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Effect of endophytic bacterium *Curtobacterium citreum* PLPL and chitosan on plant growth enhancement and management of potato black scurf and stem canker

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ABSTRACT

Rhizoctonia stem canker and black scurf of potato are the most prevalent and widespread soil-borne fungal diseases that cause notable reduction in the yield of potato globally. The purpose of the current study was to assess the effect of both *Curtobacterium citreum* and chitosan, either alone or in combination, on the prevention of potato Rhizoctonia diseases and its growth-promoting characteristics. The antagonistic capability of the endophytic bacteria *C. citreum* PLPL and chitosan against *R. solani* was evaluated and got potential responses. Following that, compatibility tests between *C. citreum* and chitosan were conducted, revealing compatibility between the natural agents. Prior to sowing, seed tubers were treated with *C. citreum* (10⁸ colony forming units per milliliter) and chitosan at 1.0 % concentration. In comparison to pathogen-inoculated plots, all individual and combined treatments increased growth and yield while reducing disease incidence and severity. The combination of *C. citreum* and chitosan resulted in the significant reduction in disease incidence (79.95% for stem canker and 65.53% for black scurf) and percent disease index (85.14% for stem canker and 66.00% for black scurf) over the pathogen-inoculated plot. The combination of *C. citreum* with chitosan resulted in a maximum yield of 43.15 t/ha, with all growth parameters significantly increased. In addition to increasing potato yield, treating tubers with a combination of *C. citreum* and chitosan may be a sustainable and environment friendly method of managing stem canker and black scurf diseases.

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Introduction

Potato (*Solanum tuberosum* L.) is one of the most important vegetable crops of the world belonging to the family solanaceae. In 2021, 376 million tons of potatoes were grown globally on 18,132,694 ha of land (FAO, 2023). In Bangladesh, 464011 ha of land are covered by potato cultivation, and 10144835 MT of potatoes were produced in 2021-2022 (BBS, 2023). The production of potatoes is restricted by a number of factors, the most significant of which are diseases caused by plant pathogens. Rhizoctonia diseases include damping off, stem canker, black scurf, and root rot, which are caused by soil-borne fungal pathogen *Rhizoctonia solani* (telomorph: *Thanatephorus cucumeris*). Black scurf and stem canker are two diseases that drastically lower potato tuber production worldwide (Brewer and Larkin, 2005). Stem canker results in quantitative losses and a decrease tuber quantity and size. In contrast, black scurf is associated with the development of malformed tubers and mycelia clustering (Das *et al.*, 2014). To mitigate the quantitative and qualitative losses due to stem canker and black scurf disease, control measures should be employed. Control of *R. solani* is extremely difficult due to its diverse host range. It also produces resting spores to overwinter in soil and plant parts (Balkali and Martin, 2006). Around the world, farmers mostly use synthetic fungicides to manage soil-borne diseases in agricultural systems. However, the unregulated application of synthetic fungicides causes ecological disruption, risks

to human health, adverse effects on aquatic environments, and a decline in soil-beneficial microorganisms (Panth *et al.*, 2020). So, eco-friendly management practices should be employed instead of synthetic fungicides to combat plant diseases (Fenta and Mekonnen, 2024). Beneficial bacteria also have a great chance of protecting against certain well-known plant diseases caused by bacteria and fungi. Recently, bacterial endophytes have exhibited favorable outcomes on host plants, including enhancing growth and control of pathogens (Tjamos *et al.*, 2004). In order to combat plant pathogenic fungi like *R. solani*, *Pythium* sp., *Alternaria alternata*, *Fusarium* sp., *Sclerotinia sclerotiorum*, endophytic bacteria can be utilized (Cao *et al.*, 2005). Like other endophytes, *Curtobacterium* can strengthen plant defenses and reduce disease symptoms (Lacava *et al.*, 2007). A bio pesticide like chitosan, which provides a unique, sustainable technique of disease management in fruits and vegetables (Maqbool *et al.*, 2010), and can be used as an alternative to chemical pesticides.

Chitosan is an antimicrobial substance with a substantial chance to combat pathogenic plant diseases through the interplay of its antimicrobial and eliciting properties (Xing *et al.*, 2015). It is non-toxic to living things in addition to being biodegradable and biocompatible. It is created by applying an alkaline substance (NaOH) to the shells of shrimp and other crustaceans that are made of chitin. Its ingredients are also capable of boosting plant growth and output while acting

