Research Inventy: International Journal of Engineering and Science
Vol.16, Issue 4 (April 2026), PP 29-35
Issn (e): 2278-4721, Issn (p):2319-6483, www.researchinventy.com

Effectiveness of Maggot Pupal Biopesticide Enriched with Bidara (*Ziziphus mauritiana*Lam.)Against for *Phytophthora*sp. in Red Chili (*Capsicum annuum* L.)

Frudent Aulia Anjani Surahman, Bambang Irawan, Mahfut, Yulianty
Department ofBiology, FacultyMathematicsandNaturalSciences, UniversityofLampung, Jl.Prof.Dr. Ir. SumantriBrojonegoroNo. 1, KotaBandarLampung, Lampung, 35141..

Abstract:*Red chili pepper (Capsicum annuum L.) is a high-value horticultural crop that is highly susceptible to Phytophthora sp., the causal agent of stem, root, and fruit rot. The excessive use of synthetic fungicides has led to environmental contamination and increased pathogen resistance, highlighting the need for environmentally friendly control alternatives. This study aimed to identify the active compounds and evaluate the effectiveness of a biopesticide formulation based on maggot pupae (Hermetia illucens) combined with ethanolic extract of bidara leaves (Ziziphus mauritiana Lam.) against Phytophthora sp. The experiment was arranged in a Completely Randomized Design (CRD) consisting of six treatment concentrations (0–2.5%) with four replications.Both in vitro and in vivo assays were conducted to assess fungal growth inhibition and disease severity. Phytochemical screening revealed the presence of flavonoids, alkaloids, saponins, tannins, and terpenoids. The results demonstrated that increasing the formulation concentration significantly inhibited mycelial growth and reduced disease severity, with the highest effectiveness observed at the 2.5% treatment. The combination of maggot pupae extract and bidara leaf extract shows strong potential as an environmentally friendly biopesticide for controlling Phytophthora sp. in red chili pepper plants.*

***Keywords: Red chili pepper, Capsicum annuum L., Phytophthora sp. Biopesticide, Hermetia illucens, Ziziphus mauritiana* Lam.**

Date Of Submission: 07-04-2026 Date of Acceptance: 20-04-2026

I. INTRODUCTION

Red chili pepper (Capsicum annuum L.) is a high-value horticultural commodity widely cultivated in Indonesia due to its importance as a food ingredient and spice (Prajnanta, 2007; Pawar et al., 2011). However, chili productivity is often reduced by soil-borne pathogens, particularly *Phytophthora* sp., which causes root rot, stem rot, and fruit rot (Lourenço et al., 2020). The development of this disease is influenced by infection dynamics and environmental conditions that favor pathogen growth (Putaporntip et al., 2023).

Control of *Phytophthora* sp. is commonly carried out using synthetic fungicides; however, their continuous application can lead to pathogen resistance and negative environmental impacts (Agriculture and Horticulture Development Board, 2020). Therefore, more sustainable control strategies are needed, including the use of biopesticides derived from natural materials (Penaud et al., 2025). Bidara leaves (Ziziphus mauritiana Lam.) are known to contain secondary metabolites such as flavonoids, alkaloids, saponins, tannins, and terpenoids, which function as antifungal compounds (Nurlita et al., 2025; Putri et al., 2023). Flavonoids and alkaloids can disrupt fungal cell membranes and inhibit pathogen mycelial growth (Dewi & Wuryandari, 2019). In addition, plant extracts have been reported to suppress the growth of pathogenic fungi under in vitro conditions (Fitriyah et al., 2023; Syaifudin et al., 2023).

The utilization of biological materials as plant disease control agents has gained increasing attention due to their safety and environmental friendliness (Elnahal et al., 2022). The combination of bioactive compounds from different natural sources may enhance inhibitory effectiveness through synergistic mechanisms against plant pathogens (Rafiq et al., 2025). Therefore, this study was conducted to evaluate the effectiveness of a maggot pupae-based biopesticide formulation enriched with bidara leaf extract in controlling *Phytophthora* sp. in red chili pepper plants under both in vitro and in vivo conditions (Nugroho et al., 2021).

