NANOEMULSION BIOPESTICIDE FORMULATED FROM *Zingiber Purpureum* EXTRACT AND PATCHOULI OIL DISTILLATION WASTE FOR CONTROLLING BACTERIAL LEAF BLIGHT ON RICE

FORMULASI BIOPESTISIDA NANOEMULSI DARI EKSTRAK *Zingiber Purpureum* DAN LIMBAH PENYULINGAN MINYAK NILAM UNTUK MENGENDALIKAN HAWAR DAUN BAKTERI PADA TANAMAN PADI

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ABSTRACT

Bacterial leaf blight, caused by *Xanthomonas oryzae* pv. *oryzae* (Xoo), is one of the most destructive diseases in rice crops. This study aimed to assess the efficacy of nanoemulsion biopesticides derived from bangle extract (*Zingiber purpureum*) and patchouli oil distillation waste in controlling Bacterial Leaf Blight (BLB) in rice. The experiment used a Randomized Block Design with six replications, including a control, nanoemulsion biopesticide concentrations of 500, 1000, 1500, and 2000 ppm, and a bactericide as a positive control. The observed parameters included pathology (incubation period, disease intensity, infection rate, and area under disease progression curve) and agronomic traits (plant height, number of tillers, number of panicles, number of grains, 100-grains weight, root and shoot dry weight). The results showed that the application of biopesticide nanoemulsions, although it does not prolong the incubation period, was able to reduce disease intensity up to 8.7% compared to the control at a concentration of 2000 ppm and also lowered the rate of disease infection. The treatment did not have a significant effect on vegetative growth, however nanoemulsion biopesticides at concentrations above 1000 ppm increased production components, including grain number by 32% and 100-grain weight by 6.1% compared to the control. This study demonstrates that the nanoemulsion biopesticide formulation of bangle extract and patchouli oil distillation waste has the potential to be used as an environmentally friendly biopesticide to control Bacterial Leaf Blight while simultaneously increasing rice productivity.

Key words: Bacterial leaf blight, Bangle extract, Nanoemulsion, Patchouli oil distillation waste

ABSTRAK

Hawar daun bakteri yang disebabkan oleh *Xanthomonas oryzae* pv. *oryzae* merupakan salah satu penyakit paling merusak pada tanaman padi. Penelitian ini bertujuan mengevaluasi

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efektivitas biopestisida nanoemulsi berbahan ekstrak bangle *(Zingiber purpureum*) dan limbah penyulingan nilam dalam menekan hawar daun bakteri pada padi. Percobaan dilakukan dengan Rancangan Acak Kelompok yang terdiri atas enam perlakuan (kontrol, biopestisida nanoemulsi dengan konsentrasi 500, 1000, 1500, dan 2000 ppm, serta bakterisida sebagai kontrol positif) dengan enam ulangan. Parameter yang diamati meliputi aspek patologi (masa inkubasi, intensitas penyakit, laju infeksi, dan *area under disease progression curve*) serta aspek agronomi (tinggi tanaman, jumlah anakan, jumlah malai, jumlah biji, bobot 100 biji, bobot kering akar dan tajuk). Hasil penelitian menunjukkan bahwa meskipun tidak memperpanjang masa inkubasi, aplikasi nanoemulsi biopestisida tetapi mampu menekan intensitas penyakit mencapai 8.7% dibandingkan dengan kontrol pada konsentrasi 2000 ppm serta menurunkan laju infeksi penyakit. Perlakuan tidak menunjukkan perbedaan respon terhadap pertumbuhan vegetatif, namun biopestisida nanoemulsi dengan konsentrasi di atas 1000 ppm meningkatkan komponen produksi berupa jumlah biji sebesar 32% dan bobot 100 biji mencapai 6,1% dibandingkan dengan kontrol. Penelitian ini menunjukkan bahwa formulasi biopestisida nanoemulsi ekstrak bangle dan limbah nilam berpotensi digunakan sebagai biopestisida ramah lingkungan untuk mengendalikan hawar daun bakteri sekaligus meningkatkan produktivitas padi.

Kata kunci: Ekstrak bangle, Hawar daun bakteri, Limbah penyulingan nilam, Nanoemulsi

INTRODUCTION

Bacterial leaf blight (BLB) is a significant disease impacting rice crops globally, caused by *Xanthomonas oryzae* pv. *oryzae* (Xoo) (Ritbamrung et al., 2025). This disease can result in yield losses of up to 100% in Indonesia (Safni et al., 2021). The disease is recognized as a vascular wilt in the initial phases of plant development and as leaf blight throughout the flowering period. The gram-negative bacterium Xoo infiltrates the host plant via wounds or hydatodes, infecting xylem tissue and resulting in a systemic infection. This rod-shaped motile bacterium generates yellowish slime and was initially identified as a bacterial cluster in dewdrops on rice plants (Teja et al., 2025). Xoo transmits from diseased leaves to the roots and thereafter disseminates throughout the rhizosphere inside the soil and nearby water. Xoo was identified in rice seeds, leaves, stems, roots, soil and water throughout three-month period in controlled pots or at the rice fields (Ritbamrung et al., 2025).