as antibiotics against a broad range of microbes (Román-Doval *et al.*, 2003). Chitosan has been widely used in the field to induce resistance against late and early blight diseases of potatoes and root rot diseases of tomato plants (Abd-El-Kareem *et al.*, 2001, Abd-El-Kareem *et al.*, 2006). Numerous authors also encouraged using bioagents in combination to achieve maximum benefit from them. Pastucha (2005) reported that the number of bacteria in the soybean rhizosphere soil increases after using chitosan. The addition of chitosan in soil enhances the efficacy and cell number of antagonistic *Lysobacter enzymogenes* against *Pythium aphanidermatum* (Postma *et al.*, 2009). Mishra *et al.* (2014) observed a substantially higher number of bacterial cells in tomato roots after the application of *Pseudomonas* sp., in combination with chitosan. The natural biopolymer, chitosan, promotes the activity of beneficial microbes like *Bacillus*, *Pseudomonas*, actinomycetes, mycorrhiza, and rhizobacteria in the soil (Bell *et al.*, 1998). As a result, this changes the balance of microbial community in rhizosphere which makes the plant pathogens more vulnerable. Additionally, the application of chitosan with antagonistic bacteria increases growth and yield of strawberry (Mukta *et al.*, 2017). The development of a suitable pathogen control strategy, such as the combination of biocontrol agents, is currently required because the use of biological control agents alone is less successful than their combined use.

The aforementioned parameters were considered in the design of the study, which aimed to examine the effects of chitosan and antagonistic bacteria both individually and in

combination on the suppression of *R. solani* causing stem canker and black scurf diseases, as well as on the enhancement of potato production.

Materials and Methods

R. solani isolation, virulent isolate selection and inoculum preparation

Potato tubers exhibiting characteristic symptoms of black scurf were utilized for pathogen isolation. Infected potato samples were gathered from the fields of farmers in the Gazipur district, including the research field of Bangabandhu Sheikh Mujibur Rahman Agricultural University (BSMRAU). Diseased portions of the tuber were sliced into 1-2 cm pieces, surface sterilized for 30 seconds in 70% ethanol, and incubated in water agar. The mycelial tips of the *R. solani* isolates were then transferred to PDA medium by observing its morphological characters like tan to light brown septate hyphae, branching at 90-degree angles, and the presence of constrictions at the base of hyphal branching. Subcultures of *R. solani* isolates in the PDA plate were carried out to get pure culture of the fungus. After purification, *R. solani* isolates were kept in PDA slant for further use (Wang *et al.*, 2015). In a pot culture trial, the virulence of the isolates was assessed by monitoring potato mortality before and after emergence employing a soil infestation technique (Datar *et al.*, 2011). Two kilograms of sterilized soil were added to each earthen pot and two grams of wheat bran inoculum were spread out around the tuber. For each

isolate, three replications were used to record disease development in order to determine the pathogen's activity in causing pre- and post-emergence seedling death. Re-isolation of pathogens from infected tubers and seedlings provided proof of the causative agent. At last, the most virulent *R. solani* isolate was chosen for further study. Wheat bran inoculum of the *R. solani* isolate was prepared following the methods of Zhang *et al.* (2014), where 100 g of wheat bran and 200 mL of distilled water were taken in a 500 mL conical flask and autoclaved for 40 min at 121°C. Ten mycelial discs of *R. solani* having 0.8 cm diameter grown in Potato Sucrose Agar (PSA) were added in the wheat bran containing conical flask for 4 days at 25°C under dark light condition. Thirty days later, the wheat bran containing hyphae and sclerotia was collected, dried, and ground into powder.

Collection, culture and in vitro assessment of *C. citreum* against *R. solani*

Endophytic antagonistic bacterium, *C. citreum* PLPL (accession number MT084037) was collected from Plant Pathology Department of BSMRAU. Then the bacterium was cultured on Yeast Peptone Dextrose Agar (YPDA) medium and incubated for 3 days. A dual culture assay was conducted to assess the effect of *C. citreum* against *R. solani*. The PDA plate had a 5 mm *R. solani* disk at one side of the petri dish, and antagonistic bacterium *C. citreum* was streaked at the middle. The dual culture assay was repeated three times to confirm the results regarding the mycelial inhibition of *R. solani* (Elkahoui *et al.*, 2012).

Collection and in vitro assessment for the selection of effective dose of chitosan against *R. solani*

Chitosan was procured from the Bangladesh Atomic Energy Commission (BAEC). The effect of chitosan on mycelial growth was determined by Chronological preliminary analyses, performed on PDA plates supplemented with 0.125, 0.25, 0.5, 1.0 percent concentrations of chitosan using three replications against *R. solani* isolates. The 5 mm mycelial discs of *R. solani* were placed in the center of the chitosan-supplemented PDA plates and incubated at 25 °C for 5 days (Akter *et al.* 2018). Finally, a calculation was performed to determine the percentage of radial growth inhibition following the formula:

% inhibition of radial growth of *R.*

$$\text{solani} = \frac{X - Y}{X} \times 100$$

Where, X = Mycelial growth of *R. solani* in absence of chitosan (control)

Y = Mycelial growth of *R. solani* in presence of chitosan

Preparation of cell suspension of *C. citreum*

Cells were extracted from nutrient broth cultures that had been maintained at 28± 1°C for 48 hours, and then centrifugation performed at 6000 rpm for 15 minutes to prepare cell suspension of *C. citreum*. After re-suspending the inoculum in sterile distilled water, the concentration was measured with a spectrophotometer and maintained at 10⁸ CFU/mL (Kumar *et al.* 2015).

