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In Vitro Antagonistic and Fungicidal Activity against *Agroathelia rolfsii* (syn. *Sclerotium rolfsii*), the Causal Pathogen of Stem Base Rot Disease in Brassica Plants

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Abstract

Plants of the Brassica genus, such as cabbage, cauliflower, and broccoli, are high-value horticultural commodities that are susceptible to collar rot caused by the fungus Agroathelia rolfsii (syn. Sclerotium rolfsii). This soil-borne pathogen is difficult to control due to its ability to form sclerotia that can survive for long periods in the soil, thus requiring more effective and environmentally friendly alternative control strategies. This study aimed to evaluate the antagonistic activity of several Trichoderma spp. isolates and the effectiveness of two commercial fungicides, chlorothalonil and pencycuron, against A. rolfsii in vitro. The antagonism test was conducted using the double culture method, while the fungicide test used the toxic agar technique. The results of the study showed that Trichoderma sp. isolate T01 showed the highest antagonistic activity with a PGI value of 62.05%. However, this value was not significantly different from Trichoderma sp. isolate T2 (59.87%) and Trichoderma asperellum MHR1 strain (59.09%) based on the DMRT test. Meanwhile, among the fungicides tested, chlorothalonil at a concentration of 100 mg/L provided the highest inhibition of 21.98% and 10 mg/L (14.60%). These findings indicate that Trichoderma sp. isolate T01 has the potential to be used as an effective biological agent, while chlorothalonil can play a role as a supporting fungicide in the application of Integrated Pest Management (IPM) strategies. Further research is recommended to evaluate the effectiveness of both agents under greenhouse and field conditions to ensure consistency in their performance.

Keywords: *Agroathelia rolfsii, antagonistic activity, basal stem rot, brassica plants, fungicidal activity.*

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INTRODUCTION

Horticultural crops from the Brassica genus, such as cabbage, cauliflower, and broccoli, are important commodities that play a role in meeting food and nutritional needs, as well as national market demand (Handayani et al., 2022). The increasing demand and strategic economic value make the productivity of Brassica plants an important factor in the sustainability of horticultural agribusiness in Indonesia. Therefore, efforts to maintain production stability are very necessary.

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The sustainability of Brassica crop production is often hampered by plant pests, particularly soil-borne diseases. One of the most damaging diseases is stem rot caused by the fungus *Agroathelia rolfsii* (syn. *Sclerotium rolfsii*). This pathogen is aggressive, capable of inhibiting growth, reducing yield quality, and causing significant yield losses, especially during the early vegetative phase (You et al., 2021).

The pathogen *A. rolfsii* has a high survival rate through the formation of sclerotia that can survive in the soil for years (Paul et al., 2017). In addition, this pathogen is polyphagous and capable of attacking various types of plants in tropical and subtropical regions (Kamel et al., 2020). Indonesia's warm and humid climate further supports the development of pathogens, there by increasing the risk of disease outbreaks.

Control of stem base rot disease has so far been dominated by the use of synthetic fungicides. Although effective, the continuous use of fungicides can have negative impacts, such as pathogen resistance, environmental pollution, soil degradation, and residues in agricultural products (Kumari et al., 2024). This situation necessitates the development of more environmentally friendly and sustainable control strategies.

Within the framework of Integrated Pest Management, the use of biological agents such as *Trichoderma* spp. is a potential alternative. This fungus is able to suppress pathogens through mechanisms of competition, antibiosis, mycoparasitism, and induction of plant resistance (Cui et al., 2019; (Wang et al., 2024). However, the effectiveness of Trichoderma is greatly influenced by isolate differences and environmental conditions, so in vitro testing is required to determine the most effective isolate.

In addition to biological agents, fungicides such as chlorothalonil and pencycuron are still needed in certain conditions. In vitro evaluation of Trichoderma isolates and commercial fungicides is important to determine their antagonistic and fungicidal capabilities against *A. rolfsii.* Therefore, this study aims to evaluate the in vitro activity of several Trichoderma isolates and fungicides in inhibiting the growth of *A. rolfsii* as a basis for developing effective and sustainable control strategies for stem base rot disease in Brassica plants.

MATERIALS AND METHODS

Place and Time

This study was conducted at the Microbial Taxonomy Laboratory, Horticulture Research Centre (HR) Building, Malaysian Agricultural Research and Development Institute (MARDI), Serdang, Selangor, Malaysia, from 21 September 2025 to 10 October 2025.

Materials, Tools and Composition

This study use various material biological and chemical requirements for perform antagonistic tests and effectiveness testing fungicide to *Agroathelia rolfsii*. The main ingredients used in the form of isolated *A. rolfsii* culture from plant cabbage China (*Brassica rapa* L. ssp. *pekinensis*) which shows symptom rotten base stem. In addition, this study utilized five Trichoderma isolates obtained from the Plant Pathology Laboratory, Horticulture Research Center, MARDI Serdang.

