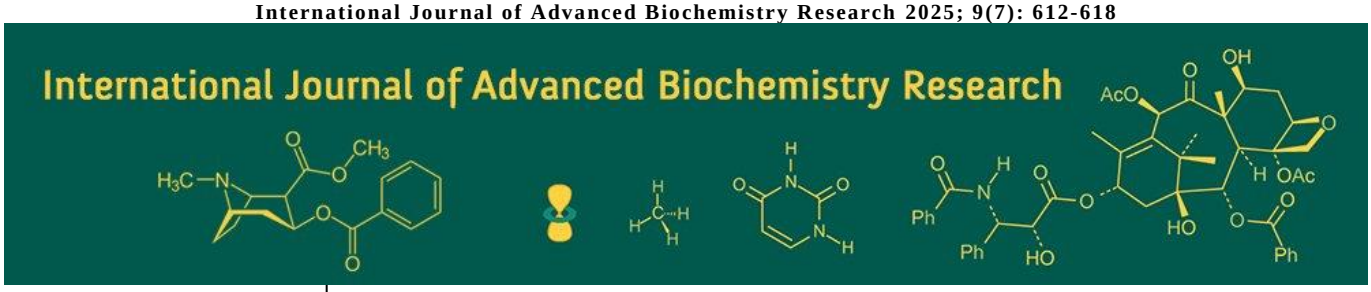

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Integrated application of *Trichoderma harzianum* and *Pseudomonas fluorescens* for the biological management of Botrytis gray mould in chickpea (*Cicer arietinum* L.)

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Abstract
Botrytis gray mould (BGM), caused by *Botrytis cinerea* Pers. ex Fr., is a major yield-limiting disease in chickpea (*Cicer arietinum* L.) globally. In recent years, biological control has gained prominence as a sustainable and environmentally friendly approach to disease management. This study evaluated the efficacy of two biological control agents, *Trichoderma harzianum* and *Pseudomonas fluorescens*, against BGM through laboratory and field trials. *in vitro* dual culture assays demonstrated significant antagonistic activity of selected strains against *B. cinerea*. Field experiments were conducted at the Crop Research Centre (CRC), G.B. Pant University of Agriculture and Technology, Pantnagar, during the rabi seasons of 2007-08 and 2008-09, using the highly susceptible cultivar H-208 in a Randomized Block Design (RBD). Treatments included foliar applications of *T. harzianum*, *P. fluorescens*, and a combination of both agents at 15-day intervals. All biocontrol treatments significantly reduced disease severity as well as increase the grain yield and 1000-seed weight compared to the untreated control. The combined application of *T. harzianum* and *P. fluorescens* was most effective, indicating synergistic effects in suppressing gray mould and promoting plant health. These findings align with recent advancements in biological control research emphasizing integrated microbial consortia for enhanced pathogen suppression (Kumar *et al.*, 2023; Zhang *et al.*, 2024).

Keywords: Biological control, *Botrytis cinerea*, *Trichoderma harzianum*, *Pseudomonas fluorescence*, chickpea

1. Introduction
Chickpea (*Cicer arietinum* L.) is a major legume crop cultivated globally for its high nutritional value and contribution to sustainable agriculture through nitrogen fixation. However, its productivity is severely hampered by various biotic stresses, among which Botrytis gray mould (BGM) caused by the necrotrophic fungus *Botrytis cinerea* Pers. ex Fr. is one of the most destructive diseases, particularly in humid and cool environments during the flowering and pod stages (Pande *et al.*, 2010; Khan *et al.*, 2011) [35, 23]. Botrytis can cause 70-100% yield loss in Northern India, under favourable conditions. In the absence of resistant cultivar, growers rely largely on fungicides to combat the disease but development of resistance in *B. cinerea* to fungicides is very common and widespread. The pathogen attacks flowers, leaves and mature fruits in the pre and post-harvest stages (Agrios, 1997) [2]. The effective method for control of gray mould has been chemical sprays with benzimidazole, dicarboximide and other fungicides in most greenhouses. However, *B. cinerea* has developed resistance to these fungicides by their repeated applications (Choi *et al.*, 1995; Grindle *et al.*, 1981) [8, 17]. In addition, public concern about fungicide residues in edible products and environment has accelerated the search for alternative disease control strategies. Biological control using antagonistic microorganisms is an alternative control method to the fungicide use and provides an ecologically based approach to integrated pest management in sustainable agriculture in crop production systems (Cook and Granados, 1991; Singh *et al.*, 1999; Sutton and Peng, 1993) [9, 40, 41]. Antagonistic microorganisms such as *Trichoderma*, *Bacillus*, and *Pseudomonas* species have been often evaluated for the control of *B. cinerea* and developed as commercial products (Roca *et al.*, 2021; Harman, 2000; Nelson and Powelson, 1988; Paulitz and Belanger, 2001; Redmond *et al.*, 1987) [38, 19,