Characterization of *C. citreum*

Some morphological observations (bacterial colony color, shape, size and texture) and biochemical tests such as Gram reaction test (Archana *et al.*, 2013), KOH solubility test (Li *et al.*, 2021) catalase assay (Xu *et al.*, 2020), glucose fermentation test (Hugh and Leifson, 1953) and production of indole acetic acid (Hemraj *et al.* 2013) were performed following standard protocols. These morphological and biochemical test were carried out to identify the specific character of the bacteria. Among the tests, indole acetic acid production test was conducted to determine the plant growth promoting character of the bacteria.

Compatibility test of *C. citreum* and chitosan

Compatibility test of *C. citreum* and chitosan was carried out by streaking of *C. citreum* on chitosan (1%) amended PDA plate. It was incubated for 5 days and the response of the bacterium was monitored carefully.

Collection of Potato tubers for field experiment

“BARI ALU-7 (Diamant)” was chosen for this experiment. Tubers of the same size and without any damage or disease symptoms were selected for seed treatment. The entire tuber was utilized for planting in the field. Prior to treatment, the tubers were briefly cleaned with 1% sodium hypochlorite for three minutes, rinsed multiple times with sterile water to

remove residual disinfectant solution, and allowed to air dry.

Field experiment with *C. citreum* and chitosan

The total experimental area was separated into three blocks, which corresponded to three replications. The plot size was 3.0 m by 1.2 m. Blocks and plots were separated by 1.0 and 0.50 meters, respectively. When treated tubers were sown, the plants were put 25 cm apart, and rows were placed 60 cm apart. To get an advantage over the weeds, weeding was done four weeks following planting. The proper amount of water was given when irrigation was needed. At 20-day intervals, three earthing ups were carried out. Before sowing in the field, healthy tubers were sprayed 3 times at 4hrs interval with *C. citreum* (10^8 CFU/mL) and chitosan (1.0%) for 24hrs. A few drops of the emulsifier Tween 20 and sticker were added. Foliar spray was done 30 days after planting. Combined treatments were done as per the requirements.

The treatment combinations were as follows: T₁: Soil without any treatment (control 1), T₂: Soil inoculation with *R. solani* (control 2), T₃: T₂ + seed treatment with 1.0 % chitosan, T₄: T₂ + foliar spray with 0.5 % chitosan, T₅: T₂ + seed treatment with *C. citreum* (10^8 CFU/mL), T₆: T₂ + seed treatment with 1.0 % chitosan + foliar spray with 0.5 % chitosan, T₇: T₂ + seed treatment with *C. citreum* (10^8 CFU/mL) + seed treatment with 1.0 % chitosan.

Data recording

Data on seedling mortality, shoot length (cm), shoot fresh and dry weight (g), number of branches per plant, disease incidence and disease severity of stem and tuber, tuber diameter (cm), weight of tuber (g), and yield (t/ha) were taken at various phases of potato plant development. The percentages of disease incidence and disease severity were calculated following the formula stated below:

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

$$\text{Percent Disease Index (PDI)} = \frac{\sum \text{of rating of stems/tubers observed}}{\text{Number of stems/tubers observed} \times \text{Highest score of the scale used}} \times 100$$

A five-point rating scale was employed to assess the severity of stem canker and black scurf disease, with 0 denoting no symptoms, 1 denoting 1-25%, 2 denoting 26-50%, 3 denoting 51-75%, and 4 denoting 76-100% of the potato stem covered in lesions and tuber with sclerotia (Grisham and Anderson, 1983).

Statistical Data analysis

A Complete Randomized Design (CRD) was employed for the in vitro investigations, while a Randomized Complete Block Design (RCBD) with three replications was utilized for the field trial. Data on several disease attributes and yield parameters were statistically assessed using the statistical software Statistix 10. At

the 5% level of significance, the means were compared after the LSD (Least Significant Difference) test.

