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In Vitro Antagonistic Activity of *Trichoderma atroviride* and *Trichoderma harzianum* against *Colletotrichum* sp. and *Fusarium* sp. Isolated from Chili (*Capsicum annuum* L.)

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ABSTRACT

Fungal diseases remain a major constraint in chili cultivation, particularly anthracnose and Fusarium wilt. This study aimed to evaluate the in vitro biocontrol potential of *Trichoderma atroviride* and *Trichoderma harzianum* against *Colletotrichum* sp. and *Fusarium oxysporum* f. sp. *capsici*. The study was conducted experimentally using the dual culture method on Potato Dextrose Agar (PDA) medium with three replications for each treatment. Results showed that both *Trichoderma* isolates exhibited strong inhibitory activity against the tested pathogens. *Trichoderma atroviride* showed inhibition percentages of 92% against *Colletotrichum* sp. and 90% against *Fusarium* sp., while *Trichoderma harzianum* inhibited pathogen growth by 73.6% and 85%, respectively. The primary antagonistic mechanism observed was competition for space and nutrients, indicated by the rapid growth of *Trichoderma* colonies covering the Petri dishes. This study indicates that *T. atroviride* and *T. harzianum* have promising in vitro biocontrol potential against *Colletotrichum* sp. and *Fusarium oxysporum* f. sp. *capsici*.

Keywords: Antagonistic, Biocontrol, Chili, Colletotrichum, Trichoderma

ABSTRAK

Penyakit yang disebabkan oleh jamur masih menjadi kendala utama dalam budidaya cabai, terutama antraknosa dan layu fusarium. Penelitian ini bertujuan untuk mengevaluasi potensi pengendalian hayati secara in vitro dari Trichoderma atroviride dan Trichoderma harzianum terhadap Colletotrichum sp. dan Fusarium oxysporum f. sp. capsici. Penelitian dilakukan secara eksperimental menggunakan metode kultur ganda (dual culture) pada media Potato Dextrose Agar (PDA) dengan tiga ulangan untuk setiap perlakuan. Hasil penelitian menunjukkan bahwa kedua isolat Trichoderma memiliki aktivitas penghambatan yang tinggi terhadap patogen yang diuji. Trichoderma atroviride menunjukkan persentase penghambatan sebesar 92% terhadap Colletotrichum sp. dan 90% terhadap Fusarium sp., sedangkan Trichoderma harzianum mampu menghambat pertumbuhan Colletotrichum sp. dan Fusarium sp. masing-masing sebesar 73,6% dan 85%. Mekanisme antagonisme utama yang diamati adalah kompetisi ruang dan nutrisi, yang ditunjukkan oleh pertumbuhan koloni Trichoderma yang cepat hingga menutupi cawan petri. Penelitian ini menunjukkan bahwa T. atroviride dan T. harzianum memiliki potensi pengendalian hayati secara in vitro yang menjanjikan terhadap Colletotrichum sp. dan Fusarium oxysporum f. sp. capsici.

Kata kunci: Antagonistik, Biokontrol, Cabai, Colletotrichum, Trichoderma

INTRODUCTION

Fungal diseases remain a major constraint in chili cultivation, particularly anthracnose caused by *Colletotrichum* spp. and Fusarium wilt caused by *Fusarium oxysporum* f. sp. *capsici*. Anthracnose leads to fruit rot and reduced yield quality, whereas Fusarium wilt is a soil-borne disease that infects

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vascular tissues and is difficult to control (Dailah et al., 2021). Conventional management of these diseases often relies on chemical fungicides, which may contribute to environmental pollution, pathogen resistance, and potential risks to human health. Therefore, environmentally friendly and sustainable disease management strategies are needed.