Approaches to manage BLB have been implemented using many chemical and cultural strategies, although their efficacy has been limited. This is caused by the variable complexity of infections a lack of enduring plant resistance (Das et al., 2025), and differential susceptibility to various pesticides and environmental factors (Safni et al., 2021). The extensive application of chemical pesticides as a solution inadvertently leads to negative impacts, such as environmental contamination, threats to human health, and the development of pathogen resistance (Li et al., 2024). Therefore, the development of natural, environmentally sustainable pesticides and integrated pathogen management techniques, including essential oils and essential oil-derived bio-actives for the control of Xoo, has been warranted due to the negative impacts of chemical pesticides.

Zingiber purpureum is an essential oil-producing plant from the Zingiberaceae family known to have antimicrobial activity against a number of plant pathogens, including *Rhizoctonia solani* and *Xanthomonas oryzae* (Devkota et al., 2021). The essential oil from this plant contains active compounds such as sabinene (27-34%), terpinen-4-ol (30-5%), β-pinene (14.3%), carryophyllene (9.5%), γ- terpinene (6-8%), and α-terpinene (4-5%) which have antibacterial activity (Devkota et al., 2021; Julaeha et al., 2021). Additionally, the patchouli plant (*Pogostemon cablin* B.) from the Lamiaceae family is an aromatic plant that produces essential oil with pogostone and patchouli alcohol as its main components (Singh & Agrawal, 2024). Both compounds have strong antimicrobial activity, primarily through mechanisms of disrupting cell membrane permeability and inhibiting microbial metabolic enzymes (Suhartono et al., 2024; Yang et al., 2013). The residue from patchouli oil distillation waste has been observed to retain bioactive components with antimicrobial properties, rendering it appropriate for subsequent utilization as a biopesticide. The antibacterial properties of *Z. purpureum* and *P. cablin* are suitable candidates for developments as active components in biopesticides against BLB caused by Xoo.

Nanoemulsion technology is a promising innovative approach to enhance the effectiveness of the bioactive compounds from both plants. Nanoemulsion facilitate the delivery of lipophilic compounds such as antibacterials and antioxidants in the form of very small particles (<100 nm), thereby augmenting the interaction of these compounds with biological membranes. Nanoemulsions also provide benefits including minimal polydispersity, controlled release of bioactive compounds, low viscosity, high kinetic, and comparatively low surfactant usage (Savitharani et al., 2024). Various studies have demonstrated that essential oil nanoemulsions help manage phytopathogens and have been extensively utilized in agriculture (Suharti et al., 2025).

This research aimed to investigate the efficacy of *Z. purpureum* extract and patchouli oil distillation waste (*P. cablin)* into nanoemulsions as a biopesticide against BLB in rice plants caused by *Xanthomonas oryzae* pv. *oryzae.* The nanoemulsion method aims to maximize the utilization of bioactive components from both plants to improve antibacterial efficacy, while also supporting the development of sustainable and environmentally friendly agriculture practices.

MATERIALS AND METHODS

Experimental Design

This study was carried out from June 2024 to February 2025 at the Plant Protection Laboratory and Greenhouse, Faculty of Agriculture, Jenderal Soedirman University, Purwokerto. The experimental design used was a Randomized Complete Block Design (RCBD) consisting of six treatments and six replications, resulting in a total of 36 experimental units. The treatments applied were different concentrations of nanoemulsion biopesticide as follows: P0: Control (no treatment), P1: 500 ppm, P2: 1000 ppm, P3: 1500 ppm, P4: 2000 ppm, and P5: Bactericide (positive control).

Preparation of Nanoemulsion Biopesticide

The nanoemulsion biopesticide was formulated using bangle extract and patchouli oil distillation waste (PODW). Bangle rhizomes were cleaned, sliced, oven-dried at 60°C for 3 days, ground into powder, and filtered through a 40-mesh sieve before being stored in a glass jars. The powder was macerated in 96% ethanol with 1:10 (w/v) ratio for 3 days at room temperature with daily stirring. The filtrate was collected and concentrated using a rotary evaporator at 78℃ for 53 minutes to obtain a crude extract (Suharti et al., 2023). The resulting thick extract of *Z. purpureum* rhizome, was stored in a refrigerator at 5℃ until used.