II. EXPERIMENTAL PROCEDURE

This study was conducted from October to December 2025 at the Botany Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, University of Lampung. Isolate rejuvenation and both in vitro and in vivo assays were carried out using standard laboratory equipment, including Petri dishes, micropipettes, an autoclave, incubator, oven, rotary evaporator, microscope, and laminar air flow cabinet. The materials used in this study consisted of a pure isolate of *Phytophthora* sp., maggot pupae extract (Hermetia illucens), bidara leaf extract (Ziziphus mauritiana Lam.), Potato Dextrose Agar (PDA) medium, 96% ethanol, distilled water, and red chili pepper seedlings (Capsicum annuum L.).

The extracts were obtained through maceration and concentrated by evaporation. They were then formulated at a total concentration of 10% with different treatment variations. The observed parameters included colony diameter, percentage of growth inhibition, disease incidence, and disease severity. Data were analyzed using a homogeneity test, followed by Analysis of Variance (ANOVA) and Tukey's HSD test at a 5% significance level.

1.1 Preparation of Bidara Leaf and Maggot Pupae Extracts

Bidara leaves and maggot pupae were first air-dried under sunlight and then ground into powder. Extraction was performed using the maceration method for five days, employing 96% ethanol as the solvent for bidara leaves and distilled water for maggot pupae. The filtrate was then filtered and evaporated using a rotary evaporator at 60°C to obtain concentrated extracts.

1.2 Biopesticide Formulation

Table 1. Biopesticide formulation of maggot pupae with the addition of bidara leaf extract

Treatment	Volume of 10% Biopesticide (mL)	Maggot Pupae Extract (mL)	Bidara Leaf Extract (g)	Solvent Volume (Water) (mL)
P0	40 mL	-	-	360 mL
P1	40 mL	2,5% = 10 mL	0,5% = 2 g	360 mL
P2	40 mL	2,5% = 10 mL	1% = 4 g	360 mL
P3	40 mL	2,5% = 10 mL	1,5% = 6 g	360 mL
P4	40 mL	2,5% = 10 mL	2% = 8 g	360 mL
P5	40 mL	2,5% = 10 mL	2,5% = 10g	360 mL

The biopesticide was formulated at a total concentration of 10%, consisting of a fixed concentration of maggot pupae extract (2.5%) combined with varying concentrations of bidara leaf extract (0.5–2.5%), and diluted with distilled water to a final volume of 400 mL.

1.3 Active Compound Screening Test

Active compound screening is an initial step to identify the presence of bioactive compounds in plant materials (Sulistyarini et al., 2020). Qualitative screening was conducted using specific reagents that react selectively with certain compounds such as alkaloids, flavonoids, tannins, saponins, terpenoids, and others. Naturally occurring bioactive compounds in plants exhibit biological activities, including antimicrobial and antifungal properties (Akhmadi et al., 2022).

1.4 In Vitro Inhibitory Activity Test of the Maggot Pupae-Based Biopesticide Formulation with Bidara Leaf Extract Against *Phytophthora* sp

Instant Potato Dextrose Agar (PDA) medium (39 g/L) was dissolved in 1000 mL of distilled water and sterilized using an autoclave at 121°C for 15 minutes (Sigma & Aldrich, 2022). A pure isolate of *Phytophthora* sp. was then characterized macroscopically based on colony color, texture, and elevation, and microscopically using lactophenol cotton blue staining to observe hyphae, sporangia, and zoospores (Mukhlis et al., 2018; Suhartina et al., 2018; Wulandari, 2024).

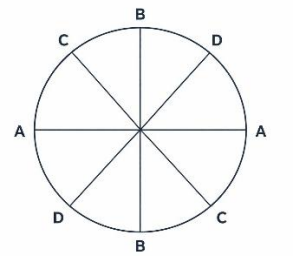
The biopesticide was formulated in a 10% solution consisting of maggot pupae extract (fixed at 2.5%) and ethanolic bidara leaf extract at varying concentrations of 0.5%, 1%, 1.5%, 2%, and 2.5%. Each formulation was mixed into 25 mL of sterile PDA medium in a Petri dish and homogenized. After solidification, one loopful (1 ose) of *Phytophthora* sp. culture was inoculated at the center of the medium and incubated at room temperature for 5–10 days.