As an environmentally friendly alternative, biological control using antagonistic microorganisms has received considerable attention. One of the fungal genera widely studied as a biocontrol agent is *Trichoderma* (Jibril, 2016). Species of *Trichoderma* are known for their antagonistic activity against various fungal pathogens through several mechanisms, including competition for space and nutrients, production of antimicrobial metabolites, and mycoparasitism. These mechanisms have been reported to effectively suppress the growth of plant pathogenic fungi in both in vitro and in vivo studies (Yassin et al., 2021).

Several studies have demonstrated the effectiveness of *Trichoderma* species against important plant pathogens. For example, *Trichoderma harzianum* exhibited antagonistic activity against *Colletotrichum* sp. through effective competition and suppression of pathogen growth (Sanothan et al., 2023). Likewise, Alamsjah et al. (2023) reported that antagonistic microorganisms, including *Trichoderma* spp., have the potential to inhibit the growth of plant pathogens under laboratory conditions, although the effectiveness and mechanisms may vary depending on the isolate and target pathogen.

Despite the growing number of studies on the biocontrol potential of *Trichoderma* spp., most investigations have focused on a single *Trichoderma* species or a single target pathogen. Comparative information regarding the antagonistic performance of *Trichoderma atroviride* and *Trichoderma harzianum* against both *Colletotrichum* sp. and *Fusarium oxysporum* f. sp. *capsici* remains limited. Differences in antagonistic mechanisms among *Trichoderma* species may result in varying levels of effectiveness against specific pathogens. Therefore, a direct comparison of these two *Trichoderma* species under the same experimental conditions is necessary to better understand their relative biocontrol potential. This study provides comparative information on the in vitro antagonistic activity of *T. atroviride* and *T. harzianum* against two major fungal pathogens of chili.

Therefore, this study aimed to evaluate and compare the in vitro biocontrol potential of *Trichoderma atroviride* and *Trichoderma harzianum* against *Colletotrichum* sp., the causal agent of anthracnose, and *Fusarium oxysporum* f. sp. *capsici*, the causal agent of Fusarium wilt in chili.

MATERIALS AND METHODS

This study was conducted experimentally under laboratory conditions to evaluate the antagonistic activity of *Trichoderma atroviride* and *Trichoderma harzianum* against *Colletotrichum* sp. and *Fusarium* sp. using the dual culture method. The experiment consisted of three replications for each treatment. The treatments included dual cultures of *T. atroviride* with each pathogen and *T. harzianum* with each pathogen, while pathogen cultures grown alone on PDA served as the control treatment. The equipment used included Petri dishes, a laminar air flow cabinet, an autoclave, and an incubator. The materials consisted of *Trichoderma* isolates, *Colletotrichum* sp., *Fusarium* sp., and Potato Dextrose Agar (PDA) medium. Antagonistic activity was evaluated based on the inhibition of

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pathogen growth compared with the control treatment. Data were presented descriptively as inhibition percentages obtained from the three replications.

Preparation of Culture Medium

Potato Dextrose Agar (PDA) medium was prepared by dissolving 39 g of ready-to-use PDA powder in 1 L of distilled water, followed by heating and homogenization. The medium was then sterilized in an autoclave at 121°C under a pressure of 2 atm for 20 minutes.

Fungal Reculture

The reculture of *Trichoderma*, *Colletotrichum*, and *Fusarium* isolates was performed in test tubes by transferring a portion of fungal hyphae previously grown on PDA medium in Petri dishes onto sterile PDA medium in test tubes. The inoculated cultures were then incubated at room temperature (25–27°C) for 7 days until abundant mycelial growth was observed.

Antagonistic Activity Assay

The antagonistic activity of *Trichoderma atroviride* and *Trichoderma harzianum* against the pathogenic fungi *Colletotrichum* sp. and *Fusarium* sp. was evaluated using the dual culture method.