[41, 36, 37]. A *Trichoderma* based bio-fungicide, TRICHODEX (Makhteshim Chemical Works Ltd., Beer Sheva, Israel) is commercially available for the control of *B. cinerea* (Elad, 2000a; 2000b) [14, 15]. Multiple mechanisms of action including antibiosis, competition, mycoparasitism *via* production of chitinases and glucanases, solubilization of inorganic plant nutrients and inactivation of pathogen's enzymes involved in the infection process have been suggested for the bio-control of *Trichoderma* species against *B. cinerea* (Altomare *et al.*, 1999; Elad and Kapat, 1999; Hjeljord *et al.*, 2001; Lorito *et al.*, 1993; Chohan *et al.*, 2024) [3, 13, 21, 29, 7]. The mechanism of induced resistance for defense responses by *T. harzianum* has also been provided (De Meyer *et al.*, 1998; Harman, 2000; Yedidia *et al.*, 1999) [10, 19, 44]. Localized and systemic resistance responses are induced by *Trichoderma* species as avirulent symbionts of plants (Harman *et al.*, 2004; Horst *et al.*, 2005) [20, 22].

Traditional control measures, such as the use of chemical fungicides, give temporary relief but pose risks of environmental contamination, fungicide resistance, and disruption of beneficial soil microbiota (Abdel-Monaim *et al.*, 2023) [1]. In recent years, attention has shifted towards biological control as a sustainable and eco-friendly alternative for disease management. Several studies have shown that these biocontrol agents, when applied individually or in combination, significantly reduce disease severity and enhance plant growth parameters in chickpea and other legumes (Frontiers in Plant Science, 2023; Sharma *et al.*, 2022) [16, 39]. The synergistic interaction between *T. harzianum* and *P. fluorescens* has particularly gained interest for its broad-spectrum antifungal efficacy and ability to improve crop yield under field conditions (Mdpi Microorganisms, 2025) [30].

The present study aims to evaluate the efficacy of *T. harzianum* and *P. fluorescens*, alone and in combination, against *B. cinerea* under both laboratory and field conditions. The goal is to identify an effective biocontrol strategy that can be integrated into chickpea disease management programs to sustainably combat BGM and improve productivity.

2. Materials and Methods

2.1 Procuring Biocontrol Agents and antagonistic assay

Various strains of *T. harzianum* and *P. fluorescens* were procured from biocontrol laboratory of the Department of Plant Pathology, College of Agriculture, G B Pant University of Agriculture and Technology, Pantnagar for this study. *in vitro* antagonistic activities of both the biocontrol agents alone and in combination against *B. cinerea* were assessed using the dual culture technique. For dual cultures, a mycelial plug of each *Trichoderma* isolate (5 mm diameter) was transferred onto a PDA plate, about 1 cm from the edge of the petri dish. Similarly, a 5 mm diameter mycelial plug of *B. cinerea* removed from the colony margin of a 3-day-old culture grown on PDA was placed 6 cm away from the plug of the *Trichoderma* isolate in the same Petri dish. A PDA containing petri dish similarly inoculated with each isolate of either *Trichoderma* or *B. cinerea* alone were used as controls. For each treatment (Trichoderma isolate), three replications were maintained and incubated at 25±1 °C. All plates were incubated at 25ºC±1 °C and were examined daily until Trichoderma strains have completely covered full PDA plates in control or overgrown/ surrounded the *B. cinerea* colony. After 14 days of incubation under dual culturing, the antagonistic potential of Trichoderma strains inhibiting the pathogen's growth was measured.

The antagonism between the bacterial bio-control agent *Pseudomonas fluorescens* and the pathogen was also studied using the dual culture technique. Twenty milliliters of sterilized medium (1:1 mixture of melted PDA and King's B medium) were aseptically poured into sterilized 90 mm Petri plates and allowed to solidify. A 5 mm mycelial disc of the test fungus was cut from the edge of a 3-5-day-old culture plate using a sterilized cork borer. A sterilized paper disc (5 mm in diameter) dipped in the bacterial suspension was placed on the solidified medium opposite the fungal disc, approximately 4 cm apart. The inoculated Petri plates were incubated at 25 ± 1 °C. Plates inoculated only with the test fungus, without the bacterial antagonist, served as the control. Periodic observations were recorded on the growth of the pathogen.

At the end of incubation period, radial growth was measured.

Radial growth reduction was calculated in relation to growth of the control as follows:

% Inhibition of radial mycelial growth (I) = [(C-T)/C] × 100

Where

I= percent inhibition in pathogen mycelium growth

C = radial growth measurement of the pathogen in control plates

T =radial growth of the pathogen in the presence of *Trichoderma*.

The percentage inhibition of radial growth (PIRG) was calculated to determine efficacy of biocontrol agents against BGM pathogen.

