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## Antagonistic effects of *Chaetomium globosum* and *Trichoderma viride* against Chili Anthracnose

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Rohaendi, P.<sup>1</sup>, Sudjatismiko, S.<sup>2\*</sup>, Pamekas, T.<sup>3</sup>, Soyotong, K.<sup>4</sup>, Song, J. J.<sup>5</sup>, Ganefianti, D. W.<sup>2</sup>, Bustamam, H.<sup>3</sup> and Sutrawati, M.<sup>3</sup>

<sup>1</sup>Faculty of Agriculture University of Bengkulu, Indonesia; <sup>2</sup>Department of Crop Production, Faculty of Agriculture University of Bengkulu, Indonesia; <sup>3</sup>Department of Plant Protection, Faculty of Agriculture University of Bengkulu, Indonesia; <sup>4</sup>Excellence Center-Research Institute of Modern Organic Agriculture (RIMOA), King Mongkut's Institute of Technology Ladkrabang, Bangkok, Thailand; <sup>5</sup>King Mongkut's Institute of Technology Ladkrabang, Prince of Chumporn Campus, Thailand.

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**Abstract** The research demonstrated that *Trichoderma viride* and *Chaetomium globosum* inhibited *Colletotrichum acutatum* by 90.30% and 88.20%, respectively, primarily via competition for nutrients/space and parasitism, with no observed antibiosis. The field trials revealed that both fungi significantly reduced anthracnose severity. *Trichoderma viride* applications at 1, 2, and 3 g/L decreased disease incidence by 80.68%, 91.16%, and 92.27%, respectively, while *Chaetomium globosum* reduced it by 87.61–92.65%. Both exceeded synthetic fungicide Antracol (83.48–89.37% reduction) as a controlled treatment. Attack intensity followed a similar trend, with *Trichoderma viride* (95.12–97.46% suppression) and *Chaetomium globosum* (92.76–96.83%) surpassing Antracol (89.95–93.80%). Additionally, *Trichoderma viride* and *Chaetomium globosum* enhanced salicylic acid levels in chili plants, suggesting induced systemic resistance. These findings highlighted the potential of antagonistic fungi as sustainable biocontrol agents which would develop to be biofungicide, offering effective anthracnose management while reducing chemical residues.

**Keywords:** Antagonism, Biocontrol, Chemical residues, Inhibitory power, Systemic resistance

### Introduction

Chilli (*Capsicum annum* L.) is one of the most important tropical and subtropical crop systems and it ranks fourth among the world crops. Approximately 400 species of chilies are grown around the world. The genus *Capsicum* was autogenous to the American tropics and has been distributed worldwide including the tropical, subtropical, and temperate zones (Pickersgill,

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\* **Corresponding Author:** Sudjatismiko, S.; **Email:** sigitsudjatismiko@unib.ac.id

1997). *Capsicum* fruit is known by different names such as ‘chilli’, ‘chilli pepper’ or simply ‘pepper’ in accordance with region (Isaac, 1992).

Besides, chilli peppers are also highly economical value in a form of raw materials for many industrial goods. In general, red chili peppers contain high values of nutrients and vitamins: caloric, protein, fat, carbohydrate, calcium (Kantar *et al.*, 2016), and recently it has been reported as antioxidant and an anti-obesity agent (Azlan *et al.*, 2022). Thus, red chili has been one of the commodities with high interest for cultivation, including in Indonesia. However, during the cultivation process, they often experience disturbances that cause a decrease in chili plant productivity and even lead to crop failure.

Anthracoze represents one of the most significant diseases affecting red chili pepper plants (*Capsicum annuum* L.), caused by fungal pathogens belonging to the genus *Colletotrichum*. It can develop on the field, during long distant transport, cold storage and shelf-life. In conventional agriculture, the whole plant including the fruits, are sprayed with fungicides as a prerequisite for post-harvest control of chilli anthracnose. This disease manifests through characteristic symptoms including by very dark, sunken lesions, containing spores cause dieback (shoot blight) in mature plants and fruit infections, leading to substantial yield reductions (Isaac, 1992). According to BPPP (2016), in Indonesia anthracnose outbreaks can result in crop losses reaching 90%, particularly during rainy seasons. Taxonomically, the causal pathogen is classified within Kingdom: Fungi; Phylum: Ascomycota; Class: Sordariomycetes; Order: Glomerellales; Family: Glomerellaceae, with its perfect stage (teleomorph) identified as *Glomerella* (Agrios, 2005). Several *Colletotrichum* species have been implicated in the disease etiology, including *C. acutatum*, *C. coccodes*, *C. gloeosporioides*, *C. atramentarium*, *C. dematium*, and *C. capsici* (Gopinath *et al.*, 2006).