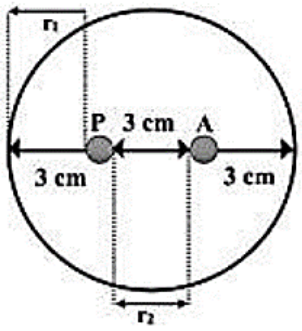

Figure 1. Schematic Arrangement of Antagonistic and Pathogenic Fungi Using the Dual Culture Method.

Description:
A = Antagonistic fungus (*Trichoderma*)
P = Pathogenic fungus (*Colletotrichum* and *Fusarium*)

Measurement of Inhibition Percentage

The percentage of inhibition (IP) was determined by measuring the radius of the pathogenic fungal colony growing toward and away from the antagonistic fungal colony. According to Ningsih *et al*. (2016), the inhibition percentage (IP) was calculated using the following formula:

$$IP(\%) = \frac{R_1 - R_2}{R_1} \times 100$$

Description:
IP = Inhibition Percentage (%)
R_1 = Diameter of pathogen growth in the control (mm)
R_2 = Diameter of pathogen growth in each treatment (mm)

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RESULTS AND DISCUSSION

Percentage of Inhibition of *Trichoderma* Against *Colletotrichum* and *Fusarium*

Based on the results of the antagonistic assay presented in Table 1, both *Trichoderma* isolates exhibited strong inhibitory activity against the tested pathogens. *Trichoderma atroviride* showed the highest antagonistic activity, with inhibition percentages of 92% against *Colletotrichum* sp. and 90% against *Fusarium* sp. Meanwhile, *Trichoderma harzianum* was also able to suppress the growth of both pathogens, with inhibition percentages of 73.6% against *Colletotrichum* sp. and 85% against *Fusarium* sp. These results indicate that *T. atroviride* has greater antagonistic potential compared to *T. harzianum*, particularly against *Colletotrichum* sp.

Table 1. Percentage of Inhibition of Antagonistic Activity

Antagonistic fungi	Pathogen	Inhibition %
Trichoderma atroviride	*Colletotrichum* sp.	92%
	Fusarium sp.	90%
Trichoderma harzianum	*Colletotrichum* sp.	73.6%
	Fusarium sp.	85%

The antagonistic activity of both *Trichoderma* isolates against the tested pathogens was found to be high. *Trichoderma atroviride* exhibited the strongest antagonistic effect, with inhibition percentages of 92% against *Colletotrichum* sp. and 90% against *Fusarium* sp. Meanwhile, *Trichoderma harzianum* also suppressed the growth of both pathogens, with inhibition percentages of 73.6% against *Colletotrichum* sp. and 85% against *Fusarium* sp. These results suggest that *T. atroviride* possesses greater antagonistic potential than *T. harzianum*, particularly against *Colletotrichum* sp.

This observation aligns with the findings of Dailah *et al*. (2020), who reported that *Trichoderma harzianum* can significantly inhibit the growth of *Colletotrichum* sp. in chili plants through antagonistic assays, as indicated by the formation of inhibition zones and the dominance of *Trichoderma* colony growth over the pathogen. Furthermore, Pasalo *et al*. (2022) noted that *Trichoderma* sp. showed optimal inhibition against *Fusarium* sp. starting from the second day after inoculation, with inhibition reaching 50%.

Antagonistic Mechanisms of *Trichoderma*

Based on the results of the antagonistic activity shown in Figures 2 and 3, *Trichoderma atroviride* demonstrated strong antagonistic ability against *Colletotrichum* sp. and *Fusarium* sp., as indicated by its rapid colony growth, nearly covering the entire Petri dish by the second day of incubation.

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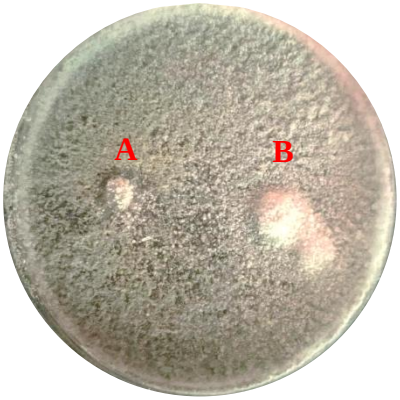

Figure 2. Antagonistic Activity of *Trichoderma atroviride* Against *Colletotrichum* sp.