2.2 Field Experiments

Field trials were conducted at the Crop Research Centre (CRC) of G.B. Pant University of Agriculture and Technology, Pantnagar, during the Rabi seasons of 2007-08 and 2008-09. The experiments employed a Randomized Block Design (RBD) with three replications, using the BGM-susceptible chickpea cultivar H-208. Each plot measured 3.0 × 1.5 m² with a row spacing of 30 cm. The treatments used were individual foliar application of *T. harzianum*, *P. fluorescens* and a combination of both the biocontrol agents at 15-day interval along with a control with absolutely no spray. Sprays were initiated at the onset of flowering, a critical period for BGM infection.

2.3 Data Collection and Analysis

Disease severity was assessed using a 0-9 scale, where 0 indicated no disease and 9 represented complete plant death. Grain yield and 1000-grain weight were recorded at harvest. Data were statistically analysed using ANOVA, and treatment means were compared using the Least Significant Difference (LSD) test at a 5% level of significance.

3. Results

3.1 *In vitro* Antagonistic Activity

The effect of bio-control agents for its antagonistic potential against the test pathogen *B. cinerea* was assessed following dual culture technique for both *T. harzianum* and *Ps. fluorescence*. The observations are presented in Table 1 and 2. Periodic observation on the colonization of the test fungus

by the selected bio-agents indicated their varied antagonistic potentiality. All tested strains of *T. harzianum* and *P. fluorescens* exhibited significant inhibitory effects against *B. cinerea* in dual culture assay. Notably, the combination treatment showed the highest PIRG, indicating synergistic interactions between the two biocontrol agents. Maximum inhibition 77.19% after 72 h of incubation was recorded in Th 34 followed by Th 41, where inhibition was 70.17%. Minimum inhibition (63.1%) was recorded in the culture treated with Th 39, Th 42, Th 43, Th 46 respectively (Fig. 1).

Different strains of *Ps. fluorescence* were tested for their antagonistic potential against *B. cinerea*. A clear zone of inhibition between *B. cinerea* and *Ps. fluorescence* was observed. In inhibition zone no mycelial growth of *B. cinerea* was visible. All the 12 strains of *Ps. fluorescence* were found effective in inhibiting the growth of test fungus. Maximum inhibition zone was observed in strain 84, where diameter of inhibition zone was 25.5mm followed by strain 82 where inhibition zone was 24.4 mm. Minimum inhibition zone 12.4 mm was observed in strain 3 and 25, respectively (Fig. 2).

3.2 Effect of bio-control agents' seed treatment and spray on the severity of grey mould, grain yield and 1000 grain weight

Biological control has been considered a potential strategy for use in combination with conventional chemical control methods. Fungal antagonists have been successfully used to control soil-borne diseases. It has been recognized that pathogen-saprophyte interactions are not confined solely to the soil environment, but also occur on the surface of plant parts. In view of the potential of *Trichoderma* spp. as effective antagonistic agents, experiments were conducted to explore the possibility of using *Trichoderma* spp. for the control of grey mould in chickpea. The observations are presented in Table 3. The results revealed that seed treatment with a mixture of *T. harzianum* and *Pseudomonas fluorescens*, followed by three foliar sprays of the same combination, resulted in effective disease control. This treatment also led to an increase in grain yield compared to the untreated control.

During 2007-08 crop season the data recorded revealed that plots treated with a mixture of *T. harzianum* and *Ps. fluorescence* showed lowest disease severity (3.9) followed by *Ps. fluorescence* (4.9) and *T. harzianum* (5.0) alone. The highest grain yield (1810 kg/ha) was recorded in *T. harzianum* + *Ps. fluorescence* treated plots followed by *Ps. fluorescence* (1550 kg/h) and *T. harzianum* (1508 kg/ha) alone. Maximum 1000 grain weight (112g) was recorded in *T. harzianum* + *Ps. fluorescence* treated plots followed by *Ps. fluorescence* and *T. harzianum* (106g) respectively while in control 1408 kg/h yield was recorded. Maximum decline in disease incidence (45.83%) and increase in grain yield (28.91%) was recorded in *T. harzianum* + *Ps. fluorescence* treated plots.

During the year 2008-09, the data recorded revealed that *T. harzianum* + *Ps. fluorescence* treated plots again resulted lowest disease incidence (4.3) followed by *Ps. fluorescence* and *T. harzianum*. Highest grain yield (1890 kg/ha) and 1000 grain weight (105g) was recorded in *T. harzianum* and *Ps. fluorescence* treated plots. 1398 kg/h yield was recorded in control plot. Maximum percent decline in disease incidence (41.66%) and increase in grain yield (35.19%) was recorded *T. harzianum* and *Ps. fluorescence* in treated plots (Fig 3). In both cropping seasons, all biocontrol treatments significantly reduced BGM severity compared to the control. The combination treatment consistently resulted in the lowest disease severity scores. Correspondingly, plots receiving the combination treatment recorded the highest grain yields and 1000-grain weight. These findings suggest that integrating *T. harzianum* and *P. fluorescens* enhances disease suppression and promotes plant health.