Antagonistic fungi represent a group of microorganisms capable of suppressing pathogen growth through multiple mechanisms, including spatial and nutrient competition, antibiosis (production of both volatile and non-volatile antibiotic compounds, as well as lytic enzymes such as chitinases, proteases, and glucanases), parasitism (hyphal coiling and penetration of pathogenic fungi), and induction of plant systemic resistance (Agrios, 2005; Vinale *et al.*, 2008). A notable example is *Chaetomium globosum*, which has demonstrated significant antagonistic activity against soil- and seed-borne pathogens (Jiang *et al.*, 2017; Zhang *et al.*, 2021). Formulated products derived from *Chaetomium* have even been evaluated as broad-spectrum mycofungicides (Soytong and Soytong, 1996). Wheat seeds coated with a suspension of *Chaetomium globosum* 12XP1-2-3 spores showed better growth and had a controlling effect on *Fusarium pseudograminearum* attacks, as well as increasing wheat yields (Feng *et al.*,

2023). *Chaetomium globosum* and *Trichoderma sp.* fungi also significantly improve cucumber seedling growth and root system development, while improving rhizosphere soil properties (Yu *et al.*, 2025). The fungus *Chaetomium cupreum* has been proven to inhibit *Fusarium oxysporum*, which causes wilt disease in Sida and Cherr tomato varieties (Sibounnavong *et al.*, 2012), and to control leaf spot disease in Sen Pidoa rice varieties caused by *Curvularia lunata* (Tann and Soyong, 2016). Additionally, *Penicillium funiculosum* has been reported to effectively inhibit *Phytophthora spp.*, the causal agents of root rot (Fang and Tsao, 1995), highlighting the critical role of these biocontrol agents in integrated disease management strategies.

*Trichoderma* species, particularly *Trichoderma viride*, constitute a group of antagonistic fungi commonly found in agricultural soils (Harman *et al.*, 2004). Their pathogen-suppressing capacity is mediated through three primary mechanisms: (1) competition (dominance in spatial colonization and nutrient acquisition), (2) mycoparasitism (hyphal coiling, penetration, and subsequent lysis of pathogenic fungi), and (3) antibiosis (production of hydrolytic enzymes including chitinases, glucanases, and pectinases) (Harman and Kubicek, 1998). These synergistic mechanisms enable *Trichoderma spp.* to effectively control the spread of fungal phytopathogens (Asad, 2022).

As established in the literature, anthracnose disease caused by *Colletotrichum spp* presents a serious threat to red chili pepper productivity, with documented yield losses reaching 90% (BPPP, 2016). Conventional chemical control methods carry inherent risks of pathogen resistance development and environmental contamination. While antagonistic fungi such as *Chaetomium globosum* and *Trichoderma viride* have demonstrated broad-spectrum pathogen suppression through multiple mechanisms (Agrios, 2005; Harman and Kubicek, 1998), their specific efficacy against *Colletotrichum spp* in chili pepper systems remains insufficiently characterized. This study therefore aimed to evaluate the inhibitory effects of *Chaetomium globosum* and *Trichoderma viride* against *Colletotrichum spp.* through both in vitro and in planta assays, and to elucidate the predominant antagonistic mechanisms involved, thereby establishing a scientific foundation for developing sustainable biocontrol strategies.