Colony diameter growth was measured according to Rahmawati (2025) using the following formula:

$$\text{Colony Diameter} = \frac{(AA) + (BB) + (CC) + (DD)}{4}$$

Description:
AA : Horizontal colony diameter
BB : Vertikal colony diameter
CC dan DD : Diagonal colony diameters

A diagram illustrating the measurement of *Phytophthora* sp. colony growth diameter is presented in **Figure 1.**



The percentage of inhibition against fungal growth was calculated using the following formula:

$$\text{Inhibition Zone} = \frac{k - p}{k} x\ 100\%$$

Description:
k : Colony diameter on control medium
p : Colony diameter on treatment medium

1.5 **Preparation of *Phytophthora* sp. Conidial Suspension**

After morphological characterization, a 7-day-old culture of *Phytophthora* sp. grown on PDA medium was suspended in 10 mL of sterile distilled water and homogenized using a vortex mixer. Spore density was determined using a hemocytometer and adjusted to 10^5 spores/mL as the standard inoculum concentration (Avin, 2019).

2.5**In Vivo Test of the Biopesticide Formulation Concentration of Maggot Pupae with the Addition of Bidara Leaf Extract**

The in vivo assay was conducted to evaluate the effectiveness of the maggot pupae-based biopesticide formulation combined with bidara leaf extract against Phytophthora sp. infection in red chili pepper (Capsicum annuum L.). Twenty-eight-day-old seedlings (5–6 leaves) were inoculated with 10 mL of a pathogen suspension containing 10^5 spores/mL per plant, except for the control treatment (Butu et al., 2022). The biopesticide was applied once a week at a dose of 100 mL per plant according to the respective treatments. Plants were maintained at a temperature of 25–30°C and relative humidity of 70–80% (Katoch & Singh, 2021).

The observed parameters included disease incidence and disease severity. Disease incidence was calculated using the formula proposed by Cruz-Rodriguez et al. (2020) as follows:

1. Disease Incidence

$$DI = n / N \times 100\%$$

Description :
DI : Disease Incidence Percentage
n : Number of plants showing disease symptoms
N : Total number of observed plants

2. Disease Severity

$$DS = \frac{\Sigma (n \cdot v)}{N \cdot V} \times 100\%$$

Description:
DS : Disease severity
n : Number of infected plants in each category
v : Score of each category
N : Score of each category
V : Maximum score on the severity scale

III. RESULT AND DISCUSSION

3.1. Results of Active Compound Screening

Table2. Results of Active Compound Screening of the Maggot Pupae-Based Biopesticide Formulation with Bidara Leaf Extract

Test	Result	Indicator
Alkaloids		
• Mayer's reagent	+	Formation of a white or yellow precipitate
• Dragendorff' reagent	+	Formation of an orange precipitate
• Bouchardat reagent	+	Formation of a brown or black precipitate
Flavonoids	+	Yellow, red, or orange color change
Saponins	+	Formation of stable foam (5 cm height for 5 minutes)
Tannins	+	Color change to greenish-black
Terpenoids	+	Formation of a reddish-orange ring
Steroids	+	Absence of green color change in the solution
phenols	+	Dark bluish-black color change

Keterangan :+ = Presence of tested compound
- = Absence of tested compound

The qualitative screening results indicated that the maggot pupae-based biopesticide formulation enriched with bidara leaf extract contained alkaloids, flavonoids, saponins, tannins, phenols, terpenoids, and steroids. These compounds are known to exhibit antifungal activity through mechanisms such as disruption of cell membranes and cell walls, inhibition of ergosterol and chitin synthesis, and interference with hyphal and mycelial growth (Dewi & Wuryandari, 2019). The combination of these bioactive metabolites may act synergistically in suppressing the growth of *Phytophthora* sp. (Nugroho et al., 2021).