Table 1. List of Trichoderma Isolates Used in the Study

Trichoderma Isolate	Plant	Location
Trichoderma sp. isolate T1	Cabbage	MARDI Serdang
Trichoderma sp. isolate T2	Durian	MARDI Serdang
Trichoderma sp. isolate T01	Orchid	MARDI Serdang
Trichoderma asperellum	Orchid	MARDI Serdang
Trichoderma ghanense	Chili	MARDI Serdang

For support growth microorganisms In this case, Potato Dextrose Agar (PDA) media was used as basic media, whereas distilled water sterile, 70% alcohol, and solution fungicide chlorothalonil (50% WP) and pencycuron (25% WP) were used as material supporters in medium preparation and treatment.

Fungicide commercially tested in study mixed to in PDA medium with concentration different. Chlorothalonil and pencycuron were each prepared in three concentration, namely 10, 50, and 100 mg/L. This composition is required to assess the effectiveness of fungicides at various dosage levels. In addition, 5 mm diameter agar discs were used as source inoculum both for Trichoderma and *A. rolfsii*.

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Various tool laboratory participate used for support all over series procedures, including sterile Petri dishes, tubes reaction, micropipette, tweezers, scalpel, and laminar air flow for ensure condition aseptic. Sterilization process done use autoclave, while culture incubation is carried out in an incubator temperature of 28 ± 2°C. Measurement growth colony done use term push or rulers, and documentation growth colony noted use camera. All data is then analyzed use device computer with help device R Statistical Software version 4.3.3.

Composition treatment in study this includes two series of tests. First, the antagonistic test or dual culture assay involving five isolates. Trichoderma was placed on PDA medium at a distance of 1.5 cm from center cup, while disc *A. rolfsii* placed six centimeters from colony Trichoderma. Every treatment repeated as much as three times, and control prepared only with grow *A. rolfsii* without existence Trichoderma. Second, effectiveness test fungicide with toxic agar method, in which the PDA medium has been mixed with each fungicide at the concentration that has been set inoculated with disc *A. rolfsii* which is placed in the section middle cup. Every treatment fungicide repeated as much as four times, and medium without fungicide used as control. The main parameters observed includes growth diameter colony pathogen as well as percentage inhibition calculated growth based on comparison with control.

Method

The method used in this study was an in vitro experimental method in the laboratory to evaluate the antagonistic activity of *Trichoderma* spp. isolates and the effectiveness of fungicides against the growth of *A. rolfsii*.

Isolation and Maintenance of Fungal Cultures

The pathogen isolate *A. rolfsii* was obtained from infected Chinese cabbage plants (*Brassica rapa* L. ssp. *pekinensis*) showing characteristic symptoms of collar rot. The pathogen culture was purified using the hyphal tip method and maintained on Potato Dextrose Agar (PDA) medium (DIFCO, USA). *Trichoderma* isolates were also maintained on PDA medium (Table 1). All cultures were incubated at 28 ± 2°C.

Testing the Antagonistic Activity of *Trichoderma* spp. against *A. rolfsii*

The inhibitory activity of Trichoderma isolates against the growth of *A. rolfsii* hyphae was evaluated using a dual culture assay (Islam et al., 2018). A 5 mm diameter agar disc containing mycelium from a 7–10-day-old Trichoderma culture was placed 1.5 cm from the centre of the PDA medium. Next, a 5 mm diameter agar disc containing pathogen mycelium was placed 6 cm from the Trichoderma colony in the same Petri dish. PDA dishes inoculated with pathogens only were used as controls. All treatments were incubated for 7 days at 28 ± 2°C. The experiment was conducted in triplicate. After the incubation period, the colony diameter (mm) was measured, and the percentage growth inhibition (PGI) was calculated using the following formula.

$$\mathrm{I}(\%) = \frac{\mathrm{C}-\mathrm{T}}{\mathrm{C}} \times 100\%$$

Explanation:
C = diameter of the pathogen colony without treatment (mm) (control); and
T = diameter of the pathogen colony in the treatment with antagonists (mm**).**

In Vitro Effectiveness Test of Fungicides against *A. rolfsii*

The efficacy of two commercial fungicides, chlorothalonil and pencycuron was tested using the poison agar technique in accordance with the method described by Hussien et al. (2022). The required concentrations of each fungicide were mixed into liquid PDA medium to achieve concentrations of 10, 50, and 100 mg/L. The medium containing the fungicides was then poured into Petri dishes. A 5 mm diameter agar disc containing *A. rolfsii* mycelium was placed in the centre of each dish. PDA dishes without fungicide were used as controls. All dishes were incubated at 28 ± 2°C until the colonies on the controls grew to cover the entire surface of the medium. The percentage of mycelium growth inhibition was calculated using the following formula.

$$\mathrm{I}(\%) = \frac{\mathrm{C}-\mathrm{T}}{\mathrm{C}} \times 100\%$$

Explanation:
C = diameter of the pathogen colony in the control dish (mm).
T = diameter of the pathogen colony in the PDA medium containing fungicide (mm).

