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RESEARCH PAPER

Efficacy analysis of nano-insecticides from essential oils in controlling *Sitophilus oryzae* L. (Coleoptera: Curculionidae) in stored rice

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ABSTRACT

This study evaluated nano-insecticides formulated from essential oils of mint (*Mentha piperita*), lemongrass (*Cymbopogon citratus*), basil (*Ocimum basilicum*), and eucalyptus (*Eucalyptus* sp.) for controlling *Sitophilus oryzae*, a major pest of stored rice. Essential oils were extracted using hydrodistillation, and nanoemulsions were prepared via a low-energy method. Efficacy tests conducted at concentrations of 1–5% showed that mint and lemongrass nano-insecticides were the most effective, achieving 100% mortality at 5% concentration within 168 hours and exhibiting superior repellency (mint oil reached 100% repellency at 5%). At 5% concentration, mint oil caused 25% mortality after 24 hours, increasing to 86% by 96 hours and reaching 100% by 120 hours, while lemongrass oil induced 10% mortality at 24 hours, 76% by 96 hours, and 100% by 144 hours. Their high efficacy is attributed to bioactive compounds such as menthol and citral, which disrupt the pest's physiological processes. In contrast, eucalyptus and basil oils demonstrated comparatively lower mortality and repellency. These results highlight the potential of mint and lemongrass nano-insecticides as eco-friendly alternatives to synthetic pesticides and provide a sustainable option for integrated pest management in stored rice.

Keywords: Biochemical, mortality, nanotechnology, repellency, sustainability

INTRODUCTION

Rice is a crucial staple food in Indonesia, and its proper storage is essential to ensure continuous availability for consumption. However, stored rice is often threatened by storage pests. *Sitophilus oryzae* L. (Coleoptera: Curculionidae) is one of the most significant pests infesting stored rice (Demis & Yenewa, 2022). The presence of this storage pest can severely degrade grain quality. According to Gowda et al. (2019), *S. oryzae* can reduce rice quality by 10–65% under standard storage conditions, with damage reaching up to 80% during prolonged storage. Similarly, Pramono et al. (2018), who observed that the degree of seed damage by *Sitophilus* infestation in sorghum genotypes was closely associated with seed physical and chemical characteristics, such as hardness, pericarp thickness, and phenolic content. Such substantial losses necessitate effective control measures to suppress pest populations.

Synthetic fumigants such as methyl bromide and phosphine are commonly used in pest management (Singh & Sharma, 2015). However, their improper use can lead to environmental pollution and oxic residues on stored grains, posing serious health risks. Therefore, the development of environmentally friendly pest control methods is urgently needed (Rajmohan et al., 2020).

Among emerging alternatives, nano-insecticides have gained attention due to their enhanced efficacy and reduced environmental impact compared to conventional pesticides. Nano-insecticides utilize nanotechnology to improve the delivery, stability, and performance of active ingredients, offering advantages such as controlled release and higher bioavailability. This approach allows targeted pest control using lower quantities of active compounds, thereby minimizing non-target effects and environmental contamination

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(Modafferi et al., 2024). Nano-formulations can also overcome limitations of conventional pesticides, including rapid degradation and poor solubility (de Oliveira et al., 2014).

One promising strategy is the use of botanical insecticides, which provide a sustainable and eco-friendly alternative to synthetic chemicals. Derived from natural plant sources, botanical insecticides can effectively manage pest populations while reducing environmental pollution (Laxmishree & Nandita, 2017). Ngegba et al. (2022) reported that botanical insecticides help mitigate environmental contamination caused by excessive synthetic pesticide use and lower the risk of toxic residues in food, thus preserving ecological balance. As consumers and regulators increasingly demand safer agricultural practices, the development and application of botanical insecticides have become an attractive solution. These natural insecticides, obtained from various plant parts such as roots, stems, leaves, and fruits, have demonstrated efficacy against storage pests (Dougoud et al., 2019; Isman, 2020).