PODW was obtained after essential oil extraction from dried *P. cablin* leaves, which were previously oven-dried at 80℃ for 3 hours. Distillation was carried out using distillation device involving a heating mantle and condenser, with 5 g dried leaves and 150 mL distilled water heated at 100℃ for 1 hour. The distillation product was collected in a burette to separate the essential oil and water phase. In this study, the water phase of the distillation product (hydrosol) was used as the base material for biopesticide nanoemulsions (Suharti et al., 2025).

The nanoemulsion was prepared using a water phase (patchouli oil distillation waste + Tween 80) and an organic phase (bangle extract + bioethanol). The first process in the preparation started with homogenizing the water phase ingredients at 250 rpm for 30 minutes, followed by the gradual addition of the organic phase and further homogenization for 45 minutes. The formula was as follows: 87 mL PODW, 3 mL Tween 80, 7.5 mL bangle extract, and 2.5 mL ethanol.

Application of Nanoemulsion Biopesticide and Inoculation of Pathogen *Xanthomonas oryzae* pv. *oryzae*

Inoculation with the bacterium *Xanthomonas oryzae* pv. *oryzae* (Xoo) was conducted once during the vegetative stage of the plant, specifically 30 days after planting (dap). The inoculation procedure was performed using the leaf clipping method, which involves cutting the tips of three leaves per plant clump using sterile scissors previously immersed in a suspension of Xoo bacteria at a concentration of 10^8 CFU mL^{-1} (Savitharani et al., 2024). Thus, across all treatments, the total number of inoculated leaves per treatment was three plants. After inoculation, the plants were allowed an acclimatization period of 48 hours to enable the pathogen to initiate early infection prior to the next biopesticide application. Inoculation was carried out between 15:00 and 17:00 WIB to avoid direct sunlight exposure and reduce high evaporation rates.

Biopesticides were applied in the morning or evening to minimise evaporation caused by direct sunlight. The biopesticide solution was administered by spraying it onto the leaf surface, according to the specified dosage for each treatment within each experimental unit. The initial application occurred one week prior to inoculation with the Xoo bacteria, followed by regular weekly applications until the end of the observation period.

Observation Parameters and Data Analysis

The observed variables in this study consisted of pathological parameters (incubation period, disease intensity, infection rate, and area under disease progression curve/AUDPC), and agronomic parameters (plant height, number of tillers, number of panicles, number of grains, 100-grain weight, root and shoot dry weight). Disease intensity was observed every 7 days by measuring the area of symptomatic leaves (Suryaningsih et al., 2023), using the formula:

$$IP = \frac{n}{N} x\ 100\%$$

N: symptom length (cm), N: total leaf length (cm).

The Xoo infection rate was calculated weekly, employing the methodology outlined by Van der Plank 1963, utilizing the following formula (Hadiwiyono et al., 2021):

$$r = \frac{2,3}{t} (\log \frac{Xt}{(1-Xt)} - \log \frac{Xo}{(1-Xo)})$$

r: the infection rate, Xt: proportion of initial disease, Xo: proportion of diseased at time t, and t: observation time.

The AUDPC computation was executed to ascertain the correlation between disease intensity and time. AUDPC was computed utilizing subsequent formula (Hadiwiyono et al., 2021):

$$AUDPC: \sum_{i=1}^{n} \left(\frac{Y_{i=1} + Y_i}{2}\right) [X_{i+1} - X_i]$$

Yi: disease intensity at the i^{-th} observation, Xi: time (days) at the i^{-th}, and n: total number of observations.

The data were then analysed using an analysis of variance (ANOVA) and Duncan multiple range test (DMRT) at confidence level of α 5%.

RESULTS AND DISCUSSION

According to the research findings, nanoemulsion biopesticides derived from bangle extract and PODW affected rice plant pathological parameters. Bacterial leaf blight (BLB) was discovered four to seven days after inoculation (DAI). Although nanoemulsion biopesticide treatments did not prolong the onset of initial infection symptoms, disease severity varied significantly between treatments. The control (P0) had the maximum intensity (97.5%), while treatment P4 had the lowest intensity (89%) (Table 1). The application of nanoemulsion biopesticide was not significantly different for AUDPC value, it is indicating that disease progression was relatively similar across treatments. However, the nanoemulsion biopesticide was able to reduce disease severity by 1.4 - 8.7% compared to the untreated control.

Table 1. Effect of nanoemulsion biopesticides from bangle extract and PODW on pathological parameters.

Treatments	Observations Variable			
	Incubation period (DAI)	Disease severity at 7 (WAI)	Infection Rate (units/day)	AUDPC
P0 (no treatment)	4.17 a	97.50 c	0.18 a	289.58 a
P1 (500 ppm)	4.33 a	96.06 bc	0.18 a	250.00 a
P2 (1000 ppm)	5.17 a	91.33 ab	0.17 a	246.70 a
P3 (1500 ppm)	7.00 a	94.06 bc	0.17 a	236.75 a
P4 (2000 ppm)	7.00 a	89.00 a	0.15 a	229.81 a
P5 (fungicide)	7.00 a	94.00 bc	0.17 a	247.50 a

Note: Numbers accompanied by the identical letter within the same treatment and variable denote no significant difference according to the DMRT 0.05 test. Dai: days after inoculation, WAI: weeks after inoculation.