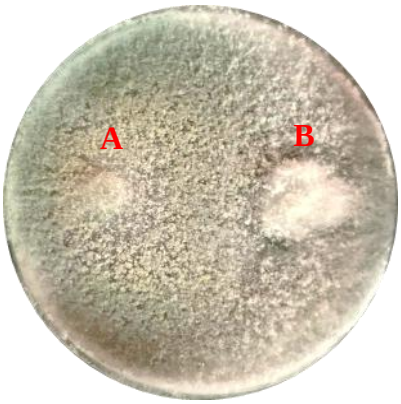

Figure 3. Antagonistic Activity of *Trichoderma atroviride* Against *Fusarium* sp.

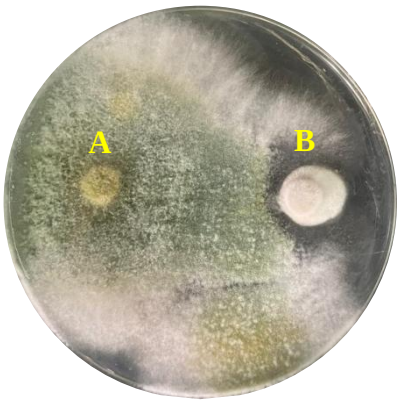

Figure 4. Antagonistic Activity of *Trichoderma harzianum* Against *Colletotrichum* sp.

Figure 5. Antagonistic Activity of *Trichoderma harzianum* Against *Fusarium* sp.

Description : A : Antagonistic fungus (*Trichoderma*)
B : Pathogenic fungus

Based on the antagonistic activity observed in Figures 2 and 3, *Trichoderma atroviride* demonstrated strong antagonistic potential against *Colletotrichum* sp. and *Fusarium* sp., as indicated by its rapid colony growth, nearly covering the entire Petri dish by the second day of incubation. This fast growth limits the space and nutrient availability for the pathogens, suggesting competition as a primary antagonistic mechanism. Gomez *et al*. (2018) reported that *T. atroviride* employs mycoparasitism, hydrolytic enzyme production, and antifungal metabolites as part of its antagonistic strategy against pathogenic fungi, supporting the notion that direct interactions between *Trichoderma* hyphae and the pathogens contribute to pathogen growth inhibition.

As a biological control agent, *T. atroviride* has been widely applied in plant disease management through seed treatment, root immersion, and application to planting media. This fungus is effective in suppressing various important diseases such as anthracnose (*Colletotrichum* spp.), Fusarium wilt (*Fusarium oxysporum*), and root rot that commonly affect horticultural crops. Its

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effectiveness is attributed to its ability to produce antifungal secondary metabolites, enhance nutrient availability, and stimulate plant growth (Kim *et al*., 2023)

Meanwhile, as shown in Figures 4 and 5, *Trichoderma harzianum* also suppressed the growth of *Colletotrichum* sp. and *Fusarium* sp., but at a slower rate compared to *T. atroviride*, with colonies covering the Petri dish only by the third day of incubation. The direct interaction between *Trichoderma* and the pathogens indicates mycoparasitic activity and further supports the observation that *T. atroviride* exhibits higher antagonistic effectiveness than *T. harzianum*.

Trichoderma spp. are known to employ diverse antagonistic mechanisms against fungal pathogens, including competition for space and nutrients, antibiosis via secondary metabolites, and mycoparasitism of pathogen hyphae (Chen *et al*., 2025). The classification of antagonistic mechanisms can be inferred from the growth rate of the fungus in occupying the Petri dish surface. If an antagonistic fungus grows rapidly to fill the Petri dish, it indicates competition as the dominant mechanism. This form of antagonism arises from the competition among microorganisms for limited nutrients and space (Cendrawati *et al*., 2020). Based on the study conducted by Puspita *et al*. (2026), *Trichoderma* sp. exhibited the highest inhibitory activity with an inhibition percentage of 92.78%, which was categorized as very high.