Trivedi et al. (2018) indicated that botanical insecticides could reduce storage pest populations by up to 90%. For example, lemongrass (*Cymbopogon citratus*) produces essential oil through steam distillation, containing citronellal and geraniol (81.67%) (Jeet et al., 2023). Citronellal acts as a respiratory toxin and inhibits the enzyme acetylcholinesterase, leading to insect mortality (Johnson et al., 2021). Similarly, basil (*Ocimum basilicum*) essential oil contains compounds such as camphor, citral, linalool, phenolics, and methyl trans-cinnamate, which act synergistically as insecticidal agents (Chaturvedi et al., 2018). The insecticidal activity of basil oil is largely attributed to its phenolic compounds, which disrupt insect growth and development (Ullah et al., 2014).

Despite their potential, botanical insecticides face challenges such as volatility, rapid degradation, and inconsistent efficacy under varying environmental conditions (Isman, 2020). The integration of nanotechnology with botanical insecticides has been proposed to address these limitations. Nano-encapsulation of essential oils enhances their stability and protects active compounds from degradation. Studies have shown that nano-formulated botanical insecticides exhibit improved penetration through insect cuticles and higher mortality rates than conventional formulations, allowing lower dosages and reduced application frequency (Modafferi et al., 2024).

The effectiveness and stability of essential oils can therefore be significantly improved through nano-insecticide formulations (Zannat et al., 2021). Nanotechnology enables encapsulation of active ingredients into nanosized particles, improving delivery efficiency and reducing harm to non-target organisms (Thakur et al., 2022). Several studies have confirmed the high insecticidal activity of such formulations. For instance, eucalyptus oil nanoemulsions combined with aqueous extracts of *Karanja* and *Jatropha* (300–1500 ppm) achieved 88–100% mortality of *Tribolium castaneum* adults within 24 hours, while *Pimpinella anisum* essential oil nanoemulsion (81.2% (E)-anethole) recorded an LC_{50} of 9.3% v/v and significantly reduced *T. castaneum* progeny. Likewise, neem oil nanoemulsions were 85–100% effective against *S. oryzae* and 74–100% against *T. castaneum*. *Mentha spicata* and *M. pulegium* nanoformulations caused 95.47% and 86.03% mortality, respectively, while *Hazomalania voyronii* essential oil nanoemulsions caused 92.1–100.0% mortality of *T. confusum*, *T. castaneum*, and *Tenebrio molitor* adults after 7 days. Similarly, *Achillea biebersteinii* essential oil nanoemulsion at 10 µL/L air achieved 100% mortality of second-instar *T. castaneum* larvae within 4 days (Jasrotia et al., 2022).

This study investigates the effects of nano-insecticides derived from lemongrass, eucalyptus, basil, and mint essential oils on the development of *S. oryzae*. The objectives are to determine the optimal concentrations for maximum pest control efficacy and to evaluate their potential as sustainable pest management strategies. The findings are expected to support the development of eco-friendly storage pest control methods that minimize environmental and health risks while maintaining rice quality during storage.

MATERIALS AND METHODS

Sample Collection. Fresh leaves of lemongrass (*Cymbopogon citratus*), eucalyptus (*Eucalyptus* sp.), basil (*Ocimum basilicum*), and mint (*Mentha piperita*) were obtained from a local market. Leaves were oven-dried at 40 ºC until reaching a moisture content of 12%. Drying time varied depending on the leaf type.

Isolation of Essential Oils. Dried leaves (200 g per species) were subjected to hydrodistillation for 4 hours using a Clevenger-type apparatus at 60 ºC. Essential oil from three replicates was dried with anhydrous sodium sulfate, filtered, and stored at 4 ºC until use (Abdellatif & Hassani, 2015).