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Table 2. List of Fungicides Used in the Study

Active Ingredients	Trade name	FRAC*	Chemical group	Mode of action
Chlorothalonil	Daconil 2787	M 05	Chloronitriles	Multisite contacts
Pencycuron	Monceren 25 WP	20	Phenylureas	Cytoskeleton and motor protein

*FRAC: Fungicide resistance classification

Statistical Analysis

All statistical analyses and graphing were performed using R Statistical Software (version 4.3.3; R Core Team, 2024) with the agricolae package. Data were analysed using Analysis of Variance (ANOVA), and comparisons of means were performed using Duncan's Multiple Range Test (DMRT) at a significance level of $p \leq 0.05$.

RESULTS AND DISCUSSION

Antagonistic Activity of Trichoderma against *Agroathelia rolfsii*

The results showed that all five Trichoderma isolates had significant antagonistic activity against *A. rolfsii* based on analysis of variance ($p \leq 0.05$). This indicates that all isolates were able to suppress pathogen growth, although the level of inhibition varied (Table 3). Of the five isolates tested, the *Trichoderma* sp. isolate T01 showed the highest antagonistic activity with a PGI value of 62.05% (Table 3). However, this value was not significantly different from *Trichoderma* sp. isolate T2 (59.87%) and *Trichoderma asperellum* MHR1 strain (59.09%) showed the highest inhibition rates after seven days of incubation. This relatively high inhibition percentage indicates that isolates T01, T2, and *Asperellum* MHR1 are capable of growing faster than the pathogen and actively suppressing the rate of colonization.

Table 3. The Results of Trichoderma Isolates Against *Agroathelia rolfsii* Using the Dual Culture Method

Trichoderma Isolate	PGI (%)
Trichoderma sp. isolate T1	53.85 ^{c}
Trichoderma sp. isolate T2	59.87 ^{ab}
Trichoderma sp. isolate T01	62.05 ^{a}
Trichoderma asperellum MHR1 strain	59.09 ^{ab}
Trichoderma ghanense MHR1 strain	52.54 ^{c}

* The score is the average of three tests.

** The same letters indicate no significant difference according to the DMRT test ($p < 0.05$).

Isolates *T. asperellum* strain MHR1 and *T. ghanense* strain MHR1 also showed fairly good effectiveness with PGI values of 59.09% and 52.54% respectively. These two isolates prove that Trichoderma species from different groups can also act as promising antagonistic agents. Meanwhile, isolates T1 and T2, although showing slightly lower inhibition capabilities compared to isolate T01, were still effective with inhibition values ranging from 53–60%. This indicates that variations between isolates within a genus can affect the effectiveness of antagonism, both in terms of growth ability, colonisation speed, and the inhibition mechanisms possessed by each isolate.

Based on the morphology of colonisation, the biological control mechanism demonstrated by isolate T01 is thought to involve antibiosis and competition for space and nutrients (Figure 1). However, over time, isolate T01 showed the ability to colonise and cover the *A. rolfsii* colony (Figure 2), indicating an antagonistic interaction between the two isolates. The biological control mechanism of Trichoderma against plant pathogens generally includes competition, antibiosis, antagonism, and mycoparasitism, as well as its ability to enhance plant growth and induce systemic plant resistance (Yao et al., 2023). The high activity of *Trichoderma* sp. T01 is likely related to a combination of competition, antibiosis, and mycoparasitism mechanisms, as described by Yao et al. (2023), who reported that Trichoderma is capable of producing lytic enzymes such as chitinase and glucanase that damage the cell walls of pathogens.

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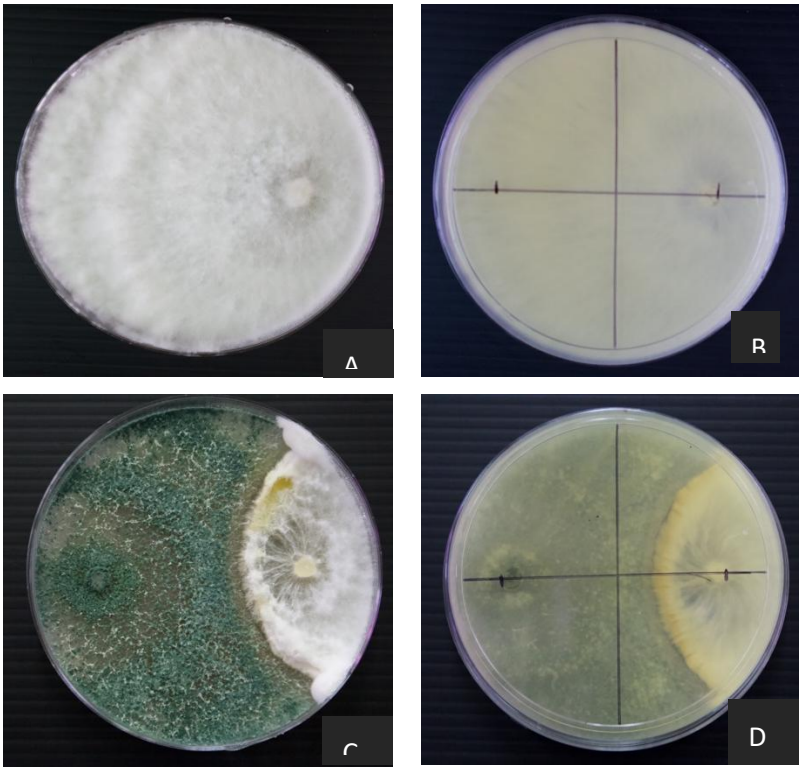

Figure 1. Dual culture assay of Trichoderma isolates against *Agroathelia rolfsii*. A) Control – top view, B) Control – bottom view, C) *Trichoderma* sp. isolate T01 – top view, D) *Trichoderma* sp. isolate T01 – bottom view.