## Materials and methods

The experiment, both in vitro and in vivo was conducted from April to November 2024. The in vivo experiment was conducted at the Plant Protection Laboratory, Faculty of Agriculture, Bengkulu University, involving several stages: Firstly, the rejuvenation of antagonistic fungi *Trichoderma viride* collected by the Agronomy Laboratory and *Chaetomium globosum* collected

from Excellence Center-Research Institute of Modern Organic Agriculture (RIMOA), King Mongkut's Institute of Technology Ladkrabang (KMITL), Bangkok, Thailand. Secondly, the purification of the pathogen *Colletotrichum acutatum*, and thirdly the antagonistic tests between *Colletotrichum acutatum* and both *Chaetomium globosum* and *Trichoderma viride*. The equipments and materials used such as Laminar Air Flow (LAF), petri dishes, cork-borer, ose needle, plastic wrap, and a Bunsen burner, anthracnose-infected chili fruit, Potato Dextrose Agar (PDA) media, 70% alcohol, and isolates of *Chaetomium globosum*, *Trichoderma viride*, and *Colletotrichum acutatum*.

### ***Rejuvenation of Chaetomium globosum and Trichoderma viride***

The isolates of *Chaetomium globosum* and *Trichoderma viride*, were rejuvenated on fresh PDA media by dissolving 39 g of PDA in 1000 mL of distilled water, boiling until homogeneous, and cooling to 36-37°C. The mixture was then sterilized in an autoclave (121°C, 15 min, 1 atm), poured into petri dishes, and allowed to solidify. Once hardened, the isolates were inoculated onto the media via surface scratching and incubated for 7 days to obtain pure cultures.

### ***Isolation of Colletotrichum acutatum***

Chili fruits displaying anthracnose symptoms were collected from farmers' fields in Lubuk Linggau, South Sumatera Indonesia, placed in labeled plastic bags, and transported for pathogen isolation. Diseased tissue samples (0.5 cm × 0.5 cm) were surface-sterilized with 70% alcohol, plated onto PDA media in sterilized Petri dishes (flame-sterilized near a Bunsen burner), and sealed with plastic wrap. After incubation, fungal growth matching anthracnose characteristics was sub-cultured on day 8 to obtain a pure isolate of *Colletotrichum* sp.

### ***In vitro antagonist test***

The antagonistic test was conducted using the dual culture method. The first test involved *Chaetomium globosum* against *Colletotrichum acutatum*. A 9 cm Petri dish containing PDA medium was prepared, and a 7-day-old pure culture of *Chaetomium globosum* was inoculated using a 0.5 cm diameter cork-borer from the colony edge. The *Chaetomium globosum* isolate was placed 2 cm from the edge of the Petri dish and labeled "C". Next, a pure culture of *Colletotrichum acutatum* was extracted similarly and inoculated 2 cm from the opposite edge of the dish, labeled "A". As a control, the pathogen was placed on

PDA medium without *Chaetomium globosum*. Colony growth was observed until contact occurred between the two fungi.

The second antagonistic test followed the same procedure, pairing *Trichoderma viride* with *Colletotrichum acutatum*. A 7-day-old *Trichoderma viride* culture was inoculated 2 cm from the edge and labeled "T", while *Colletotrichum acutatum* was placed opposite it (labeled "A"). A control was set up with the pathogen alone. Colony interactions were monitored until contact was observed between *Trichoderma viride* and *Colletotrichum acutatum*.

### **Percentage analysis of barriers**

The observation of colony diameter and growth area of an antagonist fungi (*Trichoderma viride* or *Chaetomium globosum*) and *Colletotrichum acutatum* was conducted from 1 DAI (Days After Inoculation) up to 8 DAI. The inhibition percentage was calculated using the following formula:

$$Z = \frac{(r_1 - r_2)}{r_1} \times 100\%$$

Notes:

Z = Inhibition percentage (%)

r<sub>1</sub> = Radius of *Colletotrichum acutatum* without *Trichoderma viride* (control)

r<sub>2</sub> = Radius of *Colletotrichum acutatum* with *Trichoderma viride* (treatment)

The calculation was performed by measuring the radial growth diameter of the fungus along a single straight line using a millimeter-scale ruler.