Preparation of Nanoemulsions. Nanoemulsions were prepared using a low-energy emulsification method. Each formulation consisted of 90% (w/v) distilled water, 5% (w/v) essential oil, and 5% (w/v) Polysorbate 80 (Tween 80), totaling 50 g. Essential oils and surfactant were mixed and stirred at 800 rpm for 30 min, followed by gradual addition of water at 3.5 mL/min while maintaining constant stirring for 60 min. The resulting nanoemulsion was stored at room temperature (25 ± 2 ºC) (Feng et al., 2020).

Insect Rearing. Test insects (*S. oryzae*) were obtained from the Pest Control Technology Laboratory, University of Jember. Adults were reared on organic rice in a ventilated plastic containers (14 × 14 × 7.5 cm) covered with a 100-mesh screen. Adults were allowed to oviposit for three days, after which they were removed. Containers were maintained under controlled conditions for five weeks until emergence of first-generation adults, ensuring uniform age for testing (Zhou & Wang, 2016).

Efficacy Test. The insecticidal efficacy of nano essential oils was evaluated following Sabbour (2020). Nanoemulsions were tested at concentrations of 1%, 2%, 3%, 4%, and 5%. Each treatment consisted of 10 adult *S. oryzae* and 30 g of rice placed in a test box. The nanoemulsion was applied using a foam applicator via the drop method. Mortality was recorded at 24, 48, 72, 96, 120, 144, and 168 hours after application (HAA).

Repellency Test. Repellency test for *S. oryzae* adults involved placing the insects in a container filled with rice as a food source. Nano insecticide was applied to a foam pad in the center of the test box, which was then sealed. The infested *S. oryzae* were exposed to nano insecticide treatments at 1%, 2%, 3%, 4%, and 5%, applied to the foam applicator. The control setup included 30 g of rice and 10 adult insects in a plastic container without the insecticide. Observations were conducted at 1, 3, 6, 9, 12, 15, and 18 hours post-infestation. Repellency was measured based on the emigration rate, defined as the percentage of *S. oryzae* that moved away from the treated area relative to the total number of insects present (Ouabou et al., 2024).

Experimental Design and Data Analysis. The study followed a completely randomized design (CRD) with a factorial arrangement. The first factor was the type of nano essential oil (lemongrass, eucalyptus, basil, and mint), and the second was concentration (0%, 1%, 2%, 3%, 4%, and 5%), resulting in 24 treatments with three replications (72 units total). Mortality (%) was calculated using the formula:

$$M(\%) = \frac{Nt}{Nc} \times 100$$

M = Mortality (%);
N_t = Number of dead insects in the treatment;
N_c = Number of dead insects in the control.

Mortality and repellency data were analyzed using ANOVA, and treatment means were compared using Duncan’s Multiple Range Test (DMRT) at a 5% significance level. The repellency test followed the same CRD design and replications, with repellency effectiveness categorized according to Germinara et al. (2015) into six classes ranging from weak (<0.1%) to extreme repellency (80.1–100%). Repellency data were also analyzed using ANOVA, and significant differences were further examined using DMRT at the 5% significance level, providing a comprehensive assessment of the insecticidal and repellent efficacy of the nano essential oil formulations against S. oryzae. The analysis was performed using the DSAASTAT version 1.101 program.

RESULTS AND DISCUSSION

Mortality of *S. oryzae*. The mortality of *S. oryzae* increased proportionally with nano-insecticide concentration and exposure time. Among the four formulations, mint and lemongrass oils consistently caused the highest mortality rates across all observation periods (24–168 hours) (Figures 1–4), indicating stronger insecticidal potency than eucalyptus and basil oils.

At a 5% concentration, eucalyptus oil induced gradual mortality from 3% at 24 hours to 90% at 168 hours (Figure 1). This progressive increase highlights its potential as a nano-insecticide, although its overall effectiveness was lower compared to mint and lemongrass oils. Based on the DMRT post hoc test, the 5% eucalyptus oil treatment began to show a significant difference at 48 hours after application. A similar pattern was observed at 168 hours, where the 5% concentration resulted in the highest mortality rate, significantly different from the other treatments. The steady increase in mortality suggests that while eucalyptus oil is effective, it may require longer exposure or higher concentrations to achieve mortality rates comparable to the more potent oils. Its lower effectiveness may be attributed to the specific chemical composition of eucalyptus oil, which might be less toxic to *S. oryzae*.