Meanwhile, the use of bactericide reduced disease severity by 3.58% compared to the control. This suggests a lower efficacy than applications utilizing the nanoemulsion biopesticides. These data suggest that the use of the nanoemulsion biopesticides can improve plant resistance by suppressing symptom development and reducing the severity of BLB. Treatment P4 was found to be the most effective at suppressing the disease when compared to other treatments. Biopesticides formulated with nanoemulsion technology demonstrated enhanced efficacy in inhibiting BLB on rice. This finding is consistent with Suharti et al. (2023) which states that nanoemulsion biopesticides derived from *Kaempferia galanga* extract and PODW reveal antibacterial action against *Xanthomonas oryzae* pv. *oryzae* (Xoo). Bangle extract contains essential oils with antimicrobial properties inhibiting Xoo (Lee et al., 2016), including sabinene, terpinen-4-ol, α-terpinene and γ-terpinene (Devkota et al., 2021). Sabinene the most abundant compound in the *Z. Purpureum* has antibacterial activity that inhibits bacterial growth, biofilm formation, and bacterial virulence factors (Christiane et al., 2025). Terpinene-4-ol has been suggested to disseminate and damage bacterial cell membrane structures, producing higher fluidity, disrupting membrane organization, and inhibiting membrane-bound enzymes (Mondello et al., 2022).

PODW contains active antibacterial chemicals such as caryophyllene oxide, pogostol, patchouli alcohol, and β-cyperone, which inhibit the growth of Xoo (Muhammad et al., 2025; Zhang et al., 2024). Some studies have also found that PODW can suppress the growth of pathogenic fungi including *Rhizoctonia solani* and *Synchytrium pogostemonis* (Sriwati et al., 2022). Active compounds in PODW, such as patchouli alcohol, cause damage to bacterial cytoplasmic membranes, characterized by changes in permeability, leakage of cell contents, and the formation of gaps between the cytoplasmic membrane and the cell wall. This mechanism allows patchouli oil to penetrate the cell and potentially damage cytoplasmic inclusions and DNA molecules, leading to bacterial cell death (Dai et al., 2012).

The early symptoms of bacterial leaf blight (BLB) are characterized by the appearance of gray spots on the leaf tips, which then develop into chlorosis, followed by the drying of leaf tissue until it turns brown. In the severe symptoms, necrosis spreads further from the tip toward the base of the leaf (Figure 1).

Figure 1. Symptoms of bacterial leaf blight on rice.

According to Lim et al. (2008) indicate that the distinctive symptoms at the leaf tip are closely related to the colonization of *Xanthomonas oryzae* pv. *oryzae* (Xoo) within the xylem tissue. The pathogenic mechanism of Xoo involves the formation of biofilms composed of exopolysaccharides (EPS), which disrupts water and nutrient transport in plant tissues (Dey &

Raghuwanshi, 2024). Furthermore, twitching motility facilitated by type IV pili enhances bacterial dissemination inside the xylem. The blockage of nutrient flow caused by biofilms, along with bacterial movement through type IV pili, leads to distinct symptoms, including chlorosis and necrosis, that progressively extend from the leaf tip to the base (Yu et al., 2020).

The essential protein of *Xanthomonas oryzae* pv. *oryzae*, phosphohexose mutase (PHM), functions as a key virulence factor and serves as the primary regulator of the synthesis of major exopolysaccharides including xanthan and xanthomonadin (Tan et al., 2024). Xanthan, a primary exopolysaccharide (EPS) generated by *Xanthomonas* spp., is an established virulence factor. The type III secretion system contributes to cell death at the infection site in plants. Furthermore, xanthomonadin, a yellow pigment found in bacterial cell membranes, helps to sustain contacts with the host plant. Both xanthan and xanthomonadin are required for biofilm development in plant tissues (Kim et al., 2024). Compounds present in bangle extract, namely terpinen-4-ol, γ-terpinene, and α-terpinene, were found to bind to the PHM protein, thereby contributing to the antibacterial and antibiofilm activities against Xoo (Vishakha et al., 2022).

According to the research findings, the use of nanoemulsion biopesticides did not prolong the disease's incubation period, but it decreased the infection rate at the critical phase of bacterial leaf blight growth (Figure 2). At the beginning of the observation period (weeks 1 to 2), the infection rate was relatively low for all treatments, then increased between weeks 2 and 3.