### **Field trial**

The field trial was conducted at the greenhouse of the Lubuk Linggau Agricultural Office, South Sumatra, Indonesia, using a Split Plot Design. The first factor (main plot) was the endophytic fungi *Chaetomium globosum*, *Trichoderma viride*, and the fungicide Antracol. The second factor (subplot) consisted of 3 concentration konidia levels, namely 1 g/L, 2 g/L, and 3 g/L. All treatments were repeated 3 times, and each repetition was represented by 3 sample plants. Chili pepper seedlings were transplanted to polybags consist of medium topsoil, manure and charcoal rice husks (2:1:1). The planting distance between polybags of was 50 x 50 cm. Maintenance of the chili pepper plants included daily watering, fertilization with NPK (16-16-10) at a dose of 250 kg/ha, and control of plant pests.

Inoculation of *Colletotrichum acutatum* on plants was carried out 60 days after sowing when the chili fruits were fully formed, by dissolving the pathogen

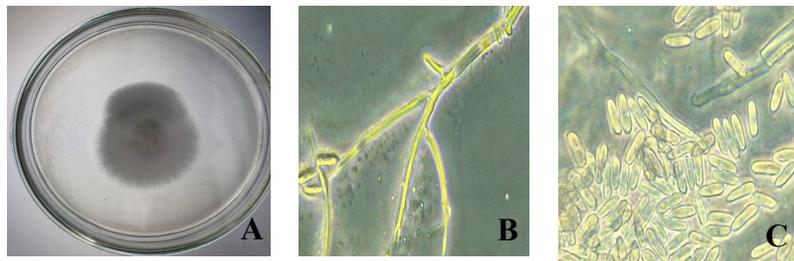
in sterile water, stirring until homogeneous and spraying all the attached chili fruits using a mini sprayer. The fungal suspension used a density of  $10^6$  spores (cells)/ml. Seven days after inoculation, the plants were treated with *Chaetomium globosum*, *Trichoderma viride*, and Antracol according to the treatment concentration.

The observation data were analyzed using analysis of variance (ANOVA) at a 95% confidence level ( $\alpha = 0.05$ ) using SAS version 9.4, followed by Least Significant Difference at a 95% confidence level.

## Results

### *Identification Colletotrichum acutatum*

Pathogenic fungus (*Colletotrichum* spp.) was isolated from anthracnose-infected fruit samples and subsequently examined under a microscope at  $100\times$  magnification. The observation results are presented in Figure 1.



**Figure 1.** Macroscopic and Microscopic Views of *Colletotrichum acutatum*  
Description: Macroscopic morphology of *Colletotrichum acutatum*; B) Hyphae of *Colletotrichum acutatum*; and C) conidia of *Colletotrichum acutatum*

*Colletotrichum acutatum* exhibited distinct macroscopic characteristics, forming white colonies with pink centers that darkened to brown or black with age, showing rapid growth that fully covered PDA plates by day 7 (Figure 1A). Microscopic examination ( $100\times$ ) of chili samples confirmed *Colletotrichum* spp. identification, revealing dark conidiophores producing elongated, rod-shaped conidia with  $\geq 3$ , along with lunate (crescent-shaped) hyaline conidia, septate branched hyphae, and appressoria. Colonies displayed white, cottony mycelium with smooth margins and cream-colored undersurfaces, demonstrating both parasitic and saprophytic behavior.

### ***Colony inhibition percentage of Chaetomium globosum on Colletotrichum acutatum***

The inhibitory effect of *Chaetomium globosum* on *Colletotrichum acutatum* was observed from day 1 to day 8 after inoculation. The antagonistic inhibition began on the first day post-inoculation, with a notable increase in inhibition (11.1%) becoming apparent by day 3. By day 8, the antagonistic fungus had completely overgrown the pathogen colony, achieving an inhibition percentage of 88.2%. These results demonstrated that *Chaetomium globosum* is found to be highly effective in suppressing the growth of *Colletotrichum acutatum*.

### ***Percentage of colony inhibition of Trichoderma viride on Colletotrichum acutatum***

The inhibitory activity of *Trichoderma viride* against *Colletotrichum acutatum* was evaluated daily from days 1 to 8 post-inoculation. Antagonistic inhibition was detectable from the first day, with a significant increase (6.7%) observed by day 3. By day 8, *Trichoderma viride* had completely overgrown the pathogen colony, achieving 90.3% inhibition. These results demonstrated *Trichoderma viride's* strong antagonistic efficacy against *Colletotrichum acutatum*. The progressive inhibition pattern between *Trichoderma viride* and *Colletotrichum acutatum* throughout the 8-day period.