3.2 *In-Vitro* Inhibitory Activity of the Biopesticide Formulation

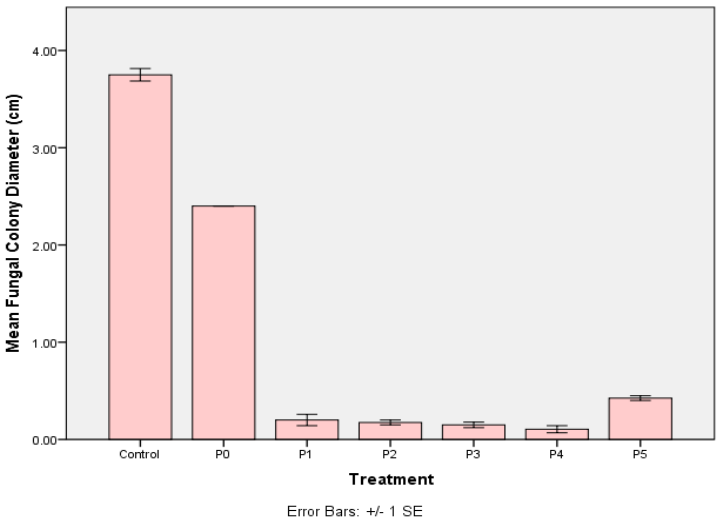


Figure 1. Colony growth diameter of *Phytophthora* sp. after application of the maggot pupae-based biopesticide formulation with the addition of bidara leaf extract.

Figure 1. Colony diameter of *Phytophthora* sp. after application of the maggot pupae-based biopesticide formulation with bidara leaf extract.Figure 1 shows that the largest colony diameter was observed in the control (P0, without formulation), while all biopesticide treatments (P1–P5; 0.5–2.5%) resulted in smaller colony diameters. The lowest diameter was recorded in P1 (0.5%), followed by P2 (1%) and P5 (2.5%), whereas

P3 (1.5%) and P4 (2%) showed relatively similar values. However, no statistically significant differences were found among the biopesticide treatments.The reduction in colony diameter in treatments P1–P5 (0.11–0.43 cm) compared to the control (3.75 ± 0.13 cm) indicates that the biopesticide formulation effectively inhibited the mycelial growth of *Phytophthora* sp. A smaller colony diameter reflects a higher level of fungal growth inhibition (Ghartavol et al., 2025).

The image of the growth of *Phytophthora* sp. colonies on PDA medium is presented in the figure below.

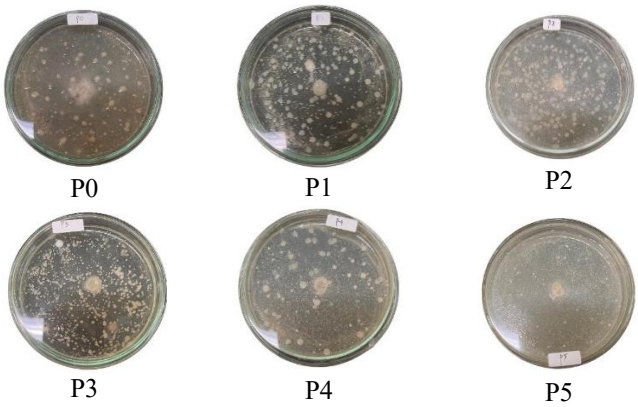


Figure 2. Comparison of *Phytophthora* sp. colony diameter on PDA medium.

The results demonstrate that the maggot pupae-based biopesticide formulation with bidara leaf extract significantly affected the colony growth of *Phytophthora* sp., as evidenced by the reduced colony diameter in all treatments compared to the control.Inhibition percentage data also showed increased suppression across all treatments, with P1 (0.5%) producing the highest inhibition, followed by P2 (1%) and P5 (2.5%). Although all treatments were effective in suppressing fungal growth, their effectiveness varied depending on formulation composition and concentration. Treatments P3 (1.5%) and P4 (2%) exhibited relatively constant values, suggesting a plateau in antifungal activity. The inhibitory effect is associated with bioactive compounds such as alkaloids, flavonoids, saponins, and tannins, which possess antifungal properties. These compounds can disrupt fungal cell wall structure, metabolism, and cell division, thereby preventing optimal mycelial development (Setiari et al., 2019). Overall, the biopesticide formulation effectively suppressed *Phytophthora* sp. colony growth, as shown by reduced colony diameter and increased inhibition percentage. Its effectiveness is influenced by formulation concentration, fungal characteristics, and experimental conditions.