4. Discussion

The study reaffirms the potential of *T. harzianum* and *P. fluorescens* as effective biocontrol agents against BGM in chickpea. Their combined application not only suppresses disease development but also enhances yield parameters, likely due to synergistic effects.

In the present investigation, different strains of *Trichoderma harzianum* and *Pseudomonas fluorescens* were screened for their antagonistic potential against *Botrytis cinerea*. The results revealed that all strains of *T. harzianum* were effective, as they overgrew and completely parasitized *B. cinerea*. The *T. harzianum* strains exhibited faster growth compared to *B. cinerea*. The suppression of pathogen growth may be attributed to nutrient competition and the production of inhibitory compounds by the antagonists. Growth inhibition could also result from hyphal parasitization and the secretion of cell wall-degrading enzymes by the test antagonists. However, these mechanisms require further investigation. The antagonism of *Trichoderma* strains against *B. cinerea* has also been reported by Mukherjee *et al.* (1997) [32] in which they obtained stable benomyl tolerant mutants of *T. viride* isolate T-15 for use in integrated biocontrol of botrytis grey mould of chickpea. Elad *et al.* (1993) [12] found that *T. harzianum* is antagonistic to *B. cinerea*. Similar observations have been reported by Mukherjee *et al.* (1995) [31] where they found both preventive and curative action of *Trichoderma*. Chaurasia and Joshi (2001) [6] also reported the antagonistic nature of *T. viride* against *B. cinerea*. Lee *et al.* (2006) [27] also observed the biological control of *B. cinerea* on cucumber by foliar sprays of *T. harzianum*. The bacterial antagonists *Ps. fluorescence* used in present investigation inhibited the growth of *B. cinerea*. Microscopic observation revealed that mycelium of test pathogen was broken and disintegrated from which cytoplasm ended to exude out. A number of biological activities have been postulated to be involved in successful inhibition of the growth of the pathogen. These include production of antagonistic compounds, such as antibiotics, siderophores, ammonia and hydrolytic enzymes (Lam and Gaffney, 1993) [26]. Involvement of siderophores in suppression of pathogen by *Ps. fluorescence* was reported by Baker and Cook (1974) [4]; Klopper *et al.* (1980) [24] and Yeole and Dube (2000) [45]. However, extensive information is lacking regarding the effectiveness of these biocontrol agents against *B. cinerea*. Effect of biocontrol agents on disease severity, grain yield and 1000-grain weight indicates that among the biocontrol agents, a combination of *T. harzianum* + *Ps. fluorescens* resulted in minimum disease severity and maximum grain yield, while *T. harziamum* alone resulted in higher disease incidence and minimum grain yield in comparison to other treatments. It provides a habitat for epiphytic saprophytic micro-organisms, many of which are capable of influencing the growth of pathogens and of reducing the incidence of

foliar diseases (Bakeman and Fokkema, 1982) [5]. Biological control of many foliar diseases has already been reported by several workers. Some workers have also reported *Trichoderma* spp. as biocontrol agent of *B. cinerea* (Dubos and Bulit, 1981; Tronsmo and Ray, 1977 and Wood, 1950) [11, 42, 43].

Recent advancements have identified additional biocontrol agents with promising activities against *B. cinerea*. For instance, *P. lilacinum* has demonstrated antifungal metabolite production effective against BGM. Moreover, rhizosphere bacteria like *P. megaterium* and *S. marcescens* have shown significant disease suppression in chickpea. These findings highlight the importance of exploring diverse microbial consortia for integrated disease management.

The mechanisms underlying biocontrol efficacy include competition for nutrients and space, production of antifungal compounds, and induction of systemic resistance in host plants. Understanding these interactions is crucial for developing effective and sustainable biocontrol strategies.

Table 1: Radial growth of *B. cinerea* and percent inhibition by selected strains of *T. harzianum* at different incubation periods in dual culture

Isolate No.	24HAI		48HAI		72HAI	
	Radial growth (mm)	Percent inhibition	Radial growth (mm)	Percent inhibition	Radial growth (mm)	Percent inhibition
Th-34	7.6	41.53	12	42.47	13	77.19
Th-37	8.3	36.15	12	42.47	19	66.66
Th-39	8.6	33.84	17	24.66	21	63.15
Th-40	8	38.46	20	11.50	20	64.91
Th-41	8	38.46	15	33.62	17	70.17
Th-42	7	46.15	14	38.05	21	63.15
Th-43	7.3	43.84	14	38.05	21	63.15
Th-44	7.5	42.30	14	38.05	19	66.66
Th-45	7.5	42.30	15	33.62	20	64.91
Th-46	7.6	41.53	15	33.62	21	63.15
Control	13	-	22.6	-	57	-
S.Em±	0.631	-	0.732	-	0.767	-
CD (P=0.05)	1.85	-	2.14	-	2.25	-
CV (%)	13.273	-	8.057	-	5.857	-