### ***Antagonistic mechanisms of fungal isolates against Colletotrichum acutatum***

The antagonistic mechanisms observed between the two fungal antagonist isolates involved competition for space, nutrients, and oxygen, as well as antibiosis and parasitism. These antagonistic interactions were assessed on day 8 post-incubation, with the results presented in Table 1.

**Table 1.** The mechanism of antagonistic fungi against the pathogen *Colletotrichum* sp.

Isolates	Competition for space, nutrients, and oxygen	Antibiosis	Lysis and Parasitism
<i>Chaetomium globosum</i>	+	-	+
<i>Trichoderma Viride</i>	+	-	+

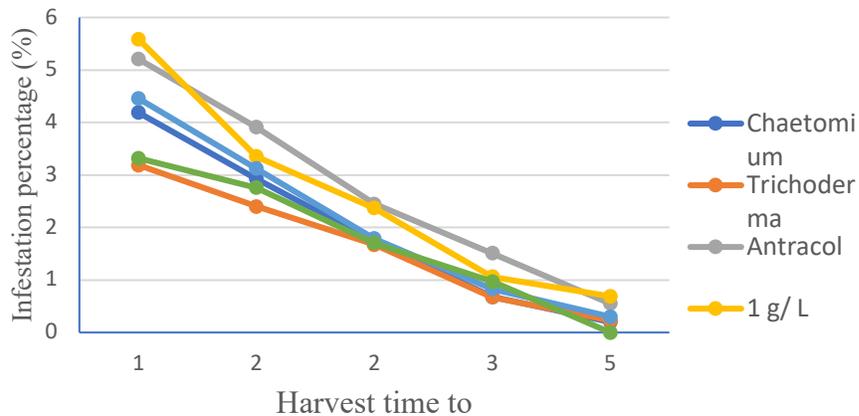
Note: (+) Antagonistic mechanism present, (-) Antagonistic mechanism absent

Both antagonistic fungi (*Chaetomium globosum* and *Trichoderma viride*) demonstrated competitive exclusion of *Colletotrichum acutatum* through space, nutrient, and oxygen competition, progressively limiting the pathogen's growth area. These macroscopic antagonistic interactions.

**Field evaluation of antagonistic fungi on disease severity and yield quality**

**Percentage of infestation**

Analysis of total attack percentage revealed a consistent reduction in disease levels from harvest 1 to harvest 5 (Figure 2) with application of antagonistic fungi treatments. Statistical analysis demonstrated a significant interaction ( $p < 0.05$ ) between fungal treatment type and concentration, with the *Trichoderma*-based treatment at 3 g/L concentration showing optimal efficacy in reducing total disease incidence (Table 2).



**Figure 2.** The percentage of *Colletotrichum acutatum*'s infestation as affected by fungicides

**Table 2.** Interaction effect of antagonistic fungi treatment and concentration on total infestation

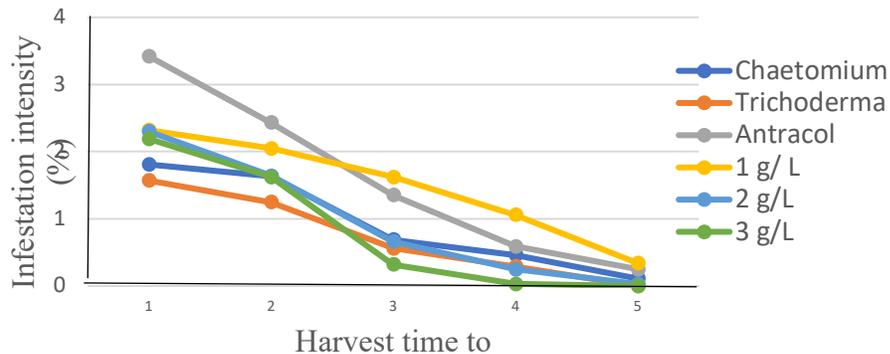
Treatments	Concentration		
	1 g/L	2 g/L	3 g/L
<i>Chaetomium globosum</i>	12,39 <sup>a</sup> B	8,93 <sup>b</sup> B	7,95 <sup>c</sup> B
<i>Trichoderma viride</i>	10,32 <sup>a</sup> C	8,84 <sup>b</sup> B	7,73 <sup>c</sup> B
Antracol	16,52 <sup>a</sup> A	13,81 <sup>b</sup> A	10,63 <sup>b</sup> A

Notes: Numbers followed by the same lowercase letter (horizontal) and the same capital letter (vertical) are not significantly different by at 5% significance level.