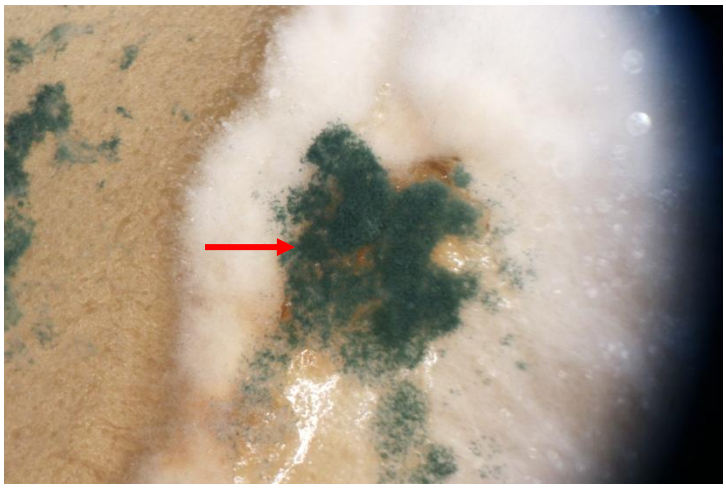

Figure 2. Interaction Between *Trichoderma* sp. Isolate T1 and *Agroathelia. rolfsii* on PDA Medium After Incubation for 9 days. The Arrows Indicate Areas of Trichoderma colonisation Covering the Growth of *A. rolfsii*, Indicating Antagonistic Activity Between the Two Isolates.

Dynamics of Growth and Patterns of Antagonism

Based on morphological observations of colonisation in dual cultures (Figure 1), it was observed that isolate T01 exhibited aggressive growth patterns with rapidly spreading mycelium covering most of the medium even before the pathogen reached the other side of the dish. In the early stages of interaction (days 3rd to 5th), this isolate exhibited a fairly clear zone of inhibition, namely the area around the Trichoderma colony boundary where the growth of *A. rolfsii* appeared to be halted or slowed down. This indication suggests the possible production of antibiosis compounds released by Trichoderma into the medium (Wells, 2023).

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As the incubation time increased, especially after days 7th to 9th, the T01 mycelium began to show the ability to colonise and coat the surface of the *A. rolfsii* colony (Figure 2). This behaviour is one of the characteristics of the mycoparasitism mechanism, in which Trichoderma utilises the pathogen as a source of nutrients. At this stage, changes in the colour and texture of the pathogen mycelium were observed, indicating enzymatic cell degradation. This is consistent with the report by Yao et al. (2023), which states that Trichoderma produces lytic enzymes such as chitinase, β-1,3-glucanase, and protease that are involved in the breakdown of the pathogen cell wall, causing lysis and death of the target cells.

Observations of other isolates show slightly different interaction patterns. For example, *T. asperellum* strain MHR1 produces a less thick colony edge but has a fairly clear antibiosis ability, as seen from the formation of a wider inhibition zone compared to other isolates. Meanwhile, *T. ghanense* strain MHR1 exhibited uniform radial growth patterns but was not as aggressive as T01. The inhibition produced appeared to be more influenced by competition for space and nutrients than by direct physical interaction with the pathogen.

Possible Mechanisms of Antagonism

The antagonistic mechanisms of Trichoderma against soil-borne pathogens have been widely reported and include several methods, namely competition for space and nutrients, antibiosis, mycoparasitism, and induction of plant resistance. Based on the results of this study, at least two main mechanisms can be observed directly, namely competition and antibiosis. This is evident from the pattern of opposing colony margins, where the growth of *A. rolfsii* stops as soon as it approaches the Trichoderma colony (Junaidi and Sarjan, 2025).

Mycoparasitism is also indicated by the penetration of Trichoderma colonies into pathogen colonies. At several contact points, especially in isolate T01, morphological changes in *A. rolfsii* mycelium were found, becoming thin, irregular, or showing a brownish colour. These conditions lead to enzymatic activity produced by Trichoderma, such as chitinase or glucanase enzymes that play a role in hydrolysing the main components of fungal cell walls (Ghasemi et al., 2020).

In addition to direct mechanisms, the possible release of volatile compounds or secondary metabolites may also explain the formation of inhibition zones without direct contact. This mechanism is common in several Trichoderma species, particularly the *T. harzianum*, *T. asperellum*, and *T. virens* groups. Metabolites such as gliotoxin, viridin, and trichodermin are known to effectively suppress pathogen growth.