Table 2: Diameter of inhibition zone in Dual culture of Fluorescent *Pseudomonads* against *B. cinerea*

Isolate No.	Inhibition Zone (mm)
FL P 2	18.5
FL P 3	12.4
FL P 4	21.1
FL P 11	20.1
FL P 12	15.7
FL P 18	19.2
FL P 25	12.4
FL P 27	17.6
FL P 31	13.8
P f 82	24.4
FL P 90	12.8
FL P 84	25.5
Control	0.00
S Em±	0.673
CD(P=0.05)	1.95

Table 3: Effect of biocontrol agents on Disease Severity, grain yield and 1000-grain weight during 2007-08 and 2008-09 crop seasons

Treatment	Dose (g/kg)	Disease Severity (1-9 scale)		Disease decline (%)		Grain yield (kg/ha)		Percent increase over check		1000-grain weight (g)	
		2007-08	2008-09	2007-08	2008-09	2007-08	2008-09	2007-08	2008-09	2007-08	2008-09
Pseudomonas fluorescens	5.0	4.9	5.1	31.9	30.55	1550	1651	10.39	17.88	106	103
Trichoderma harzianum	5.0	5.0	5.3	30.5	27.77	1508	1503	7.40	6.15	106	100
Trichoderma harzianum + *Pseudomonas fluorescens*	5.0	3.9	4.3	45.83	41.66	1810	1890	28.91	35.19	112	105
Control	-	7.2	7.3	-	-	1404	1398	-	-	95	92
S Em±		0.073	0.076	-	-	5.1	5.5	-	-	1.13	0.88
CD(P=0.05)		0.25	0.26	-	-	17.6	18.9	-	-	1.8	3.04

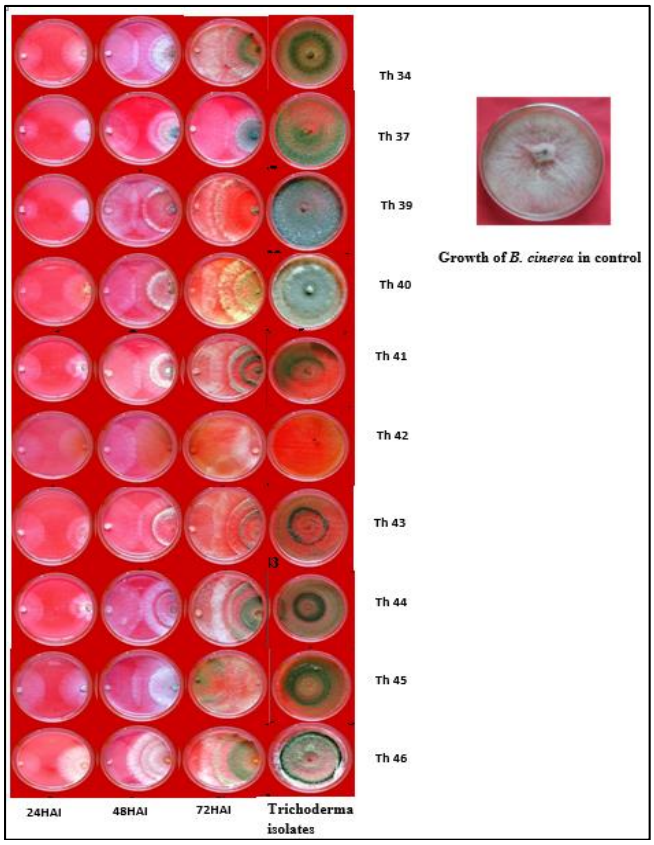

Fig 1: Antagonistic activity of selected *Trichoderma* isolates against BGM of chickpea after 24, 48, and 72 hours, respectively.