There was significantly interacted the effects between antagonistic fungi treatments and concentration levels on total infestation percentage (Table 2). Both antagonistic fungi (*Chaetomium globosum* and *Trichoderma viride*) showed a concentration-dependent reduction in infestation, with the lowest infestation rates observed at the highest concentration (3 g/L). Notably, *Trichoderma viride* at 3 g/L showed the best performance with only 7.73% infestation, followed closely by *Chaetomium globosum* at the same concentration 7.95%. All concentration levels within each fungicide treatment was significantly differed results which indicated by lowercase letters, with higher concentrations consistently yielding better control. When comparing across treatments at equivalent concentrations, both antagonistic fungi performed significantly better ( $p < 0.05$ ) than the chemical fungicide Antracol which indicated by capital letters, particularly at lower concentrations.

### Infestation intensity

Analysis of total disease severity revealed a consistent decline in infection levels from harvest 1 to harvest 5 (Figure 3) with the application of antagonistic fungicides. Statistical analysis demonstrated a significant interaction ( $p < 0.05$ ) between fungicide type and concentration, with the *Trichoderma viride* treatment at 3 g/L concentration showing optimal efficacy in reducing disease severity (Table 3).



**Figure 3.** The infestation intensity of *Colletotrichum acutatum* as affected by fungicides

There was significantly interacted ( $p < 0.05$ ) between fungicide type and concentration on infestation intensity, with both antagonistic fungi (*Chaetomium globosum* and *Trichoderma viride*) outperforming the chemical fungicide Antracol at all concentrations (Table 3). *Trichoderma viride* at 3 g/L showed the strongest suppression.

**Table 3.** Interaction effect of antagonistic fungicide treatment and concentration on infestation intensity

Treatments	Concentration		
	1 g/L	2 g/L	3 g/L
<i>Chaetomium globosum</i>	7,24 <sup>a</sup> B	3,17 <sup>b</sup> B	3,73 <sup>b</sup> B
<i>Trichoderma viride</i>	4,88 <sup>a</sup> C	3,62 <sup>b</sup> B	2,54 <sup>c</sup> C
Antracol	10,05 <sup>a</sup> A	7,86 <sup>b</sup> A	6,20 <sup>c</sup> A
-			

Notes: Numbers followed by the same lowercase letter (horizontal) and the same capital letter (vertical) are not significantly different by at 5% significance level.

### *Salicylic acid*

There was significantly interacted between control agent type and concentration on salicylic acid (SA) induction in anthracnose-infected chili plants (Table 4). While increasing concentration to 3 g/L universally elevated SA levels, the antagonistic fungi *Trichoderma viride* and *Chaetomium globosum* was significantly differed in efficacy at 1 g/L, yet both outperformed the chemical fungicide Antracol at this low dose, confirming their role as potent plant defense elicitors even at minimal rates.

**Table 4.** Concentration interaction on salicylic acid content in chili plants

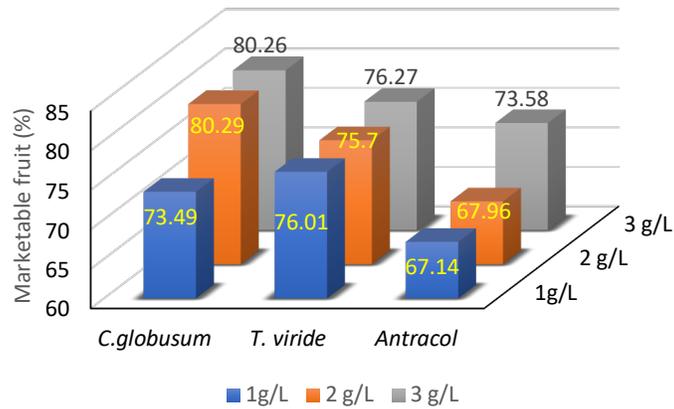
Treatments	Concentration		
	1 g/L	1 g/L	1 g/L
<i>Chaetomium globosum</i>	34,69 <sup>c</sup> A	35,16 <sup>b</sup> B	37,71 <sup>a</sup> B
<i>Trichoderma viride</i>	34,38 <sup>c</sup> A	36,71 <sup>b</sup> A	51,32 <sup>a</sup> A
Antracol	30,10 <sup>c</sup> B	31,91 <sup>b</sup> C	36,07 <sup>a</sup> C

