBacDOB-S24, when tested in greenhouse trials, significantly suppressed both rhizome rot (by 16.4 %) and leaf blight (by 15.5 %) caused by *P. aphanidermatum* and *R. solani*, respectively (Prabhukarthikeyan *et al.*, 2018; Vinayarani & Prakash, 2018; Saryanah *et al.*, 2021). This strain also promoted plant height to 89.09 cm and rhizome yield to 392 g under pathogen challenge, reflecting a similar performance to TuR6 in our study.

Both TuR6 and the previously studied strains share several PGP traits, such as production of IAA, siderophores, and lytic enzymes, including cellulase and protease (Vinayarani & Prakash, 2018; Kavitha *et al.*, 2024). These traits likely contribute to their dual function in

Table 2: *In vitro* antagonistic activity of bacterial strains against plant pathogenic *F. solani* under laboratory conditions

Isolates	Growth inhibition zone of <i>F. solani</i> (%)
TuR1	0
TuR2	0
TuR3	11.57 ± 0.51^b
TuR4	0
TuR5	0
TuR6	78.12 ± 0.27^a
TuR7	0
TuR8	0
TuR9	12.41 ± 0.37^b
TuR10	11.0 ± 0.15^b

Values represent the mean of three replicates $\pm$ standard error (SE). Means followed by the same letter within a column are not significantly different at $p \leq 0.05$ according to Tukey's Honest Significant Difference (HSD) test.

Table 3: Effect of endophytic strain TuR6 on turmeric growth and disease suppression under *Fusarium solani* challenge in greenhouse conditions

Treatment	Fresh rhizome mass (g per plant)	Plant height (cm)	PDI
<i>B. cereus</i> TuR6	371 ± 1.38 ^a	80.57 ± 1.43 ^a	17.7 ± 0.31 ^b
Carbendazim (0.1 %) + Mancozeb (0.25 %)	283 ± 1.29 ^b	66.27 ± 0.58 ^b	19.1 ± 0.52 ^b
Uninoculated control	253 ± 1.26 ^c	53.01 ± 0.53 ^c	0.0
Pathogenic control	202 ± 1.37 ^d	40.76 ± 0.59 ^d	78.97 ± 0.43 ^a

Values represent the mean of three replicates ± standard error (SE). Means followed by the same letter within a column are not significantly different at $p \leq 0.05$ according to Tukey's Honest Significant Difference (HSD) test. PDI: Percent Disease Index.

promoting plant growth and inhibiting pathogen colonization. The capacity of these bacteria to enhance enzymatic defenses and compete for critical nutrients like iron via siderophore secretion plays a central role in their biocontrol activity (Vinayarani & Prakash, 2018; Kavitha et al., 2024).

Taken together, the performance of *B. cereus* TuR6 not only confirms the feasibility of employing endophytic bacteria for biological control of *F. solani* in turmeric but also supports its use as a multifunctional bioinoculant. Compared to chemical fungicides, which showed lesser growth promotion and similar levels of disease suppression, TuR6 presents a safer, eco-friendly alternative for sustainable turmeric cultivation in Vietnam and beyond.

3.4 EFFECT OF *B. CEREUS* TUR6 ON PLANT GROWTH AND DISEASE SUPPRESSION UNDER FIELD CONDITIONS

Under field conditions challenged with *F. solani*, turmeric plants treated with *B. cereus* TuR6 exhibited substantially improved growth and disease resistance compared to untreated and pathogen-inoculated controls (Figure 2).

In terms of disease suppression, TuR6 reduced the PDI by 73 % relative to the pathogenic control (Figure 2A). Notably, its level of disease control was comparable to that of the chemical fungicide treatment, despite being a biological agent. This underscores the biocontrol efficacy of TuR6, likely due to its antagonistic activity via production of antibiotics, siderophores, or hydrolytic enzymes - mechanisms well documented in *B. cereus* strains (Ongena & Jacques, 2008).