3.3 *In-Vitro* Inhibition Percentage

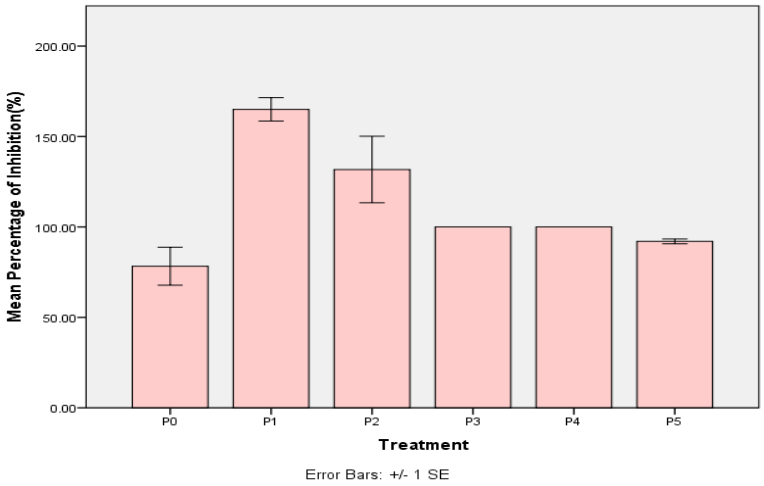


Figure 3. Inhibition percentage of *Phytophthora* sp.

Figure 3 indicates that the control (P0) exhibited the highest disease incidence (100.00 ± 0.00%). Treatments P1 (0.5%) and P2 (1%) resulted in the lowest disease incidence (16.50 ± 19.05%) and were not significantly different from each other. Treatments P3 (1.5%) and P4 (2%) showed values of 33.00 ± 0.00% and 41.25 ± 16.50%, respectively, while P5 (2.5%) showed a relatively high value (91.50 ± 17.00%) and was not

significantly different from the control.Overall, the biopesticide formulation reduced disease incidence, particularly in P1 and P2. Data were analyzed on the 17th day after inoculation, when symptoms had fully developed and differences among treatments were clearly observable.

3.4 *In-Vivo* Test Results of the Biopesticide Formulation

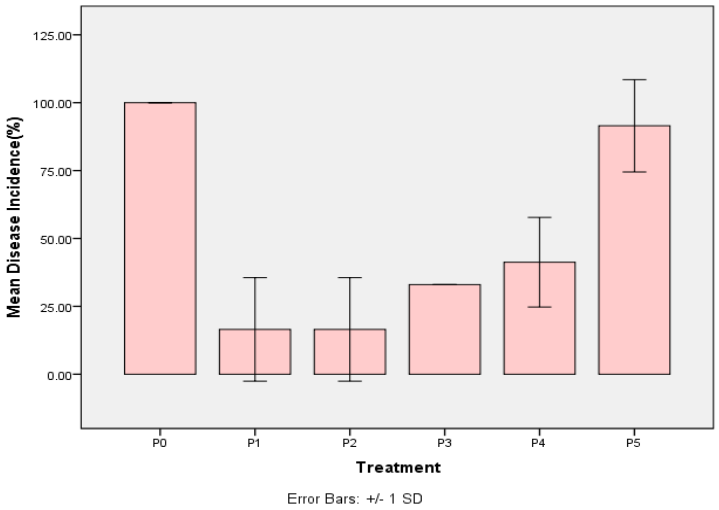


Figure 4. Disease incidence of *Phytophthora* sp. in red chili pepper plants.

Based on the graph, the highest disease incidence was observed in the control (P0), reaching nearly 100%. Treatments P1 and P2 showed a sharp decrease, with mean incidence values of approximately 16–17%, and relatively large error bars (±1 SE) due to variation among replicates. Treatments P3 and P4 showed increased incidence compared to P1 and P2, at approximately 33% and 41%, respectively. Meanwhile, P5 showed a high disease incidence (±90%), visually approaching the control.Thus, low to moderate concentrations (particularly P1 and P2) were most effective in suppressing initial *Phytophthor*a sp. infection, whereas increasing the concentration up to P5 did not enhance disease control effectiveness.