Overall, the results of this study confirm that the isolates *Trichoderma* sp. T01, T2, and *Asperellum* MHR1 are suitable for development as biological control agents against *A. rolfsii* on Brassica crops. The combination of competition, antibiosis, and mycoparasitism provides the biological basis for the high level of inhibition observed.

Effectiveness of Fungicides on the Growth of *A. rolfsii*

In addition to antagonistic testing, this study also evaluated the effectiveness of two fungicides, chlorothalonil and pencycuron, using the toxic agar method. These fungicides were selected because they represent different mechanisms of action. Chlorothalonil is a contact fungicide with a multisite mechanism, while pencycuron is a systemic fungicide that specifically controls fungal diseases.

The results showed considerable variation in the inhibitory activity of the tested fungicides against the mycelial growth of *A. rolfsii*. Overall, both fungicides produced relatively low to moderate inhibition levels, ranging from 1.48% to 21.98% (Table 4). Chlorothalonil showed the highest inhibition at a concentration of 100 mg/L (21.98%) and 10 mg/L (14.60%), while lower inhibition was observed at 50 mg/L (11.48%). The non-linear response across concentrations suggests that fungal sensitivity to chlorothalonil may vary depending on the distribution and interaction of the active compound in the agar medium. Statistical analysis indicated that chlorothalonil at 100 mg/L produced significantly higher inhibition compared with most other treatments (Table 4).

Meanwhile, pencycuron showed lower inhibitory activity overall, with the highest inhibition observed at 50 mg/L (14.51%), whereas inhibition decreased markedly at 100 mg/L (1.48%). Chlorothalonil is a broad-spectrum fungicide with a multisite contact mode of action that interferes with multiple enzymatic processes in fungal cells (Hussien et al., 2022). In contrast, pencycuron, which belongs to the phenylurea group (FRAC 20), acts by disrupting cytoskeleton and motor protein functions in fungal cells. However, under in vitro conditions its inhibitory effect against *A. rolfsii* appeared relatively limited. These findings are consistent with Singh et al. (2019), who reported that chlorothalonil effectively reduced diseases caused by *Sclerotium rolfsii* in mustard plants. It was found that the technique employed is appropriate for supporting sustainable and environmentally friendly control of stembase rot disease.

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Table 4. Effectiveness of Fungicides on the Growth of *A. rolfsii* Using the Toxic Agar Method

Treatment	Concentration	Inhibition (%)
Control	–	0.00 [c]
Pencycuron	10	9.99 [b]
Pencycuron	50	14.51 [ab]
Pencycuron	100	1.48 [c]
Chlorothalonil	10	14.60 [a]
Chlorothalonil	50	11.48 [b]
Chlorothalonil	100	21.98 [a]

[†] The scores are the average of four tests.
[Š] The same letters indicate no significant difference according to the DMRT test ($p < 0.05$).