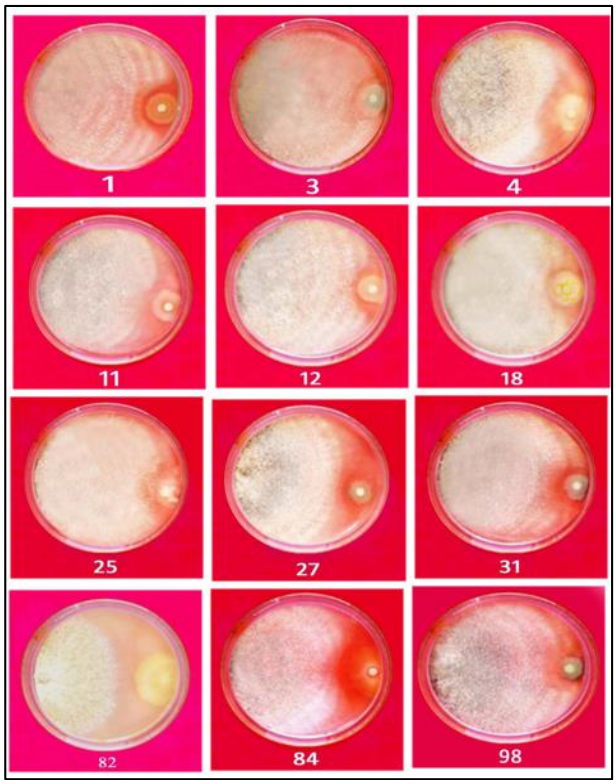

Fig 2: Antagonistic activity of Fluorescent *Pseudomonas* isolates against *B. cinerea* showing inhibition zone formed by Fluorescent pseudomonad isolates 1, 3, 4, 11, 12 and 18, 25, 27, 31, 82, 84 and 98

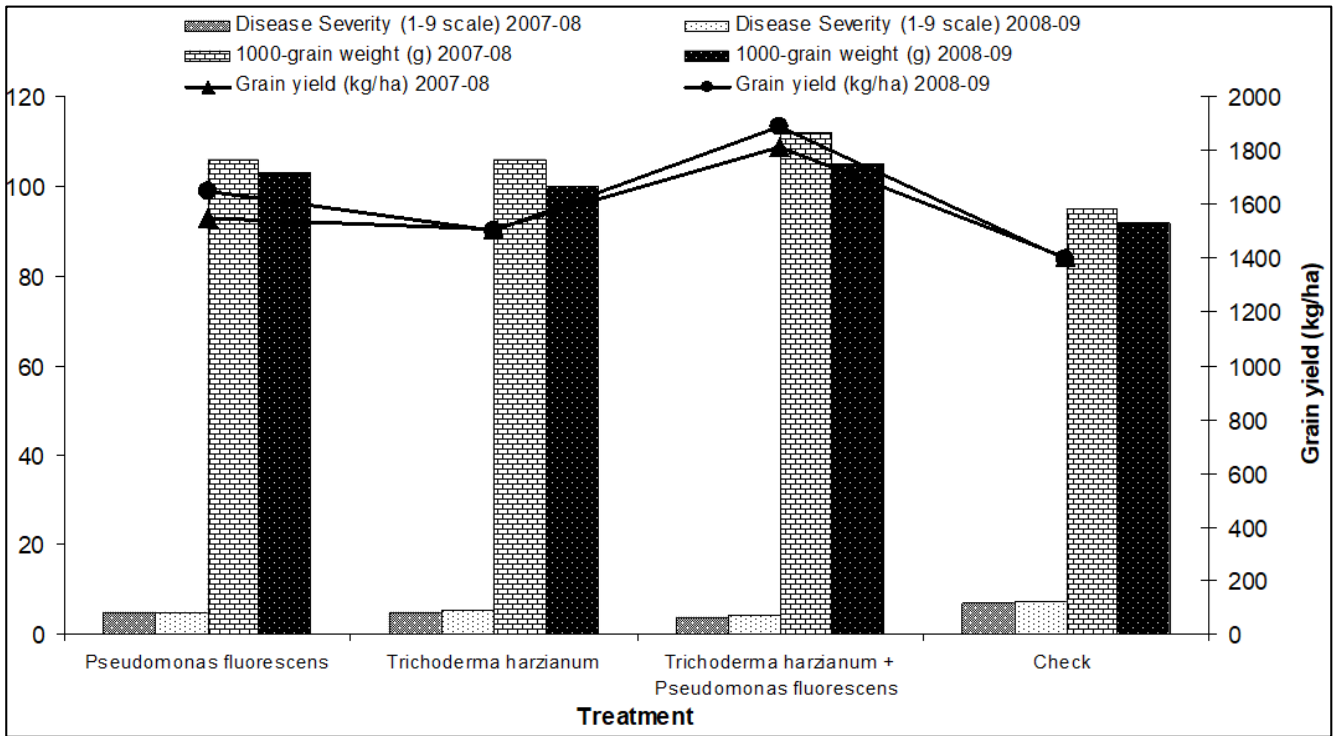

Fig 3: Effect of biocontrol agents on disease severity, grain yield and 1000-grain weight during 2007-08 and 2008-09 crop seasons

5. Conclusion

The integration of *T. harzianum* and *P. fluorescens* offers a viable and eco-friendly approach to managing BGM in chickpea. Their combined application not only reduces disease severity but also enhances yield attributes. Future research should focus on field validation of other promising biocontrol agents, such as *P. lilacinum*, *P. megaterium*, and *S. marcescens*, and explore their synergistic interactions for comprehensive disease management. pubmed.ncbi.nlm.nih.gov