Results

Exploring pathogenicity test to select virulent *R. solani* isolate

A total of ten *R. solani* isolates were isolated from different fields of BSMRAU, from which Rh-1, Rh-2, and Rh-3 isolates were selected randomly for the pathogenicity test to find out the most virulent isolate. Although all of the test pathogen isolates were virulent, their respective rates of overall potato seedling mortality varied, ranging from 35-88%. With the highest total seedling mortality of 88 %, the *R. solani* isolate Rh-2 appeared to be the most virulent, followed by isolate Rh-3, which had a death rate of 55 % (Fig. 1 and Table 1). Notably, isolate Rh-1 had the lowest total seedling mortality rate of 35%. In the untreated control pot, there was no evidence of seedling mortality either before or after emergence. Thus, the isolate Rh-2 was selected for further study. Numerous researchers have also confirmed that *R. solani* causes pre-emergence and post-emergence mortality in potatoes (Rauf *et al.*, 2007). Yang *et al.* (2017) found that the pathogenicity of the *R. solani* isolates was indicated by black scurf and stem cankers on potatoes. Our findings are consistent with their findings.

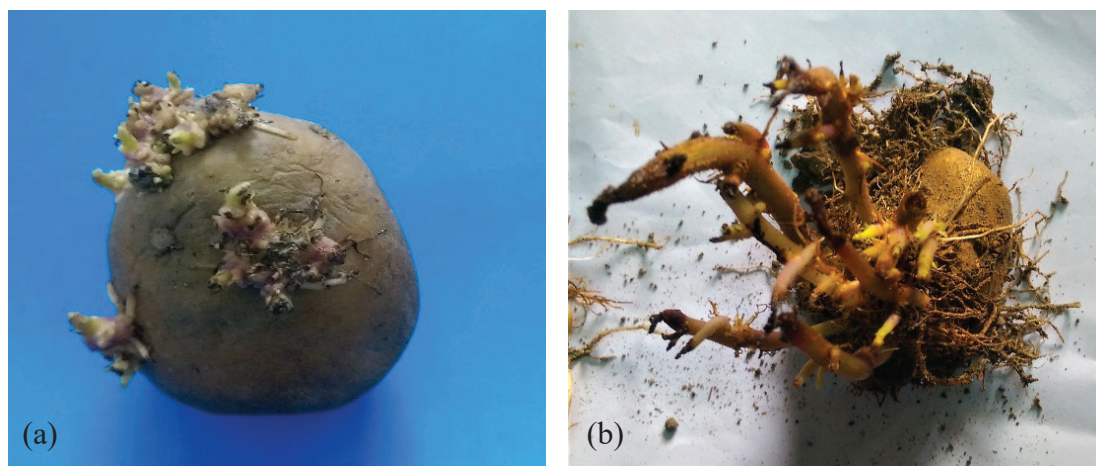


Fig. 1. Seedling mortality of potato plant. a. Pre-emergence seedling mortality, b. Post-emergence seedling mortality

Table 1. Pathogenicity test of *Rhizoctonia solani* against potato plants

<i>R. solani</i> isolates	Seedling mortality (%)		
	Pre-emergence	Post-emergence	Total
Rh-1	12.00	23.00	35.00 c
Rh-2	28.00	60.00	88.00 a
Rh-3	20.00	35.00	55.00 b
Control	0.00	0.00	0.00 d
SE	4.90	7.22	7.88

*Means in columns with different letters show significant differences ($p < 0.05$) as assessed by LSD.

In vitro* assessment of *C. citreum* against *R. solani

The collected bacteria *C. citreum* PLPL was rejuvenated on YPDA medium and a dual culture assay was performed against *R. solani* Rh-2 (Fig. 2). The results revealed that *C. citreum* inhibited the mycelial development of *R. solani* by 90.25%. According to Pal and Gardener (2006), antagonistic bacteria

are also said to be effective in controlling a number of harmful fungi, including *Rhizoctonia*, *Pythium*, *Fusarium*, *Aspergillus*, *Phytophthora*, *Pyricularia*, and *Alternaria*. These bacteria produce some secondary metabolites that inhibit the fungal mycelia. Our findings supported earlier research showing that endophytic bacteria, *C. pusillum* effectively inhibited *R. solani*'s mycelial proliferation (Hassanin *et al.*, 2007).



Fig. 2. In vitro antagonistic activity of *Curtobacterium citreum* against *Rhizoctonia solani*

Exploring in vitro antagonistic action to select an effective dose of chitosan against *R. solani*

The mycelial growth of *R. solani* was reduced by chitosan in a dose-dependent way. Chitosan at 0.125, 0.25, 0.5 and 1.0 % greatly inhibited

the mycelial development of the test pathogen compared to the non-treated control. About 100 % inhibition of the mycelial development of *R. solani* was observed by 1.0 % compared to the control PDA plate. The PDA plate amendment with 0.5 % chitosan exhibited the second-highest reduction (88.92 %) of the growth of *R. solani* mycelium (Fig. 3). Thus, 0.5 % and 1.0 % dosages of chitosan were selected for the field trial based on the in vitro assessment. Previously some researchers in their publication mentioned that chitosan effectively suppressed the mycelial growth of fungal pathogens. Akter *et al.* (2018) and Chatterjee *et al.* (2022) reported that 1.0 % concentration of chitosan completely inhibited the mycelial growth of *Colletotrichum* and *Fusarium*. Our experiment also revealed similar results. The reason behind this may be the antifungal mechanism of chitosan, where