Characteristics of the Pathogens *Colletotrichum* sp. and *Fusarium* sp.

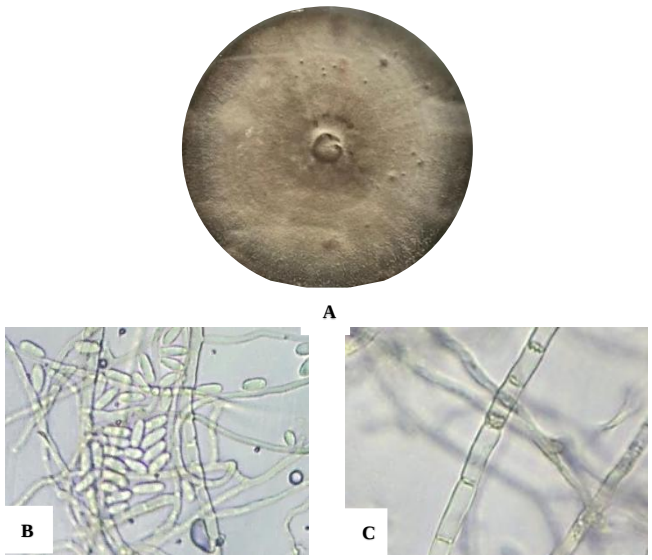

Figure 6. Macroscopic and Microscopic Characteristics of *Colletotrichum* spp. (40× Magnification)
Note: (a) Front view, (b) Conidia, (c) Septate hyphae.

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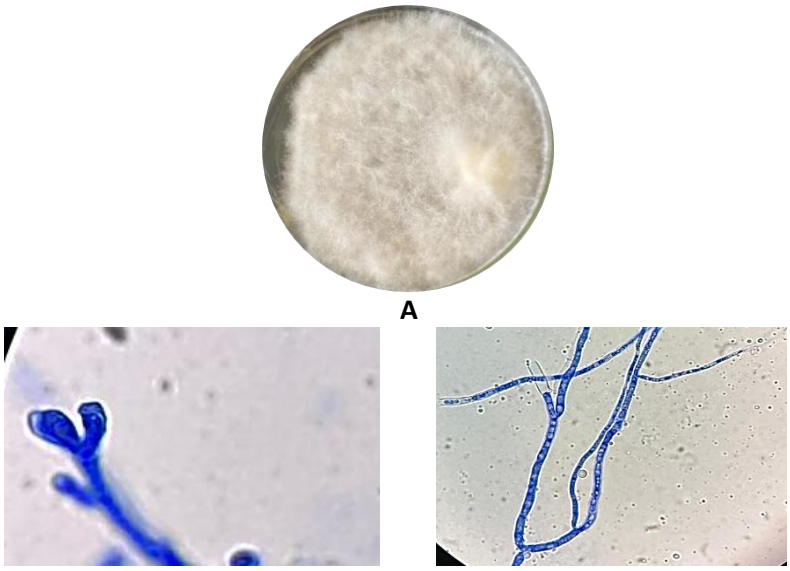

Figure 7. Macroscopic and Microscopic Characteristics of *Fusarium* spp. (40× Magnification)
Note: (a) Front view, (b) Conidiophore (c) Septate hyphae

Based on Figure 6, the results showed that the *Colletotrichum* isolate exhibited typical morphological characteristics, with colonies appearing whitish-gray and a darker central area. The colony texture was smooth, showing radial growth with even margins and a denser central zone. In the early stages of growth, the conidia appeared orange in color. Microscopically, the isolate showed septate hyphae with smooth and branched structures. The conidiophores appeared clustered, with terminal ends producing chains of conidia. The conidia were oval to fusiform in shape, small in size, and evenly distributed along the conidiophores.