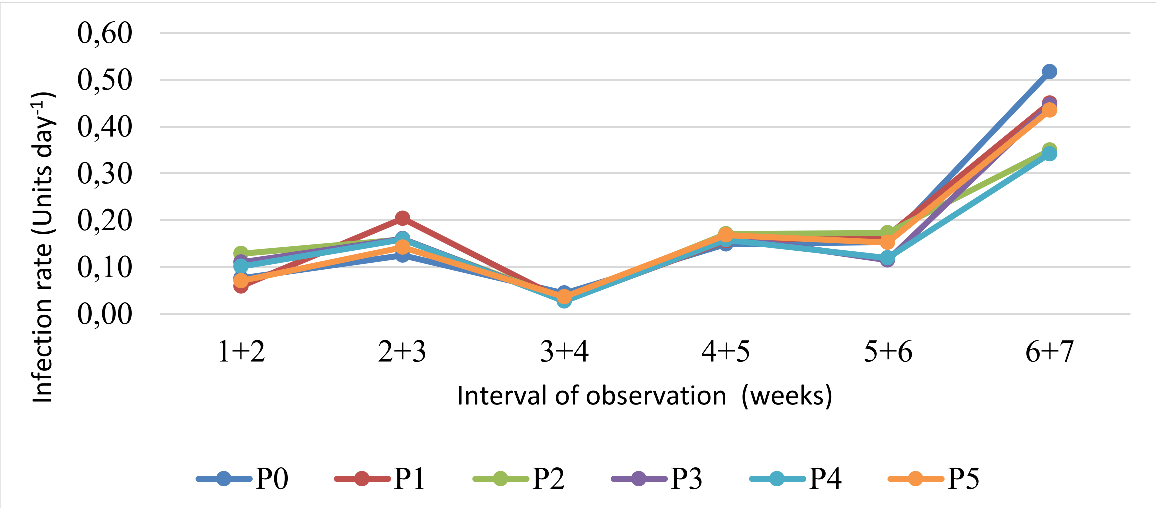

Figure 2. Infection rate of Xoo on Rice

The infection rate peaked between weeks 6 and 7, with the control group (P0) having the highest value, whereas treatments P4 and P5 had reduced infection rates. This suggests that, while infection does occur, the disease spreads more slowly in plants treated with nanoemulsion biopesticides, reducing disease severity. This effectiveness is due to the presence of bangle extract and PODW in the nanoemulsion biopesticide formulation, which have the capacity to act as an elicitor and generate plant resistance via the systemic acquired resistance (SAR) signaling pathway. According to research findings (Thepbandit et al., 2023) the application of elicitors can enhance rice plant resistance to bacterial leaf blight by generating reactive oxygen species (ROS). ROS activation promotes defense gene expression and H_2O_2 accumulation, leading to programmed cell death as a plant response to biotic and abiotic stress. Thus, the use of these nanobiopesticides not only controls disease but also activates the plant defence mechanisms. According to Lee et al. (2016) plant-derived chemical compounds can activate indirect defense pathways in plants by inducing the expression of PR genes.

Table 2. Effect of nanoemulsion biopesticide from bangle extract and PODW on agronomic parameters

Treatments	Observations Variable		
	Plant height (cm)	Number of tillers	Number of panicles
P0 (no treatment)	104.37 ab	24.67 ab	22.41 ab
P1 (500 ppm)	107.34 ab	27.50 a	23.24 a
P2 (1000 ppm)	108.09 ab	25.78 a	23.19 a
P3 (1500 ppm)	108.61 ab	27.94 a	23.45 a
P4 (2000 ppm)	109.86 a	26.94 a	23.17 a
P5 (fungicide)	100.30 b	20.17 b	18.45 b

Note: Numbers accompanied by the identical letter within the same treatment and variable denote no significant difference according to the DMRT 0.05 test.

Based on the observation results, the addition of nanoemulsion biopesticides derived from bangle extract and PODW had no significant effect on agronomic variables compared to the control (Table 2). The lack of improvement in vegetative growth variables (plant height, number of tillers, number of panicles) suggests that nanoemulsion biopesticides derived from bangle extract and PODW did not act as a growth-stimulating biofertilizer, but rather played a role in suppressing biotic disturbances during the generative phase. Bangle extract and PODW include bioactive substances such as flavonoids, phenols, and terpenoid which have antibacterial (Devkota et al., 2021; Tandirogang et al., 2022), and can reduce disease infection throughout the blooming and grain filling stages. With less biotic stress, the photosynthetic activity is more efficiently directed toward grain development and filling, increasing the quantity of grains per panicle and the weight of 100 grain. This suggests that, while vegetative growth did not differ considerably, the yield of rice in the nanoemulsion biopesticide treatment at doses more than 1000 ppm could rise significantly (Table 3).