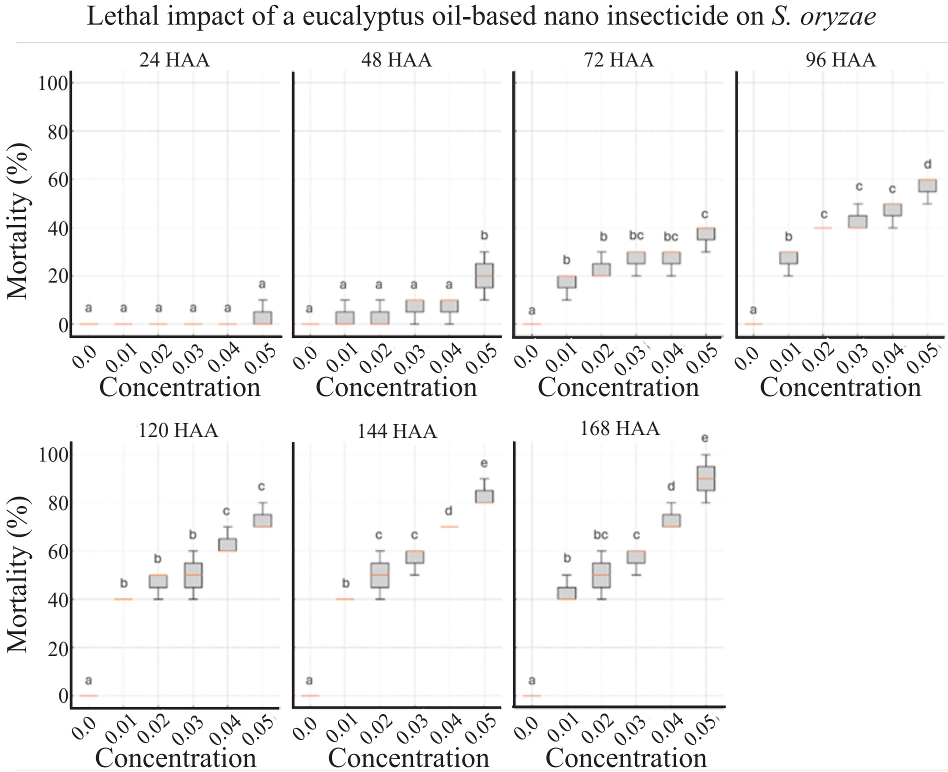

Figure 1. Mortality of *S. oryzae* after treatment with a eucalyptus oil-based nano-insecticide. HAA = Hours after application. Different letters above the boxplots indicate statistically significant differences among treatments at the same time point, as determined by the DMRT post hoc test at a 95% confidence level.