According to Alvarez *et al*. (2020), the conidial mass of *Colletotrichum* sp. is orange in color and is formed from single conidia after 10 days of incubation, spreading or forming concentric rings on the colony surface. The conidial mass represents the asexual fruiting body tissue of *Colletotrichum* spp., consisting of a cluster of conidiophores where conidia are produced (Moore *et al*., 2020). The conidial mass or conidiomata of *Colletotrichum* spp. is characterized as acervular (Kumari, 2017). *Colletotrichum* exhibits colony morphology with a whitish-gray upper surface and a dark brown to black coloration on the reverse side of the colony.

Based on Figure 7, macroscopically (a) the colony of *Fusarium* spp. exhibits radial growth with white to grayish-white mycelia and a smooth to slightly cottony texture. The central area of the colony appears denser than the margins, indicating active growth. Microscopically at 40× magnification, branched conidiophores (b) are observed, with terminal ends serving as sites of conidia formation. In addition, elongated and branched septate hyphae (c) are visible, which are characteristic of the Ascomycota group. The hyphae appear hyaline with clearly defined septa, representing common morphological features of *Fusarium* spp.

Fusarium spp. colonies grown on artificial media such as Potato Dextrose Agar (PDA) typically show rapid radial growth with whitish mycelia that may develop pink, orange, or reddish pigmentation depending on the isolate and environmental conditions; colony color and texture can also be influenced by incubation temperature and medium composition (Cavusoglu *et al*., 2025).

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Microscopically, *Fusarium* produces macroconidia and microconidia from slender phialides; macroconidia are hyaline, fusiform to sickle-shaped with multiple septa, while microconidia are smaller and variable in shape. Chlamydospores may also form under certain conditions, serving as important diagnostic features in morphological identification (Ekwomadu & Mwanza, 2023).

CONCLUSION

This study demonstrated that *Trichoderma atroviride* and *Trichoderma harzianum* possess promising in vitro biocontrol potential against *Colletotrichum* sp. and *Fusarium oxysporum* f. sp. *capsici*. Among the tested isolates, *T. atroviride* showed higher inhibition activity than *T. harzianum*, particularly against *Colletotrichum* sp., with an inhibition percentage of 92%. The rapid growth of both antagonistic fungi suggests that competition for space and nutrients plays an important role in suppressing pathogen development. These findings provide preliminary evidence supporting the potential use of *Trichoderma* spp. as environmentally friendly alternatives for disease management in chili cultivation. Further greenhouse and field evaluations are needed to validate their effectiveness under practical growing conditions.

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REFERENCES

Alamsjah, F., Noli, Z. A., Marcellinna, H. D., Agustien, A., & Ilham, K. (2023). Antagonist Test of Bacillus subtilis ATTC 6633 and *Trichoderma harzianum* on the Growth of *Magnaphorte oryzae* on Several Varieties of Priming Rice Seeds. *Jurnal Biologi Tropis*, 23(1), 371-379.

Alvarez, L. V., Hattori, Y., Deocaris, C. C., Mapanao, C. P., Bautista, A. B., Cano, M. J. B., Naito, K., Kitabata, S., Motohashi, K., & Nakashima, C. (2020). *Colletotrichum asianum* causes anthracnose in Philippine mango cv. Carabao. *Australasian Plant Disease Notes*, 15(1), 1–5.

Çavusoglu, A., Ünal, F., & ÖZER UYAR, G. Ü. L. S. Ü. M. (2025). Comparative Study of Morphological Characteristics of Phytopathogenic Fungi Fusarium oxysporum from Allium sativum at Two Different Temperatures. *Journal of Advances in Microbiology*, *25*(10).