Notes: Numbers followed by the same lowercase letter (horizontal) and the same capital letter (vertical) are not significantly different by at 5% significance level.

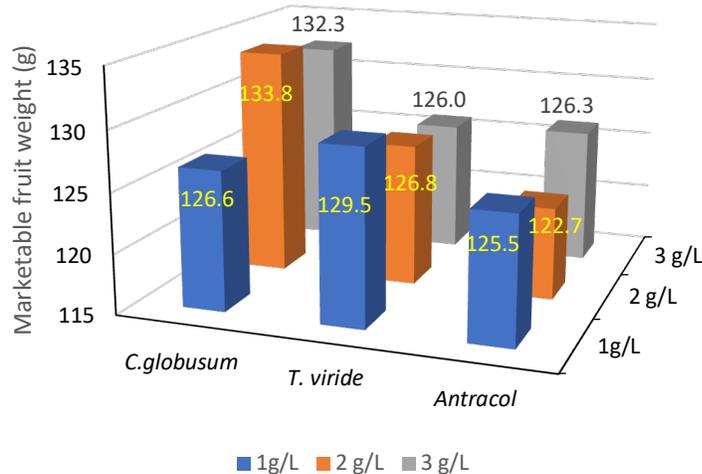
### *Marketable fruit*

The comparative effects of antagonistic fungi and concentration on marketable fruit yield was significantly ( $p < 0.05$ ) differed than the control. *Chaetomium globosum* at 2 g/L produced the highest fruit yield, outperforming both *Trichoderma viride* and Antracol at equivalent concentrations. Notably, *Chaetomium globosum* maintained consistent yield enhancement across all

concentrations, while *T. viride* and Antracol reduced efficacy at lower doses (Figure 4).



**Figure 4.** Interaction effect of antagonistic fungi and concentration on marketable fruit yield



**Figure 5.** Interaction effect of antagonistic fungi and concentration on marketable fruit weight

The comparative efficacy of *Chaetomium globosum*, *Trichoderma viride*, and Antracol at varying concentrations (1 g/L, 2 g/L, 3 g/L) on harvestable fruit weight, with *Chaetomium globosum* exhibited a peak performance at higher concentrations (2 g/L and 3 g/L) (Figure 5). In contrast, *Trichoderma viride* achieved optimal results at 1 g/L, with reduced efficacy at higher concentrations.

Antracol displayed a non-linear response, where 3 g/L matched 1 g/L but underperformed at 2 g/L.

## Discussion

The pathogen isolated from symptomatic chili plants was identified as *Colletotrichum acutatum*, similar to the established morphological descriptions (Barnett, 1960; De Silva *et al.*, 2017; Dawit *et al.*, 2025; Wakhidah *et al.*, 2021). This species is particularly prevalent in humid, mid- to high-altitude environments, where delayed intervention can result in severe yield losses and drive overreliance on chemical fungicides.

Both *Chaetomium globosum* and *Trichoderma viride* significantly suppressed *Colletotrichum acutatum*, primarily through competitive exclusion for space, nutrients, and oxygen (Mukarlina *et al.*, 2010). The absence of inhibition zones indicates that diffusible antibiosis was not the dominant mechanism under the experimental conditions (Mejia *et al.*, 2008). Instead, microscopic observations revealed hyphal coiling, entanglement, and abnormal growth of *Colletotrichum acutatum*, particularly in treatments involving *Trichoderma viride*, supporting a mycoparasitic mode of action (Sari, 2020).