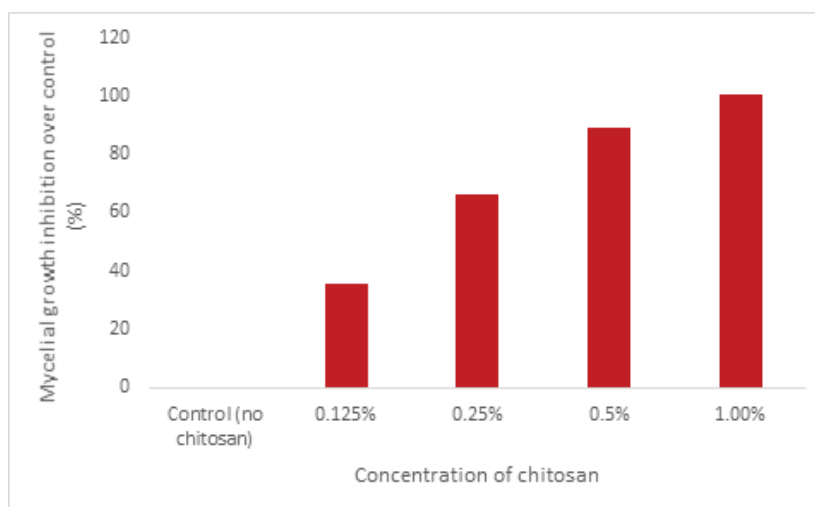


Fig. 3. Impact of different dosage of chitosan on mycelial growth of *Rhizoctonia solani*. Bars on the column indicate (\pm) Standard Error of the Mean. Means in columns with different letters show significant differences ($p < 0.05$) as assessed by LSD

chitosan molecules directly impede fungal development through cell wall morphogenesis. Again, chitosan oligomers penetrate hyphae and disrupt the function of the enzymes that cause the fungus to spread (Eweis *et al.*, 2006).

Characterization of *C. citreum*

Morphological observation revealed the colony characters of the bacteria. It was observed that the selected antagonistic bacteria formed round, small, smooth textured and yellow color colonies. The bacterium was rod in shape. Biochemical assay revealed the tested antagonistic bacterium as Gram positive. It was found that the bacterium was aerobic in nature, positive for catalase, produced indole acetic acid but not fermented glucose (Table 2). These biochemical tests provide results to identify the nature of the bacteria along with their growth promoting traits.

Compatibility assessment of *C. citreum* and Chitosan

Before field application, compatibility of *C. citreum* and chitosan was confirmed. In the *in vitro* experiments *C. citreum* and chitosan showed favorable responses (Fig. 4). When

cultured in the PDA medium *C. citreum* and chitosan guarantee their compatibility.

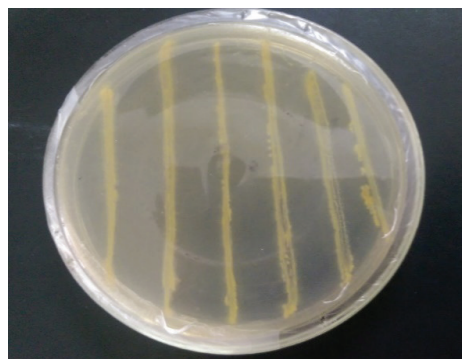


Fig. 4. Compatibility test of *Curtobacterium citreum* and chitosan

Evaluating *C. citreum* and chitosan's ability to prevent potato stem canker and black scurf disease

The disease incidence (DI) and severity (PDI) of potato stem canker and black scurf decreased in all treatments compared to the pathogen-inoculated plot. In case of Rhizoctonia stem canker, significantly the highest DI (35.33%) and PDI (17.94%) were recorded in pathogen inoculated soil while the lowest DI (7.08%) and PDI (2.67%) were recorded in T₇ treatment where *C. citreum* and chitosan treated seeds were used. Treatment T₇ revealed

Table 2. Morphological and biochemical characterization of *Curtobacterium citreum*

Antagonistic bacterial isolate	Colony and cell morphology	Gram reaction Test	KOH test	Catalase test	Fermentation of glucose test	IAA production test
<i>C. citreum</i> PLPL	Round, small, smooth, yellow colony, rod shaped	+	-	+	-	+