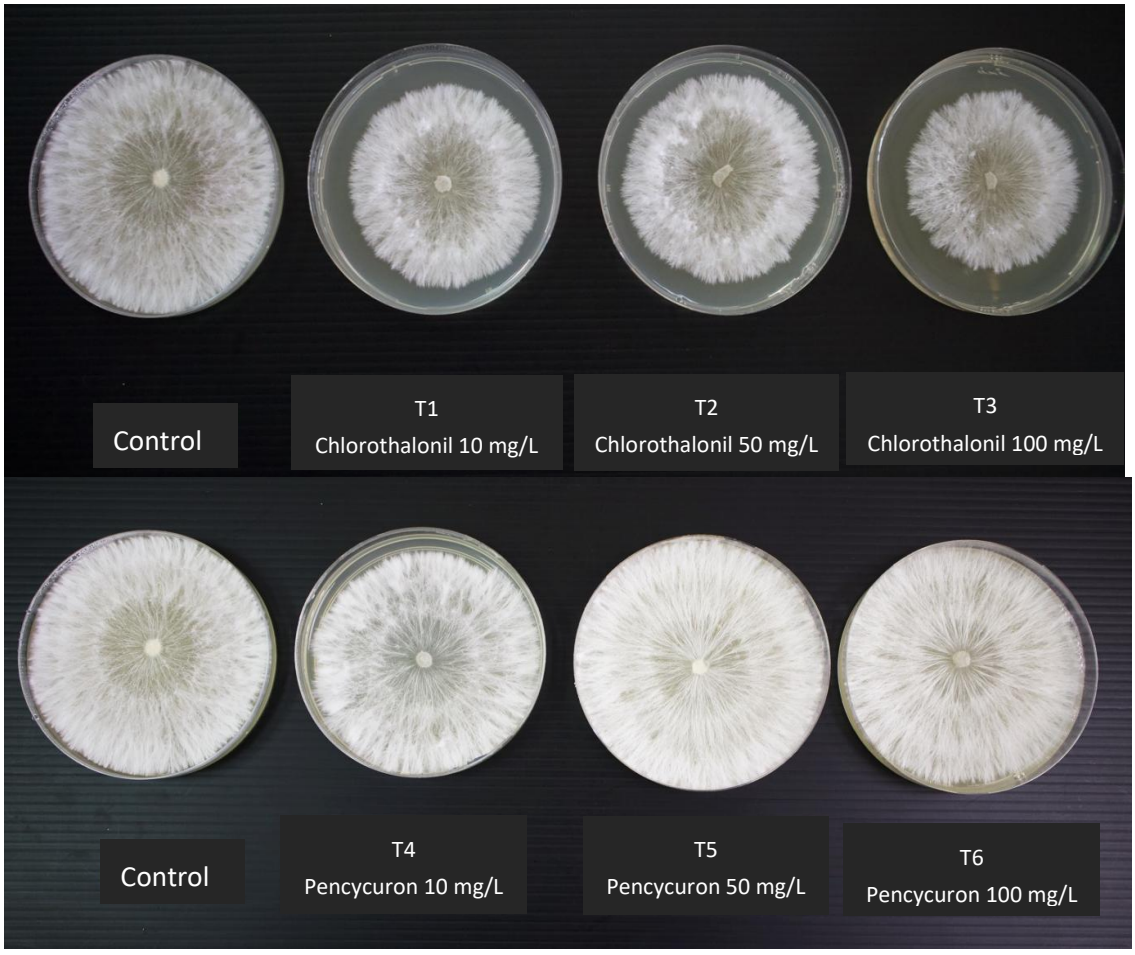

Figure 4. Representative colony growth of *A. rolfsii* using the toxic agar method. The image shows representative dishes from four replicates.

Comparison of the Effectiveness of Chlorothalonil and Pencycuron

Pencycuron, tested at concentrations of 10, 50, and 100 mg/L, appears to have low efficacy. At a concentration of 50 mg/L, the value of 14.51% was significantly different from that at 100 mg/L (1.48%), but not significantly different from that at 10 mg/L (9.99%). Interestingly, the higher the concentration administered, the lower the inhibition. This may be due to several factors, such as molecular instability at certain concentrations, possible structural adaptation of the pathogen to the active ingredient, or less effective interaction with agar components at excessively high concentrations (Suganda & Widiastuti, 2025).

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Chlorothalonil exhibited the highest inhibition at a concentration of 100 mg/L (21.98%). As the concentration decreases, the level of inhibition also decreases. This is because contact fungicides are known to work by inactivating various enzymes important in fungal metabolism through interaction with thiol groups on cellular proteins. Due to its multisite nature, chlorothalonil does not easily induce resistance. This is consistent with the findings of Hussien et al. (2022), which showed that chlorothalonil can effectively inhibit the growth of several plant pathogenic fungal species.

As the concentration of pencycuron increases, the level of inhibition decrease, in contrast to chlorothalonil. This is because pencycuron acts systemically by inhibiting the biosynthesis of fungal cell walls, particularly the chitin component. However, *A. rolfsii* is known to have cell walls rich in sclerotia and other defensive structures, so its response to certain fungicides may differ from that of other fungi. Furthermore, pencycuron's more specific nature allows resistance to develop more rapidly, particularly under in vitro conditions that do not present the environmental stresses typically found in the field.

Colony Growth Patterns as Indicators of Pathogen Response

Figures 3 and 4 show differences in the growth patterns of *A. rolfsii* mycelium under various fungicide treatments. Under chlorothalonil treatment, mycelium density was reduced and radial growth appeared asymmetrical. This indicates that the fungicide effectively influenced the pathogen's basic metabolism. At low concentrations (10 mg/L), the inhibitory effect is seen at the edges of the thinner colonies, while at high concentrations (100 mg/L), growth almost stops.

In the pencycuron treatment, the growth pattern appeared more varied and did not show a consistent decrease with increasing concentration. This indicates that A. *rolfsii*'s sensitivity to pencycuron is relatively low. The pathogen's ability to maintain hard sclerotia that are resistant to environmental conditions may be one reason why systemic fungicides do not significantly affect its mycelium growth.

Comparison with Previous Research

The results of this study are in line with the findings of Singh et al. (2019), who reported that chlorothalonil is effective in suppressing diseases caused by *A. rolfsii* in mustard plants. Meanwhile, the effectiveness of pencycuron against soil-borne pathogens is generally reported to vary and is greatly influenced by formulation, environmental conditions, and application methods. In some cases, pencycuron is more effective against leaf disease pathogens than soil-borne pathogens such as *A. rolfsii*.

Implications of the Results for the Control of Clubroot Disease in Brassica Plants

The results of this study provide an overview of the potential use of biological agents and fungicides in managing stem base rot disease caused by *A. rolfsii*. Among the tested isolates, *Trichoderma* sp. isolate T01 showed the highest antagonistic activity with a PGI value of 62.05% (Table 3). However, this value was not significantly different from *Trichoderma* sp. isolate T2 (59.87%) and *Trichoderma asperellum* MHR1 strain (59.09%) based on the DMRT test. These results indicate that several *Trichoderma* isolates possess strong antagonistic potential against *A. rolfsii*, although isolate T01 exhibited the highest inhibition among them.