Disease suppression was strongly concentration-dependent, with both antagonists achieving maximal efficacy at 3 g/L. This response aligns with reports that optimal metabolite production and enzymatic activity in fungal biocontrol agents occur at concentrations of 2.5–3 g/L (Zhang *et al.*, 2021). Importantly, both antagonistic fungi outperformed the chemical fungicide Antracol across all concentrations tested ( $p < 0.05$ ), corroborating earlier evidence of their superior control of *Colletotrichum* spp. (Aggarwal *et al.*, 2004; Batta, 2004). The robust performance of *Trichoderma viride* is consistent with its multimodal antagonism, including mycoparasitism, enzyme-mediated lysis, and host defense induction (Khan *et al.*, 2019). These findings reinforce the importance of dosage optimization in biological control and support current recommendations for reducing chemical fungicide dependence (FAO, 2023), particularly in the context of rising fungicide resistance (Yin *et al.*, 2023; EPPO, 2025).

In addition to disease suppression, antagonistic fungi significantly enhanced chili productivity. *Chaetomium globosum* performed optimally at higher concentrations (2–3 g/L), producing the greatest fruit weights, likely due to sustained competitive dominance and chitinase-mediated inhibition of pathogen hyphae (Chang *et al.*, 2003; Mukarlina *et al.*, 2010). In contrast, *Trichoderma viride* was most effective at lower concentrations (1 g/L), reflecting its reliance on parasitism and antifungal metabolites (Sari, 2020; Vinale *et al.*,

2008). Both biofungicides consistently outperformed Antracol, supporting earlier reports of their dual role as disease suppressors and plant growth promoters (Harman, 2006; Zhang *et al.*, 2021).

Patterns of infestation intensity further highlighted species-specific dose responses. *Trichoderma viride* at 3 g/L achieved the strongest suppression, coinciding with peak activity of lytic enzymes such as chitinase and  $\beta$ -1,3-glucanase (Hossain and Sultana, 2024). *Chaetomium globosum* exhibited stable efficacy at concentrations  $\geq 2$  g/L, consistent with its adaptability under nutrient-limited conditions (Mulaw *et al.*, 2019). By contrast, Antracol required substantially higher effective doses to achieve comparable suppression, in line with meta-analytical evidence showing higher efficacy thresholds for chemical fungicides against *Colletotrichum* spp. (Reglinski *et al.*, 2023).

Both antagonistic fungi also significantly increased salicylic acid (SA) levels in chili plants, confirming their role in activating systemic acquired resistance (SAR). The highest SA induction was observed with *Trichoderma viride* at 3 g/L, indicating its superior capacity to stimulate SA-dependent defense signaling and pathogenesis-related protein expression (Khan *et al.*, 2023A; Mishra *et al.*, 2024). Nevertheless, the comparable immune activation achieved at lower concentrations suggests practical flexibility for IPM systems aiming to reduce chemical inputs while maintaining strong disease control.

From an applied perspective, these results emphasize the need for precision in biocontrol deployment. The higher-dose efficacy of *Chaetomium globosum* and lower-dose efficiency of *Trichoderma viride* reflect fundamental differences in antagonistic strategy—competition versus parasitism—necessitating agent-specific application strategies based on pathogen pressure and field conditions. The inconsistent performance and potential phytotoxicity observed with Antracol further underscore the limitations of chemical fungicides and the advantages of biologically based alternatives.

In conclusion, *Trichoderma viride* and *Chaetomium globosum* are effective, environmentally sustainable biocontrol agents for managing chili anthracnose caused by *Colletotrichum acutatum*. Both fungi suppressed pathogen development more effectively than the synthetic fungicide Antracol, with maximal control achieved at 3 g/L. At this concentration, *Trichoderma viride* reduced disease incidence and attack intensity by 92.27% and 97.46%, respectively, while *Chaetomium globosum* achieved reductions of 92.65% and 96.83%. In addition to direct pathogen suppression, both antagonists enhanced salicylic acid accumulation, indicating activation of host systemic resistance. Collectively, these findings position *Trichoderma viride* and *Chaetomium globosum* as potent, residue-free alternatives to chemical fungicides and support their integration into sustainable, resistance-resilient disease management

strategies for chili production. It is needed to develop these effective species to be biofungicide and to evaluate in the field trials.

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### **Conflicts of interest**

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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