‘+’= Positive, ‘-’= Negative

the highest decrease in DI (79.95%) and PDI (85.14%) compared to treatment T_2 . In case of black scurf of potato, the considerably highest DI (36.55%) and PDI (25.57%) were found in *R. solani* inoculated soil while the lowest DI (12.60%) and PDI (8.69%) were obtained in treatment T_7 . The highest decrease in DI (65.53%) and PDI (66.00%) was observed in treatment T_7 compared to treatment T_2 . Potato stem canker and black scurf disease incidence and severity were significantly reduced in treatment T_7 where seed treatment was done with *C. citreum* and chitosan (Fig. 5). Our findings indicate that *C. citreum* and chitosan can successfully manage potato stem canker and black scurf disease. *C. citreum* and chitosan individually and in combination can reduce DI and PDI of Rhizoctonia diseases of potato. Bacterial endophytes use Plant growth-promoting Rhizobacteria like mechanisms where they synthesis secondary metabolites, volatile chemicals, and antibiotics to combat infections' detrimental effects (Lodewyckx *et al.*, 2002). According to earlier reports, tomato plant southern blight disease can be managed by endophytic bacteria *Burkholderia* sp. and *Bacillus* sp. (Hari *et al.*, 2023). Furthermore, *C. citreum* has the ability to trigger the plant's defense mechanisms, including the synthesis of phytoalexins. Additionally, *Curtobacterium* can colonize plant surfaces, forming a physical barrier that prevents plant diseases, such as citrus-variegated chlorosis. (Lacava *et al.*, 2007). Chitosan is commonly employed in controlling plant diseases due to its strong elicitor properties, rather than its direct antimicrobial effects (El-Mohamedy *et al.*, 2013).

According to Akter *et al.* (2018) and Jannat *et al.* (2018) chitosan effectively reduces DI and PDI of anthracnose of chilli and Phomopsis blight and fruit rot of eggplant. According to a prior publication, Akter *et al.* (2018) stated that anthracnose of chilli can be effectively controlled by the seed treatment and foliar application of chitosan. Our results also revealed similar outcomes. Algam *et al.* (2010) reported that chitosan and *Paenibacillus* combindly decreased the prevalence of tomato Ralstonia wilt. There is another report claiming that chitosan in combination with endophytic bacteria *Bacillus pumilus* induce resistance against Fusarium wilt of tomato (Benhamou *et al.*, 1998). Our results are consistent with these reports where combined application reduced the disease incidence and severity than the individual treatments.

Evaluating the ability of C. citreum and chitosan to increase potato plant growth and yield

The present study results pointed out that, growth promoting aspects and yield attributing factors were significantly enhanced in all the treatments by the combined use of *C. citreum* and chitosan except T_2 . The longest shoot length (54.07 cm), plant fresh and dry weight (111.11 g and 28.38 g, respectively), maximum number of branches per plant (5.85), greatest tuber weight per plot (15.53 kg), and the highest tuber diameter (5.12 cm) was obtained with T_7 treatment, which involved treating seeds with *C. citreum* and chitosan. Significantly, pathogen inoculated