Meanwhile, the fungicide assay showed that chlorothalonil generally produced higher inhibition than pencycuron in suppressing the mycelial growth of *A. rolfsii* (Table 4). Chlorothalonil at 100 mg/L resulted in the highest inhibition (21.98%) and 10 mg/L (14.60%) was significantly different from most other treatments. Moderate inhibition was observed at 50 mg/L (11.48%). In contrast, pencycuron showed relatively lower inhibitory activity, with the highest inhibition at 50 mg/L (14.51%), while the inhibition decreased markedly at 100 mg/L (1.48%). These findings indicate that the fungicides tested provided only low to moderate inhibition under in vitro conditions.

The integration of biological agents and fungicides may therefore represent a more sustainable strategy for disease management. Biological agents such as Trichoderma can act as the primary component of control due to their antagonistic activity, while fungicides may serve as a complementary measure when necessary. This approach can reduce the risk of pathogen resistance while also minimizing environmental impact and production costs. Similar results were reported by Suneeta et al. (2017), who found that the combined application of fungicides and Trichoderma provided improved control of Sclerotium disease in chilli plants.

The results of this study also support previous findings that stem rot diseases in many horticultural crops are commonly caused by soil-borne fungal pathogens that can survive for long periods in the soil. Hanif and Zamriyetti (2023), for example, identified *Fusarium* sp. as the causal agent of stem rot in shallots through morphological characterization and Koch's postulates. Although the pathogen differs from the

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one investigated in this study, both pathogens share similar ecological characteristics as soil-borne fungi capable of infecting basal stem tissues and causing decay, wilting, and plant death. These similarities highlight that stem base rot represents a common challenge across various crops and is difficult to control using conventional methods due to the persistence of pathogen survival structures in the soil.

In this context, the findings of this study provide additional insights by demonstrating the antagonistic potential of Trichoderma isolates against *A. rolfsii* under in vitro conditions. The relatively higher inhibition produced by chlorothalonil compared with pencycuron also indicates that multisite fungicides may still play a supporting role in Integrated Pest Management (IPM) strategies. However, their application should ideally be integrated with biological control agents to maintain control effectiveness while reducing the risk of fungicide resistance.

Furthermore, the strong antagonistic activity observed in Trichoderma isolates is consistent with previous studies highlighting the ability of these fungi to suppress soil-borne pathogens through multiple mechanisms, including competition, antibiosis, and mycoparasitism. For example, Hasibuan et al. (2025) reported that Trichoderma isolates were effective in suppressing the growth of *Fusarium solani*. Such findings demonstrate the broad antagonistic potential of Trichoderma against different soil-borne pathogens.

Overall, the results of this study suggest that the management of stem rot disease caused by soil-borne pathogens requires an integrated and sustainable approach, this is in line with Abadi et al., (2023). The antagonistic activity of Trichoderma isolates, particularly isolate T01, combined with the moderate inhibitory effect of fungicides such as chlorothalonil, provides a conceptual basis for developing integrated disease management strategies. Nevertheless, further studies under greenhouse and field conditions are necessary to confirm the effectiveness and practical applicability of these control strategies.

CONCLUSION

This in vitro study shows that *Trichoderma* sp. T01 has the highest antagonistic ability in inhibiting the growth of *A. rolfsii*, while chlorothalonil provides the most effective level of inhibition among the fungicides tested. Both agents have the potential to be important components in an Integrated Pest Management strategy for Brassica crops. However, further field research is still needed to ensure consistency of effectiveness, determine the optimal formulation and application techniques.

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REFERENCES

Abadi, A. L., Syib'li, M. A., Aini, L. Q., Sektiono, A. W., Choliq, F. A., & Trianti, I. (2023). *Pengelolaan penyakit tumbuhan terpadu (integrated plant disease management)*. Universitas Brawijaya Press.

Cui, W., He, P., Munir, S., He, P., He, Y., & Li, X. (2019). Biocontrol of Soft Rot of Chinese Cabbage Using an Endophytic Bacterial Strain. *Frontiers in Microbiology*, *10*(July), 1–12. https://doi.org/10.3389/fmicb.2019.01471

Ghasemi, S., Safaie, N., Shahbazi, S., Shams-Bakhsh, M., & Askari, H. (2020). The role of cell wall degrading enzymes in antagonistic traits of Trichoderma virens against Rhizoctonia solani. *Iranian journal of biotechnology*, *18*(4), e2333.

Handayani, I. T., Milka, & Tubul, M. (2022). Analisis Peran Pemuda dalam Ragam Organisasi pada Komite Nasional Pemuda Indonesia Kalimantan Tengah dalam Pembangunan Daerah di Kalimantan Tengah. *Jurnal Darma Agung*, *30*(3), 159–165.

10.30596/agrium.v29i1.28103

Hanif, A., & Zamriyetti. (2023). Karakterisasi Morfologi Cendawan Penyebab Penyakit Busuk Pangkal Batang Pada Bawang Merah (*Allium cepa*) Characterization Pathogenic Fungi Of Fusarium Basal Rot On Shallot (*Allium cepa*). *Agrium*, *26*(1), 76–82.