3.5 Disease Severity of *Phytophthora* sp. in Red Chili Pepper Plants

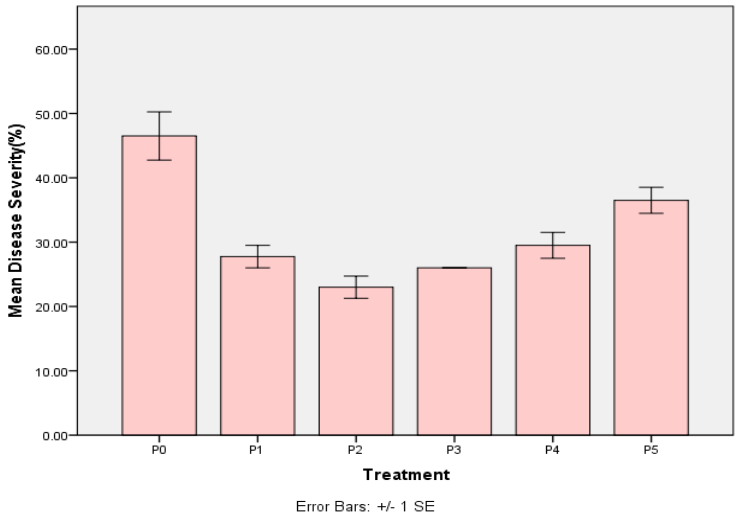


Figure 5. Disease severity of *Phytophthora* sp. in red chili pepper plants.

The disease severity graph showed a more stable pattern compared to disease incidence. The control (P0) had the highest mean severity (±46%). Treatment P2 showed the lowest severity (±23%), nearly half that of the control, with relatively small error bars, indicating low variability among replicates. Treatments P1, P3, and P4 also reduced disease severity compared to the control, with values ranging from 26–30%. Treatment P5 showed increased severity (±36%), higher than other formulated treatments but still lower than the control.

Graphically, the 1% concentration (P2) appeared to be the optimal point for suppressing disease development after infection. Increasing the concentration to 2.5% (P5) did not improve control effectiveness, indicating that formulation efficacy is not always directly proportional to concentration.

IV. CONCLUSION

Based on the results and discussion, it can be concluded that the maggot pupae-based biopesticide formulation enriched with bidara leaf extract contains active compounds, including alkaloids, flavonoids, saponins, tannins, phenols, steroids, and terpenoids, which play a role in inhibiting the growth of *Phytophthora* sp. Furthermore, concentrations of 0.5–1% (P1 and P2) were the most effective in suppressing the growth and development of *Phytophthora* sp., both under in vitro and in vivo conditions

ACKNOWLEDGEMENT

The author would like to thank the Research and Development Laboratory of PT. Great Giant Pineapple for providing facilities for this research and thank the laboratory staff who helped the author complete this research.