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7. References

1. Abdel-Monaim MF, *et al.* Fungicide resistance and biocontrol options in pulse crops. Egyptian Journal of Biological Pest Control. 2023;33(1):14.
2. Agrios GN. Plant Pathology. 4th ed. New York, NY: Academic Press; 1997. 606 p.
3. Altomare C, Norvell WA, Björkman T, Harman GE. Solubilization of phosphates and micronutrients by the plant-growth-promoting and biocontrol fungus *Trichoderma harzianum* Rifai 1295-22. Applied and Environmental Microbiology. 1999;65(7):2926-2933.
4. Baker KF, Cook RJ. Biological Control of Plant Pathogens. San Francisco, CA: W.H. Freeman and Co.; 1974. 433 p.
5. Bakeman JP, Fokkema NJ. Potential for biological control of plant diseases on the phylloplane. Annual Review of Phytopathology. 1982;20:167-192.
6. Chaurasia PCP, Joshi S. Management of Botrytis Gray Mould (*Botrytis cinerea* Pers. ex. Fr.) of chickpea at Tarahara, Nepal. Nepal Agricultural Research Journal. 2001;5:21-37.
7. Chohan SA, Imran M, Ahmad F, *et al. Trichoderma*-based formulations control wilt disease of chickpea. PeerJ. 2024;12:e17835. https://doi.org/10.7717/peerj.17835
8. Choi IS, Chung YR, Cho KY. Variations in phenotypic characteristics, pathogenicity and fungicide resistance of *Botrytis cinerea*, gray mould rot fungus, isolated from various host plants. Korean Journal of Mycology. 1995;23:246-256.
9. Cook RJ, Granados RR. Biological control: making it work. In: MacDonald MJ, editor. Agricultural Biotechnology at the Crossroads. Ithaca, NY: National Agricultural Biotechnology Council; 1991. p. 213-227.
10. De Meyer G, Bigirimana J, Elad Y, Höfte M. Induced systemic resistance in *Trichoderma harzianum* T39 and biocontrol of *Botrytis cinerea*. European Journal of Plant Pathology. 1998;104:279-286.
11. Dubos B, Bulit J. Filamentous fungi as biocontrol agents on aerial plant surfaces. In: Blackman JP, editor. Microbial Ecology of the Phylloplane. London: Academic Press; 1981. p. 353-367.
12. Elad Y, Zimand G, Zaqs Y, Zuriel S, Chet I. Use of *Trichoderma harzianum* in combination or alternation with fungicides to control cucumber grey mould (*Botrytis cinerea*) under commercial greenhouse conditions. Plant Pathology. 1993;42:324-332.
13. Elad Y, Kapat A. The role of *Trichoderma harzianum* protease in the biocontrol of *Botrytis cinerea*. European Journal of Plant Pathology. 1999;105:177-189.
14. Elad Y. *Trichoderma harzianum* T39 preparation for biocontrol of plant diseases—control of *Botrytis cinerea*, *Sclerotinia sclerotiorum* and *Cladosporium fulvum*. Biocontrol Science and Technology. 2000a;10:499-507.
15. Elad Y. Biological control of foliar pathogens by means of *Trichoderma harzianum* and potential modes of action. Crop Protection. 2000b;19:709-714.
16. Frontiers in Plant Science. Biocontrol of *Botrytis cinerea* by *Pseudomonas protegens*. 2023;14:1172349.
17. Grindle M. Variations among field isolates of *Botrytis cinerea* in their sensitivity to antifungal compounds. Pest Science. 1981;12:305-312.
18. Hadar Y, Chet I, Hennis Y. Biological control of *Rhizoctonia solani* damping off with wheat bran culture of *Trichoderma harzianum*. Phytopathology. 1979;69:64-68.
19. Harman GE. Myths and dogmas of biocontrol: changes in perceptions derived from research on *Trichoderma harzianum* T-22. Plant Disease. 2000;84:377-393.
20. Harman GE, Howell CR, Viterbo A, Chet I, Lorito M. *Trichoderma* species—opportunistic, avirulent plant symbionts. Nature Reviews Microbiology. 2004;2:1-14.
21. Hjeljord LG, Stensvand A, Tronsmo A. Antagonism of nutrient-activated conidia of *Trichoderma harzianum* (atroviride) P1 against *Botrytis cinerea*. Phytopathology. 2001;91:1172-1180.
22. Horst LE, Locke J, Krause CR, McMahon RW, Madden LV, Hoitink HAJ. Suppression of *Botrytis* blight of begonia by *Trichoderma hamatum* 382 in peat and compost-amended potting mixes. Plant Disease. 2005;89:1195-1200.
23. Khan MR, *et al.* Management of gray mould of chickpea with bacterial and fungal biopesticides. Biological Control. 2011;57(1):13-23.
24. Klopper JW, Leong J, Teintze M, Schroth MN. Enhanced plant growth by siderophores produced by plant growth promoting rhizobacteria. Nature. 1980;286:885-886.
25. Kumar A, Sharma R, Singh P. Integrated application of microbial antagonists for management of foliar pathogens in legumes. Frontiers in Plant Science. 2023;14:1172349. https://doi.org/10.3389/fpls.2023.1172349
26. Lam ST, Gaffney TD. Biological activities of bacteria used in plant pathogen control. In: Biotechnology in Plant Disease Control. North Carolina: Wiley-Liss; 1993. p. 291-320.
27. Lee SK, Sohn HB, Kim GG, Chung YR. Enhancement of biological control of *Botrytis cinerea* on cucumber by foliar sprays and bed potting mixes of *Trichoderma harzianum* YC459 and its application on tomato in the greenhouse. Plant Pathology Journal. 2006;22(3):283-288.
28. Lewis JA, Papavizas GC. Characterization of alginate pellets formulated with *Trichoderma* and *Gliocladium* and their effect on the proliferation of the fungi in soil. Plant Pathology. 1985;34:571-577.
29. Lorito M, Harman GE, Hayes CK, Broadway RM, Tronsmo A, Woo SL, Di Pietro A. Chitinolytic enzymes produced by *Trichoderma harzianum*: antifungal activity of purified endochitinase and chitobiosidase. Phytopathology. 1993;83:302-307.
30. Mdpi Microorganisms. Biocontrol efficacy of *Trichoderma* strains in chickpea. Microorganisms. 2025;13(1):121.
31. Mukherjee PK, Haware MP, Jayanthi S. Preliminary investigations in integrated biocontrol of *Botrytis* gray mould of chickpea. Indian Phytopathology. 1995;48:141-149.
32. Mukherjee PK, Haware MP, Raghu K. Induction and evaluation of benomyl tolerant mutants of *Trichoderma viride* for biological control of *Botrytis* gray mould of chickpea. Indian Phytopathology. 1997;50:485-489.