Bangle extract (*Zingiber purpureum*) has not been shown to increase the agronomic parameters of rice plants, and research on its potential as a plant growth stimulator is currently limited. However, bangle extract has been shown to improve rice production yields. This is thought to be linked to essential mineral composition on *Zingiber* sp, which includes potassium, phosphorus, and magnesium (Tanweer et al., 2018). Phosphorus promotes grain development, potassium increases the number of grains per panicle, the percentage of full grains, and the weight of 1,000 grains, and magnesium improves the plant's photosynthetic process (Sulton Hakim et al. 2020).

Table 3. Effect of nanoemulsion biopesticide from bangle extract and PODW on the rice yields

Treatments	Observations Variable			
	Number of grains	100 grain weight (g)	Shoot dry weight (g)	Root dry weight (g)
P0 (no treatment)	3,136.33 b	2.44 b	92.50 a	100.00 a
P1 (500 ppm)	3,221.67 b	2.56 ab	83.33 a	91.67 a
P2 (1000 ppm)	4,212.00 a	2.56 ab	91.67 a	95.00 a
P3 (1500 ppm)	4,132.33 a	2.57 a	95.00 a	85.00 a
P4 (2000 ppm)	4,142.83 a	2.59 a	92.50 a	100.00 a
P5 (fungicide)	2,888.67 b	2.47 ab	81.67 a	91.67 a

Note: Numbers accompanied by the identical letter within the same treatment and variable denote no significant difference according to the DMRT 0.05 test.

CONCLUSION

1. The application of nanoemulsion biopesticides formulated from bangle extract and patchouli oil distillation waste (PODW) effectively suppressed the severity of bacterial leaf blight (BLB) on rice without prolonging the incubation period.
2. The treatment at 2000 ppm (P4) showed the highest efficacy in reducing disease intensity by up to 8.7% and infection rate, indicating its potential role in enhancing plant resistance. Although no significant effects were observed on vegetative growth parameters, nanoemulsion concentrations above 1000 ppm improved yield components, particularly grain number by up to 32% and 100-grain weight by up to 6.1%.
3. These findings highlight the potential of bangle extract and PODW nanoemulsions as eco-friendly biopesticides for integrated management of BLB while simultaneously supporting sustainable rice production.

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REFERENCES

Christiane, S., Madonna, V., Toruan, L., & Paramita, S. (2025). Antibacterial test of bangle rhizome essential oil (*Zingiber Purpureum* Roscoe) originated from East Kalimantan against *Cutibacterium acnes* and *Staphylococcus epidermidis* in Vitro. *Jurnal Ilmu Kesehatan 13*(1), 77–86.

Dai, M., Peng, C., Wan, F., & Peng, F. (2012). Antibacterial activity and mechanism of *Pogostemon cablin* against bacteria from milk of dairy cows suffering with mastitis. *Journal of Animal and Veterinary Advances*, *11*(18), 3289–3297. https://doi.org/10.3923/javaa.2012.3289.3297

Das, P. P., Kumar, A., Mohammed, M., Bhati, K., Babu, K. R., Bhandari, K. P., Ghazi, I.

A. (2025). Comparative metabolites analysis of resistant, susceptible and wild rice species in response to bacterial blight disease. *BMC Plant Biology*, *25*(1). https://doi.org/10.1186/s12870-025-06154-0

Devkota, H. P., Paudel, K. R., Hassan, M. M., Dirar, A. I., Das, N., Adhikari-Devkota, A., Dua, K. (2021). Bioactive compounds from *Zingiber montanum* and their pharmacological activities with focus on zerumbone. *Applied Sciences (Switzerland)*, *11*(21), 1–24. https://doi.org/10.3390/app112110205

Dey, R., & Raghuwanshi, R. (2024). An insight into pathogenicity and virulence gene content of *Xanthomonas* spp. and its biocontrol strategies. *Heliyon*, *10*(14), e34275. https://doi.org/https://doi.org/10.1016/j.heliyon.2024.e34275

Hadiwiyono, Poromarto, S. H., Widono, S., & Rizal, R. F. (2021). Response of a local rice variety Pandanwangi to infection of bacterial leaf blight *Xanthomonas oryzae* pv. *oryzae* strain III, IV, and VIII. *IOP Conference Series: Earth and Environmental Science*, *905*(1). https://doi.org/10.1088/1755-1315/905/1/012068

Julaeha, E., Herlina, T., Nurzaman, M., Mayanti, T., Kurnia, D., & Sari, E. F. (2021). The antibacterial effect of β-pinene derived from *Citrus aurantifolia* against oral *Streptococcus mutans*. *Padjadjaran Journal of Dentistry*, *33*(1), 88. https://doi.org/10.24198/pjd.vol33no1.29200