Cendrawati, M. A., Suwandi, S., Herlinda, S., & Suparman, S. (2020). Potensi Jamur Asal Umbi Tanaman Terna Tahunan Sebagai Pengendali Ganoderma boninense Penyebab Penyakit Busuk Pangkal Batang Pada Kelapa Sawit. Jurnal Biotek, 8(2), 178-188.

Chen, X., Lu, Y., Liu, X., Gu, Y., & Li, F. (2025). Trichoderma: Dual Roles in Biocontrol and Plant Growth Promotion. *Microorganisms*, 13(8), 1840.

Dailah, S., Poerwanto, M. E., & Sutoto, S. B. (2021). Effectiveness of Antagonistic Fungi *Gliocladium* sp. and *Trichoderma Harzianum* to Control *Colletotrichum* Spp. on Chili (*Capsicum Annuum* L.). *Agrivet*, *26*(1), 17-22.

Article History
Received : 13-05-2025
Revised : 26-05-2026
Accepted : 23-06-2026

Ekwomadu, T. I., & Mwanza, M. (2023). Fusarium fungi pathogens, identification, adverse effects, disease management, and global food security: a review of the latest research. *Agriculture 13 (9): 1810*.

Gómez-Rodríguez, E. Y., Uresti-Rivera, E. E., Patrón-Soberano, O. A., Islas-Osuna, M. A., Flores-Martínez, A., Riego-Ruiz, L., ... & Casas-Flores, S. (2018). Histone acetyltransferase TGF-1 regulates Trichoderma atroviride secondary metabolism and mycoparasitism. PLoS One, 13(4), e0193872.

Jibril, S. M., Jakada, B. H., Kutama, A. S., and Umar, H. Y., 2016. Plant and Patogens: Patogen Recognition, Invasion and Plant Defense Mechanism. International Journal of Current Microbiology and Applied Sciences. 5(6): 247-257.

Kim, S. H., Lee, Y., Balaraju, K., & Jeon, Y. (2023). Evaluation of *Trichoderma* atroviride and *Trichoderma longibrachiatum* as biocontrol agents in controlling red pepper anthracnose in Korea. Frontiers in plant science, 14, 1201875

Kumari, P. (2017). Anthracnose of Mango Incited by Colletotrichum gloeosporioides : A Comprehensive Review. *International Journal of Pure & Applied Bioscience*, 5(1), 48–56.

Moore, D., Robson, G. D., & Trinci, A. P. J. (2020). 21st Century Guidebook to Fungi. 21st Century Guidebook to Fungi, June.

Ningsih, H., Hastuti, U.S., dan Listyorini, D. 2016. Kajian Antagonis Trichoderma spp. terhadap Fusarium solani Penyebab Penyakit Layu Pada Daun Cabai Rawit (Capsicum frutescens) Secara in Vitro. *Proceeding Biology Education Conference*, Vol 13(1) 2016: 814-817.

Pasalo, N. M., Kandou, F. E. F., & Singkoh, M. F. O. (2022). Uji Antagonisme jamur Trichoderma sp. terhadap patogen Fusarium sp. pada tanaman bawang merah Allium cepa isolat lokal Tonsewer secara in vitro. *Jurnal Ilmu Alam dan Lingkungan*, *13*(2).

Puspita, A. R., Alamsjah, F., Mildawati, M., & Agustien, A. (2026). Antagonistic activity of phyllosphere fungi isolated from medicinal plants against *Colletotrichum* sp. causing anthracnose in chilli (*Capsicum annuum* L.). *Bioeksperimen: Jurnal Penelitian Biologi*, 174-182.

Sanothan, A., Montong, V. B., & Lengkong, M. (2023). Uji antagonis jamur *Trichoderma* sp. terhadap penyakit antraknosa *Colletotrichum* sp. pada tanaman cabai keriting *Capsicum annuum* L. di Laboratorium. *Jurnal Enfit: Entomologi dan Fitopatologi*, 3(1), 15-23.

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