DAFTAR PUSTAKA

[1]. Agriculture and Horticulture Development Board. (2020). Application and management of biopesticides for efficacy and reliability (AMBER): Grower summary – fourth annual report (Project CP 158). AHDB Horticulture.
[2]. Akhmadi, C., Utami, W., & Annisa, E. (2022). Narrative review: Senyawa fitokimia dan aktivitas farmakologi family Basellaceae sebagai obat luka. Journal of Research in Pharmacy, 2(2), 77–85.
[3]. Avin, F. A. (2019). Easy way to count spores and prepare spore suspension by hemocytometer. Otis L. Floyd Nursery Research Center, 1(4), 12–15.
[4]. Butu, M. Rodino, S., and Butu, A. 2022. Biopesticide formulations-currentchallenges and future perspectives. In Biopesticides (pp. 19-29). Woodhead Publishing.
[5]. Cruz-Rodríguez, R. I., Cruz-Salomón, A., Ruiz-Lau, N., Pérez-Villatoro, J. I., Esquinca-Avilés, H. A., & Meza-Gordillo, R. (2020). Potential application of Crotalaria longirostrata branch extract to reduce the severity of disease caused by Fusarium. Agronomy, 10(4), 524.
[6]. Dewi, R. K., & Wuryandari, R. (2019). Mekanisme kerja alkaloid sebagai senyawa antifungi pada patogen tanaman. Jurnal Perlindungan Tanaman Indonesia, 23(2), 101–109.
[7]. Elnahal, A. S., et al. (2022). The use of microbial inoculants for biological control, plant growth promotion, and sustainable agriculture: A review. European Journal of Plant Pathology, 162(4), 759–792.
[8]. Fitriyah, N., Rahmawati, D., & Kurniawan, A. (2023). Aktivitas ekstrak daun tanaman terhadap penghambatan pertumbuhan jamur patogen secara in vitro. Jurnal Agroteknologi Tropika, 11(1), 45–53.
[9]. Katoch, S., & Singh, A. (2021). Screening of capsicum germplasm for resistance against Phytophthora capsici causing leaf blight and root rot. Journal of Entomology and Zoology Studies, 9(2), 627–630.
[10]. Lourenço, D., Branco, I., & Choupina, A. (2020). Phytopathogenic oomycetes: A review focusing on Phytophthora cinnamomi and biotechnological approaches. Molecular Biology Reports, 47(11), 9179–9188.
[11]. Nugroho, A. P., Santosa, B., & Wibowo, R. (2021). Mekanisme aktivitas flavonoid sebagai antifungi dan antioksidan pada patogen tanaman. Jurnal Mikologi Indonesia, 15(2), 88–96.
[12]. Nurlita, D., Sari, R. P., Handayani, S., & Pratama, A. R. (2025). Flavonoid, alkaloid, dan terpenoid sebagai metabolit sekunder tumbuhan dan perannya dalam perlindungan tanaman. Prosiding Seminar Nasional Biologi, 2(2), 7–9.
[13]. Pawar, S. S., Bharude, N. V., Sonone, S. S., Deshmukh, R. S., Raut, A. K., & Umarkar, A. R. (2011). Chillies as food, spice and medicine: A perspective. International Journal of Pharmacy and Biological Sciences, 1(3), 11–18.
[14]. Penaud, L., Martin, G., & Roche, P. (2025). Plant-derived biopesticides and their physiological effects on host–pathogen interactions. Plant Protection Science, 61(1), 14–22.
[15]. Prajnanta, F. (2007). Agribisnis cabai merah hibrida. Penebar Swadaya.
[16]. Putaporntip, C., Jongwutiwes, S., & Thongkamngam, T. (2023). Disease progress and infection dynamics of Phytophthora spp. in horticultural crops. Plant Pathology Journal, 39(2), 120–129.
[17]. Putri, A. D., Wulandari, S., & Permatasari, N. (2023). Aktivitas saponin sebagai antifungi dan perannya dalam pengendalian patogen tanaman. Jurnal Agroindustri Hayati, 10(2), 66–74.
[18]. Rahmawati, M. (2025). Uji pertumbuhan koloni jamur B. theobromae dan perhitungan diameter koloni berdasarkan empat arah. Jurnal Proteksi Agrikultura, 2(2), 165–171.
[19]. Sigma-Aldrich. (2022). Potato dextrose agar (PDA): Technical data sheet. EMD Millipore.2(1):7-13.
[20]. Setiari, N., Wicaksono, D., & Puspitasari, R. 2019. Faktor-faktor yang memengaruhi efektivitas ekstrak tanaman terhadap pertumbuhan jamur patogen. Jurnal Agroteknologi Indonesia. 14(2):95–103.
[21]. Suhartina, Febby, E., & Marina, F. (2018). Isolasi dan identifikasi jamur endofit pada tumbuhan paku Asplenium nidus. Jurnal MIPA Unsrat, 7(2), 24–28.
[22]. Sulistyarini, I., Sari, D. A., & Wicaksono, T. A. (2020). Skrining fitokimia senyawa metabolit sekunder batang buah naga (Hylocereus polyrhizus). Jurnal Ilmiah Cendekia Eksakta, 5(1), 56–62.
[23]. Syaifudin, E., Akhsan, N., & Aryubi, A. (2023). Efektivitas ekstrak gulma dalam menghambat penyakit (Phytophthora sp.) tanaman cabai (Capsicum annuum L.) secara in vitro. Jurnal Agroteknologi Tropika Lembab, 5(2), 136–142.
[24]. Vakili-Ghartavol, R., Mohammadi, A., & Rezaei, S. (2025). Evaluation of mycelial growth inhibition as an indicator of antifungal activity of botanical biopesticides. Journal of Fungi, 11(1), 45.
[25]. Wulandari, Y. (2024). Karakterisasi makroskopis dan mikroskopis jamur. Cahaya Media. I.Q., 2014. Synthesis and antimicrobial activity of Ibuprofen derivatives. Natural Science 6, 47–53.