33. Mukhopadhyay AN, Shrestha SM, Mukherjee PK. Biological seed treatments for control of seedborne plant pathogens. FAO Plant Protection Bulletin. 1992;40:1-10.
34. Nelson ME, Powelson ML. Biological control of gray mould of snap beans by *Trichoderma hamatum*. Plant Disease. 1988;72:727-729.
35. Pande S, *et al.* Integrated management of *Botrytis* gray mould of chickpea. ICRISAT Technical Bulletin. 2010.
36. Paulitz TC, Belanger RR. Biological control in greenhouse systems. Annual Review of Phytopathology. 2001;39:103-133.
37. Redmond JC, Marois JJ, Macdonald JD. Biological control of *Botrytis cinerea* on roses with epiphytic microorganisms. Plant Disease. 1987;71:799-802.
38. Roca-Couso R, Flores-Félix JD, Rivas R. Mechanisms of action of microbial biocontrol agents against *Botrytis cinerea*. Journal of Fungi. 2021;7(12):1045. https://doi.org/10.3390/jof7121045
39. Sharma P, *et al.* Efficacy of microbial consortia against chickpea root pathogens. Journal of Plant Pathology. 2022;104:15-23.
40. Singh PP, Shin YC, Park CS, Chung YR. Biological control of *Fusarium* wilt of cucumber by chitinolytic bacteria. Phytopathology. 1999;89:92-99.
41. Sutton JC, Peng G. Biocontrol of *Botrytis cinerea* in strawberry leaves. Phytopathology. 1993;83:615-621.
42. Tronsmo A, Ray J. Antagonistic action of *Trichoderma pseudokoningii* against the apple pathogen *Botrytis cinerea*. Phytopathologische Zeitschrift. 1977;89:216-220.
43. Wood RKS. The control of diseases of lettuce by the use of antagonistic organisms: The control of *Botrytis cinerea* Pers. Annals of Applied Biology. 1950;38:203-216.
44. Yedidia I, Benhamou N, Chet I. Induction of defense responses in cucumber plants (*Cucumis sativus* L.) by the biocontrol agent *Trichoderma harzianum*. Applied and Environmental Microbiology. 1999;65:1061-1070.
45. Yeole RD, Dube HC. Siderophore mediated antibiosis of rhizobacterial fluorescent pseudomonads against certain soil borne fungal plant pathogens. Journal of Mycology and Plant Pathology. 2000;30:335-338.
46. Zhang Y, Wu J, Li M. Advances in microbial consortia for plant disease control and growth promotion. Journal of Applied Microbiology. 2024;136(2):215-229. https://doi.org/10.1111/jam.16024