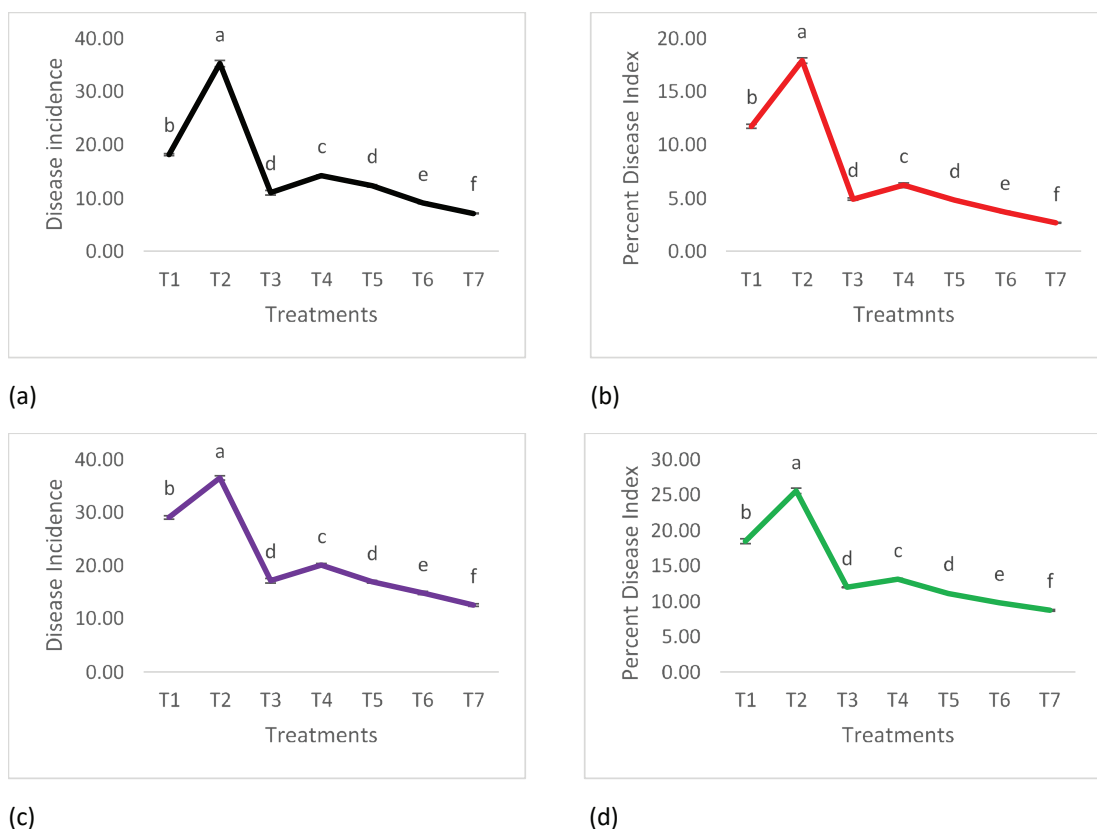


Fig. 5. Effect of *Curtobacterium citreum* and chitosan on DI and PDI of stem canker and black scurf disease of potato plants. a-b. DI and PDI of stem canker, c-d. DI and PDI of black scurf. Bars on the trend lines indicate (\pm) Standard Error of the Mean. Means in columns with different letters show significant differences ($p < 0.05$) as assessed by LSD. The treatment combinations are T₁: Soil without any treatment (control 1), T₂: Soil inoculation with *R. solani* (control 2), T₃: T₂ + seed treatment with 1.0 % chitosan, T₄: T₂ + foliar spray with 0.5 % chitosan, T₅: T₂ + seed treatment with *C. citreum* (10^8 CFU/mL), T₆: T₂ + seed treatment with 1.0 % chitosan + foliar spray with 0.5 % chitosan, T₇: T₂ + seed treatment with 1.0 % chitosan + seed treatment with *C. citreum* (10^8 CFU/mL)

soil yielded the shortest shoot length (24.33 cm), plant fresh and dry weight (43.63 g and 7.57 g respectively), minimum number of branches per plant (2.39), lowest tuber weight per plot (8.47 kg), and the shortest tuber diameter (3.70 cm) (Fig. 6). Notably, treatment T₂ had the lowest yield (23.53 t/ha), whereas

treatment T₇ produced the highest yield (43.15 t/ha) (Fig. 8). Our results also pointed out that endophytic bacteria *C. citreum* and chitosan improves the growth and yield of potato plants over the pathogen inoculated plot. Our findings are in accordance with the report of Bourles *et al.* (2019), they claimed that