Hasibuan, E. S., Sulistiani, R., & Haron, F. F. (2025). Seleksi *Bacillus* sp. sebagai Penambat NPK dan Menghambat Aktivitas Jamur Selection of *Bacillus* sp. as an NPK enhancer and inhibiting fungal activity. *Agrium*, *28*(1), 13–23.

Hussien, R. A. A., Gnedy, M. M. A., Sayed, A. A. S., Bondok, A., Alkhalifah, D. H. M., Elkelish, A., & Tawfik, M. M. (2022). Evaluation of the Fungicidal Effect of Some Commercial Disinfectant and Sterilizer Agents Formulated as Soluble Liquid against Sclerotium rolfsii Infected Tomato Plant. *Plants*, *11*(24), 3542. https://doi.org/https://doi.org/10.3390/plants11243542

Islam, A., Nain, Z., Alam, K., Banu, N. A., & Islam, R. (2018). In vitro study of biocontrol potential of rhizospheric Pseudomonas aeruginosa against *Fusarium oxysporum f. sp*. cucumerinum. *Egyptian Journal of Biological Pest Control*, *28*(90). https://doi.org/https://doi.org/10.1186/s41938-018-0097-1

Junaidi, S. H., & Sarjan, M. (2025). Pemanfaatan Trichoderma untuk Meningkatkan Ketahanan Tanaman terhadap Serangan OPT. *Journal of Multidisciplinary Science and Natural Resource Management*, *1*(3), 12-17.

Kamel, S. M., Farag, F. M., Arafa, R. A., & Essa, T. A. (2020). Bio-Control Potentials of *Trichoderma* spp. Against *Sclerotium rolfsii* the Causative of Root and Crown Rot in Tomato, Common Bean and Cabbage. *Egyptian Journal of Phytopathology*, *48*(1), 122–136. https://doi.org/https://doi.org/10.21608/ejp.2020.54217.1018

Kumari, R., Kumar, V., Arukha, A. P., Rabbee, M. F., & Ameen, F. (2024). Screening of the Biocontrol Efficacy of Potent Trichoderma Strains against *Fusarium oxysporum f . sp . ciceri* and *Scelrotium rolfsii* Causing Wilt and Collar Rot in Chickpea. *Microorganisms*, *12*(7), 1280. https://doi.org/https://doi.org/10.3390/microorganisms12071280

Paul, N. C., Hwang, E., Nam, S., Lee, H., Lee, S., Yu, G., Kang, Y., Lee, K., Go, S., Paul, N. C., Hwang, E., Nam, S., Lee, H., Lee, J., & Yu, G. (2017). Phylogenetic Placement and Morphological Characterization of *Sclerotium rolfsii* (Teleomorph: *Athelia rolfsii*) Associated with Blight Disease of Ipomoea batatas in Korea Mycobiology Phylogenetic Placement and Morphological Characterization of *Sclerotium*. *Mycrobiology*, *45*(129–138). https://doi.org/10.5941/MYCO.2017.45.3.129

Suganda, T., & Widiastuti, A. (2025). *Optimalisasi Pengelolaan Fungisida dalam Penanganan Resistensi Jamur Patogen Tanaman*.

Singh, P., SinghRamesh, & Triphati, P. (2019). Evaluation of certain fungicides and biopesticides against stem rot of mustard caused by *Sclerotinia sclerotiorum*. *J.of Plant Development Science*, *11*(6), 367–371.

Suneeta, P., Aiyanathan, K., & Nakkeeran, S. (2017). Evaluation of *Trichoderma* spp. and Fungicides in the Management of Collar Rot of Gerbera Incited by *Sclerotium rolfsii. J Pure Appl Microbiol*, *11*(2), 1161–1168.

Wang, S.-Y., Jiang, Y.-H., Chen, X., Herrera-Balandrano, D. D., Simoes, M. F., Shi, X.-C., & Laborda, P. (2024). Biocontrol strategies for the management of *Sclerotinia sclerotiorum* in Brassica species: A review. *Physiological and Molecular Plant Pathology*, *130*, 102239. https://doi.org/https://doi.org/10.1016/j.pmpp.2024.102239

Wells, H. D. (2023). *Trichoderma as a biocontrol agent. In Biocontrol of plant diseases*. CRC Press.

Yao, X., Guo, H., Zhang, K., Zhao, M., Ruan, J., & Chen, J. (2023). Trichoderma and its role in biological control of plant fungal and nematode disease. *Frontiers in Microbiology*, *10*(1), 24–33. https://doi.org/10.3389/fmicb.2023.1160551

You, J., Tang, T., Wang, F., Mao, T., Yuan, B., Guo, J., Guo, X., Duan, Y., & Huang, J. (2021). Baseline Sensitivity and Control Efficacy of Strobilurin Fungicide Pyraclostrobin Against *Sclerotium rolfsii*. *Plant Disease*, *105*(11), 3503–3509. https://doi.org/https://doi.org/10.1094/PDIS-01-21-0176-RE

10.30596/agrium.v29i1.28103