Kim, H., Kim, M. H., Choi, U.-L., Chung, M.-S., Yun, C.-H., Shim, Y., Lee, G. W. (2024). Molecular and Phenotypic Investigation on Antibacterial Activities of Limonene Isomers and Its Oxidation Derivative against *Xanthomonas oryzae*pv. oryzae. *Journal of Microbiology and Biotechnology*, *34*(3), 562–569. https://doi.org/10.4014/jmb.2311.11016

Lee, G. W., Chung, M. S., Kang, M., Chung, B. Y., & Lee, S. (2016). Direct suppression of a rice bacterial blight (*Xanthomonas oryzae* pv. *oryzae*) by monoterpene (S)-limonene. *Protoplasma*, *253*(3), 683–690. https://doi.org/10.1007/s00709-015-0904-4

Li, L., Wang, Y., Liu, H., Liu, W., Zhang, X., An, M., Wang, J. (2024). Antibacterial Activities and Underlying Mechanisms of the Compound SYAUP-491 against *Xanthomonas oryzae* pv. *oryzae*. *Molecules (Basel, Switzerland)*, *29*(6). https://doi.org/10.3390/molecules29061413

Lim, S. H., So, B. H., Wang, J. C., Song, E. S., Park, Y. J., Lee, B. M., & Kang, H. W. (2008). Functional analysis of pilQ gene in *Xanthomanas oryzae* pv. *oryzae*, bacterial blight pathogen of rice. *Journal of Microbiology*, *46*(2), 214–220. https://doi.org/10.1007/s12275-007-0173-9

Mondello, F., Fontana, S., Scaturro, M., Girolamo, A., Colone, M., Stringaro, A., Ricci, M. L. (2022). Terpinen-4-ol, the Main Bioactive Component of Tea Tree Oil, as an Innovative Antimicrobial Agent against *Legionella pneumophila*. *Pathogens*, *11*(6), 1–17. https://doi.org/10.3390/pathogens11060682

Muhammad, S., Isnaini, N., Ginting, B., Aiman, H. R., Ernawati, Prajaputra, V., & Sufriadi, E. (2025). Chemical characterization and antimicrobial potential of purified aceh patchouli oil (*Pogostemon cablin* Benth.) residue for floor cleaning product innovation. *South African Journal of Chemical Engineering*, *54*, 335–347. https://doi.org/https://doi.org/10.1016/j.sajce.2025.08.010

Ritbamrung, O., Inthima, P., Ratanasut, K., Sujipuli, K., Rungrat, T., & Buddhachat, K. (2025). Evaluating *Xanthomonas oryzae* pv. *oryzae* (Xoo) infection dynamics in rice for distribution routes and environmental reservoirs by molecular approaches. *Scientific Reports*, *15*(1), 1–12. https://doi.org/10.1038/s41598-025-85422-3

Safni, I., Lubis, K., & Lisnawita. (2021). Screening of Indonesian rice cultivars against bacterial leaf blight disease under acidic soil condition. *IOP Conference Series: Earth and Environmental Science*, *756*(1). https://doi.org/10.1088/1755-1315/756/1/012007

Savitharani, M., Kumar, V., Manjunatha, T., Ruksana, F., Vishala, E., & Prathap, H. M. (2024). Biopesticide potential of *Anisomeles indica* Kuntze leaf essential oil and its nanoemulsion against bacterial blight - *Xanthomonas* strains. *Journal of Natural Pesticide Research*, *8*, 100075. https://doi.org/https://doi.org/10.1016/j.napere.2024.100075

Singh, R., Singh, R., Soni, S. K., Singh, S. P., Chauhan, U. K., & Kalra, A. (2013). Vermicompost from biodegraded distillation waste improves soil properties and essential oil yield of *Pogostemon cablin* (patchouli) Benth. *Applied Soil Ecology*, *70*, 48–56. https://doi.org/https://doi.org/10.1016/j.apsoil.2013.04.007

Singh, S., & Agrawal, N. (2024). Exploring the pharmacological potential and bioactive components of *Pogostemon cablin* (Blanco) Benth, traditional Chinese medicine. *Pharmacological Research - Modern Chinese Medicine*, *10*, 100382. https://doi.org/https://doi.org/10.1016/j.prmcm.2024.100382

Sriwati, R., Pratiwi, V., & Oktarina, H. (2022). Integration of Patchouli Waste Compost, Antagonistic Microbes and Essential Oils to Control Budok Disease (*Synchytrium pogostemonis*). *Jurnal Fitopatologi Indonesia*, *18*, 153–159. https://doi.org/10.14692/jfi.18.4.153-159

Suharti, W. S., Tini, E. W., & Istiqomah, D. (2023). Antimicrobial activity of *Kaempferia galanga* against plant pathogen on rice. *Biodiversitas*, *24*(2), 1320–1326. https://doi.org/10.13057/biodiv/d240275