TuR6 treatment led to a 72 % increase in fresh rhizome mass and more than a two-fold increase in plant height compared to the pathogenic control (Figure 2B and 2C). Even compared to the chemical treatment (carbendazim + mancozeb), TuR6 improved rhizome yield by approximately 28 % and plant height by 23 %. In rela-

tion to the uninoculated control (plants not exposed to the pathogen), TuR6 still outperformed by 35 % in yield and 56 % in height, indicating its growth-promoting ability independent of disease pressure. This growth-promoting effect is consistent with previous reports on

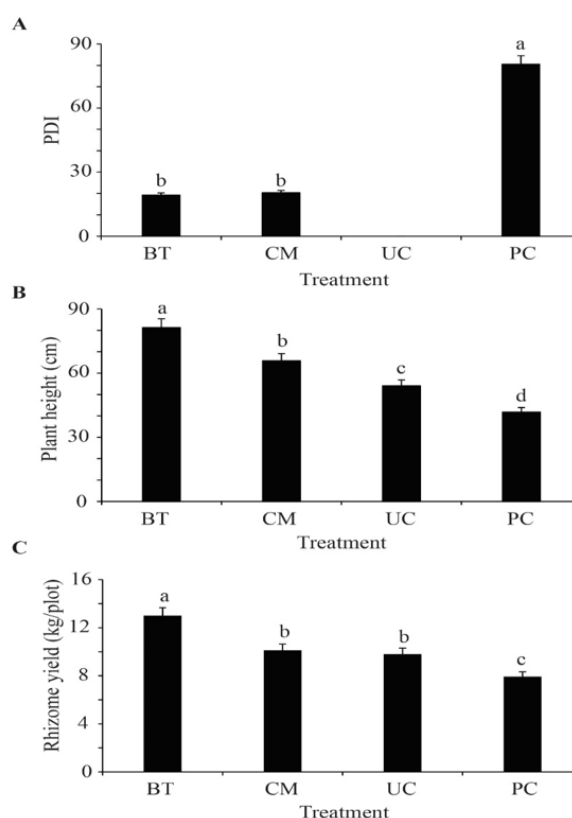


Figure 2: Effect of endophytic strain TuR6 on disease suppression (A), plant height (B), and rhizome yield (C) of turmeric plant under *F. solani* challenge in field conditions. Plotted values represent the mean of three replicates ± standard error (SE). Means followed by the same letter are not significantly different at $p \leq 0.05$ according to Tukey's Honest Significant Difference (HSD) test. PDI: Percent Disease Index, BT: *B. cereus* TuR6, CM: Carbendazim (0.1 %) + Mancozeb (0.25 %), UC: Uninoculated control, PC: Pathogenic control

PGPR, including strains of *Bacillus* spp., which enhance nutrient availability, stimulate phytohormone production, and improve plant vigor under both normal and stress conditions (Vinayarani & Prakash, 2018; Saryanah *et al.*, 2021).

The superior performance of TuR6 over chemical fungicides also highlights the potential for reducing chemical inputs in turmeric cultivation. Besides being environmentally sustainable, the use of endophytic PGPR like TuR6 aligns with integrated disease management approaches, enhancing the resilience of turmeric cropping systems.

Interestingly, while the uninoculated control showed 0 % disease incidence, it performed significantly lower in terms of both yield and plant height (Figure 2). This suggests that TuR6 not only plays a protective role under pathogen pressure but also acts as a biofertilizer, contributing to overall plant health and productivity. Similar dual effects of *Bacillus* strains have been observed in turmeric and other crops (Do *et al.*, 2025).

4 CONCLUSIONS

This study identified *B. cereus* TuR6 as a promising endophytic bacterium with dual capabilities for promoting plant growth and controlling biocontrol. TuR6 demonstrated high IAA production, phosphate solubilization, siderophore synthesis, and enzymatic activity, contributing to enhanced plant vigor and pathogen resistance. It showed *in vitro* antagonism against *F. solani* and significantly improved plant height, rhizome yield, and disease suppression under both greenhouse and field conditions. These results support TuR6 as a sustainable alternative to chemical inputs in turmeric cultivation. Further multi-location and long-term trials are recommended to validate its consistency and formulation potential across agro-ecological zones.

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Data Availability

The datasets generated and analyzed during the current study are available in a trusted public repository and include replicate-level measurements underlying all tables and figures of the manuscript. The data are openly

accessible at Zenodo under accession number 10.5281/zenodo.18251744.

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