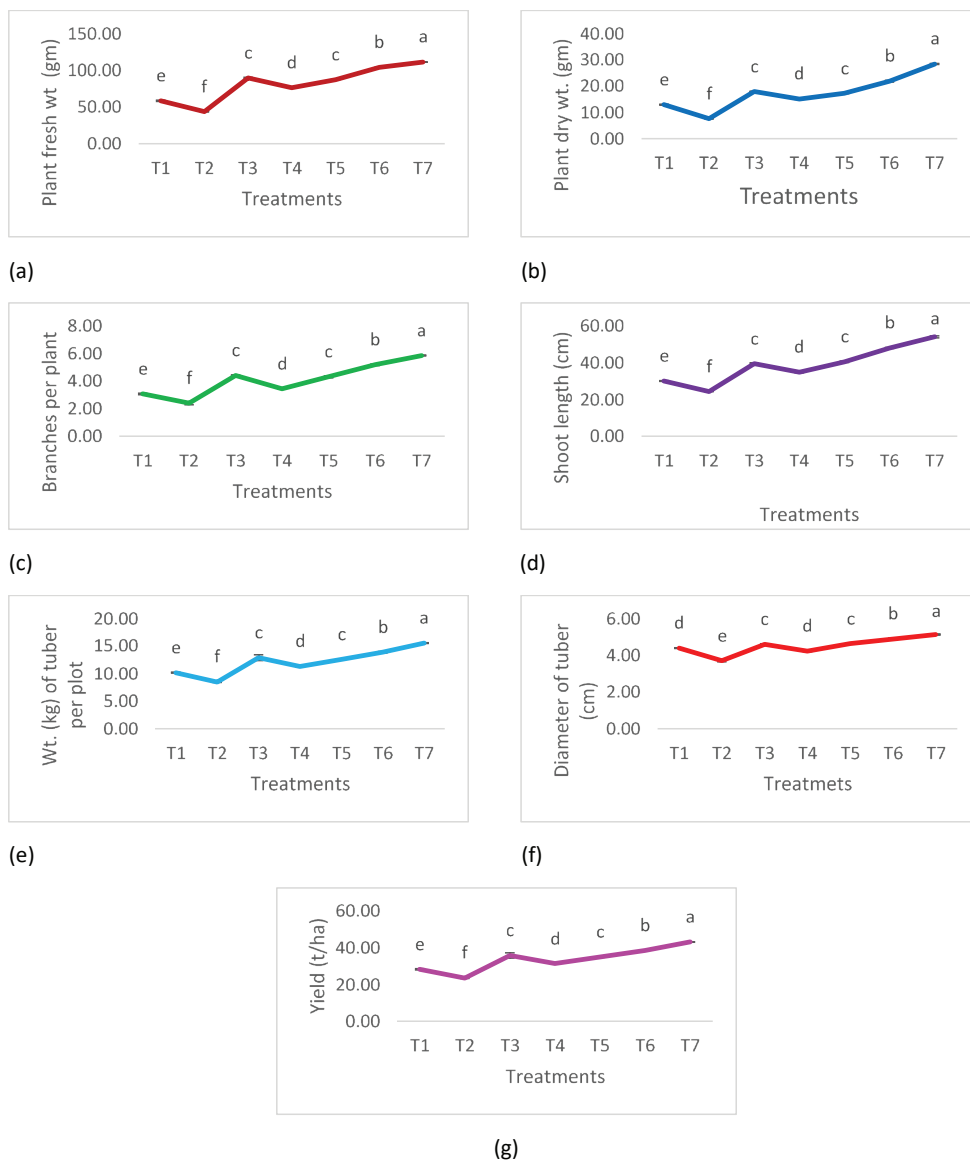


Fig. 6. Effect of *Curtobacterium citreum* and chitosan on growth parameters and yield contributing characters of potato plants. a, b, c and d- growth parameters, e, f, g- yield and yield contributing characters. Bars on the trend line indicate (\pm) Standard Error of the Mean. Means in columns with different letters show significant differences ($p < 0.05$) as assessed by LSD. The treatment combinations are T₁: Soil without any treatment (control 1), T₂: Soil inoculation with *R. solani* (control 2), T₃: T₂ + seed treatment with 1.0 % chitosan, T₄: T₂ + foliar spray with 0.5 % chitosan, T₅: T₂ + seed treatment with *C. citreum* (10^8 CFU/mL), T₆: T₂ + seed treatment with 1.0 % chitosan + foliar spray with 0.5 % chitosan, T₇: T₂ + seed treatment with 1.0 % chitosan + seed treatment with *C. citreum* (10^8 CFU/mL).

rhizosphere isolated bacterium *C. citreum* also promotes the growth of plants. Previously it is reported that bacterial endophytes benefit host plants by promoting growth and controlling diseases (Carrie *et al.*, 2023). Bacterial endophyte *Burkholderia* sp. and *Bacillus* sp. can increase growth and yield of tomato plants by producing indole acetic acid (Hari *et al.*, 2023). Our results are in consistent with several reports where chitosan enhances growth and yield qualities with increasing concentration in various crops (Mondal *et al.*, 2013). Akter *et al.* (2018) and Jannat *et al.* (2018) found that chitosan boosts the development and productivity of chillies and eggplant. There are several report claiming that chitosan can promote growth and yield of brinjal and tomato (Chaterjee *et al.*, 2021 and Nitu *et al.*, 2016). Foliar application of chitosan along with seed treatment also enhanced the plant growth and increased yield of chilli (Akter *et al.*, 2018). Mondal *et al.* (2013) pointed out in their publication that foliar application of chitosan increased the growth parameters and yield attributes of mung bean. The growth characteristics and biomass production of maize plants are enhanced by the combination of chitosan and rhizobacteria (Agbodjato *et al.*, 2016). Chitosan and *Paenibacillus* together promotes the growth of tomato plants (Algam *et al.*, 2010). Our results are in accordance with the report of Mukta *et al.* (2017). They claimed that the application of chitosan with antagonistic bacteria increases growth and yield of strawberry.

Conclusions

The present investigation indicated that *Curtobacterium citreum* and chitosan individually and in combination were effective in suppressing stem canker and black scurf diseases along with increasing potato production. Combined application of *C. citreum* (10^8 CFU/mL) and chitosan (1%) as seed treatment was more effective in terms of disease control, growth promotion and yield of potato among all the treatments. So ecofriendly management of Rhizoctonia diseases of potato can be obtained by the combined application of *C. citreum* and chitosan.

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Author's contribution

Research implementation, data collection, analysis, drafting and editing the manuscript (FT), Design and revision of the manuscript (AAK), Design and revision of the manuscript (GKMMR), Conception, Research design, drafting and reviewing (RJ).

Conflict of Interest

The authors state that they have no conflicts of interest.

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