Suharti, W. S., Yusuf, F. K., & Istiqomah, D. (2025). Nanoemulsion Biopesticides from *Kaempferia Galanga* Extract and Patchouli Oil Distillation Waste to Control Bacterial Leaf Blight Disease of Rice. *International Journal of Agriculture and Biology*, *33*(6). https://doi.org/10.17957/IJAB/15.2331

Suhartono, S., Adhyani, L., & Amalia, A. (2024). Crude Aceh patchouli alcohol (*Pogostemon cablin* Benth.) elucidated antibiofilm activity of against *Staphylococcus aureus*. *BIO Web of Conferences*, *94*, 4–10. https://doi.org/10.1051/bioconf/20249402006

Sulton Hakim, M., Dewanti, P., Hartatik, S., Handoyo, T. (2020). Application effect of potassium on Rice (*Oryza sativa* L.) Recovery After Drought Stress. *Jurnal Ilmu Dasar*, *21*(2), 115.

Suryaningsih, A. S., Triwidodo, H., & Wiyono,

S. (2023). Resistance of Six Lines from Farmers against Bacterial Leaf Blight (*Xanthomonas oryzae* pv. *oryzae*). *Jurnal Fitopatologi Indonesia*, *19*(4), 176–181. https://doi.org/10.14692/jfi.19.4.176-181

Tan, N., Huang, Y., Miao, W., Zhang, Q., & Wu, T. (2024). Type III Secretion Effectors of *Xanthomonas oryzae* pv. *oryzicola*: The Arsenal to Attack Equivalent Rice Defense for Invasion. *Agronomy*, *14*(9), 1–14. https://doi.org/10.3390/agronomy14091881

Tandirogang, N., Anitasari, S., Arung, E. T., Paramita, S., & Shen, Y. K. (2022). Evaluations of Antibacterial Properties of *Zingiber purpureum* Essential Oil Against 13 Different Gram-positive and Gram-negative Bacteria. *Indonesian Biomedical Journal*, *14*(3), 303–308. https://doi.org/10.18585/inabj.v14i3.1967

Tanweer, S., Shehzad, A., Ahmed, W., & Sciences, A. (2018). Compositional and mineral profiling *of zingiber officinale*. *Pakistan Journal of Food Science 24*(1), 21–26.

Teja, B. S., Jamwal, G., Gupta, V., Verma, M., Sharma, A., Sharma, A., & Pandit, V. (2025). Biological control of bacterial leaf blight (BLB) in rice–A sustainable approach. *Heliyon*, *11*(2), e41769. https://doi.org/https://doi.org/10.1016/j.heliyon.2025.e41769

Thepbandit, W., Papathoti, N. K., Daddam, J. R., Hoang, N. H., Le Thanhn, T., Saengchan, c., & buensanteai, K. (2023). In vitro and in silico studies of salicylic acid on systemic induced resistance against bacterial leaf blight disease and enhancement of crop yield. *Journal of Integrative Agriculture*, *22*(1), 170–184. https://doi.org/https://doi.org/10.1016/j.jia.2022.08.112

Vishakha, K., Das, S., Das, S. K., Banerjee, S., & Ganguli, A. (2022). Antibacterial, anti-biofilm, and anti-virulence potential of tea tree oil against leaf blight pathogen *Xanthomonas oryzae* pv. *oryzae* instigates disease suppression. *Brazilian Journal of Microbiology : [Publication of the Brazilian Society for Microbiology]*, *53*(1), 19–32. https://doi.org/10.1007/s42770-021-00657-2

Yang, X., Zhang, X., Yang, S. P., & Liu, W. Q. (2013). Evaluation of the antibacterial activity of patchouli oil. *Iranian Journal of Pharmaceutical Research*, *12*(3), 307–316.

Ye, T., Li, Y., Zhang, J., Hou, W., Zhou, W., Lu, J., Li, X. (2019). Nitrogen, phosphorus, and potassium fertilization affects the flowering time of rice (*Oryza sativa* L.). *Global Ecology and Conservation*, *20*, e00753. https://doi.org/https://doi.org/10.1016/j.gecco.2019.e00753

Yu, C., Nguyen, D.-P., Ren, Z., Liu, J., Yang, F., Tian, F., Chen, H. (2020). The RpoN2-PilRX regulatory system governs type IV pilus gene transcription and is required for bacterial motility and virulence in *Xanthomonas oryzae* pv. *oryzae*. *Molecular Plant Pathology*, *21*(5), 652–666. https://doi.org/10.1111/mpp.12920

Zhang, Q., Zhang, J., Zhang, Y., Sui, Y., Du, Y., Yang, L., & Yin, Y. (2024). Antifungal and anti-biofilm activities of patchouli alcohol against *Candida albicans*. *International Journal of Medical Microbiology*, *314*, 151596. https://doi.org/https://doi.org/10.1016/j.ijmm.2023.151596