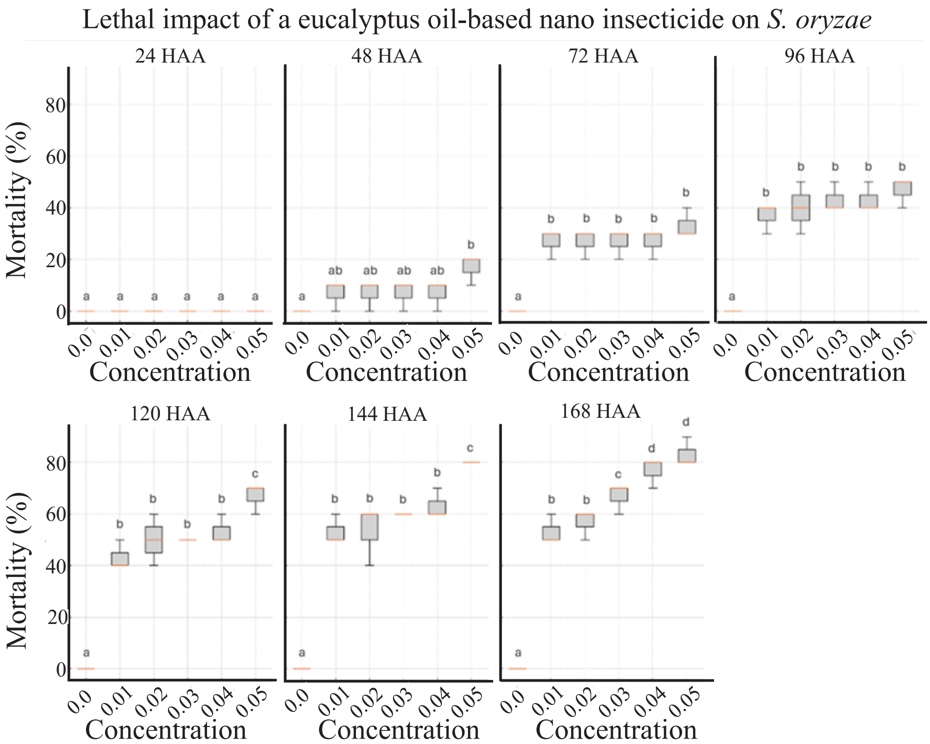

Figure 2. Mortality of *S. oryzae* after treatment with a basil oil-based nano-insecticide. HAA = Hours after application. Different letters above the boxplots indicate statistically significant differences among treatments at the same time point, as determined by the DMRT post hoc test at a 95% confidence level.

Similarly, the basil oil nano-insecticide at a 5% concentration induced mortality rates of 0%, 16%, 33%, 46%, 66%, 80%, and 83% at the respective observation times (Figure 2). According to the DMRT post hoc test, the 5% concentration showed no significant difference from all treatments at 24 hours after application. At 48 hours, concentrations of 1–4% did not differ significantly from the control, while the 5% concentration differed significantly from the control but not from the lower concentrations. By 168 hours post-treatment, the 4% and 5% concentrations were statistically similar, although the 5% treatment produced a higher average mortality rate. Despite the significant increase in mortality over time, basil oil was less effective than mint and lemongrass oils. The initial lack of impact at 24 hours suggests that basil oil has a slower mode of action, requiring longer exposure to affect the pests. However, the gradual increase in mortality indicates that it possesses insecticidal properties that become more pronounced with extended exposure.

In contrast, the 5% mint oil nano-insecticide produced much higher mortality rates—25%, 46%, 66%, 86%, and 100% at 120, 144, and 168 hours post-application (Figure 3). The effectiveness of mint oil was evident as early as 24 hours after application. The DMRT post hoc test showed that the 5% concentration performed significantly better than the control and other treatments. This trend persisted, and by 168 hours, concentrations of 2–5% achieved 100% mortality, indicating that even a 2% concentration was highly effective. These results demonstrate the superior efficacy of mint oil in controlling *S. oryzae*, achieving complete mortality within a shorter period. The rapid increase in mortality suggests that the active compounds in mint oil are highly toxic to *S. oryzae*. The consistent 100% mortality observed at later intervals underscores its potential as a highly effective nano-insecticide. Mint oil's potent insecticidal properties make it a promising candidate for integrated pest management programs.

Similarly, at a 5% concentration, lemongrass oil resulted in high mortality rates of 10%, 36%, 53%, 76%, 83%, and 100% at 144 and 168 hours, respectively (Figure 4). Lemongrass oil exhibited effectiveness from 24 hours after application, with the 4% and 5% concentrations performing significantly better than other treatments based on the DMRT test. This pattern continued, and by 168 hours, the 5% concentration achieved the highest mortality, significantly differing from the control, 1%, 2%, and 3% treatments, but not from the 4% treatment. The strong insecticidal activity of lemongrass oil, particularly its ability to achieve 100% mortality, highlights its potential for effective

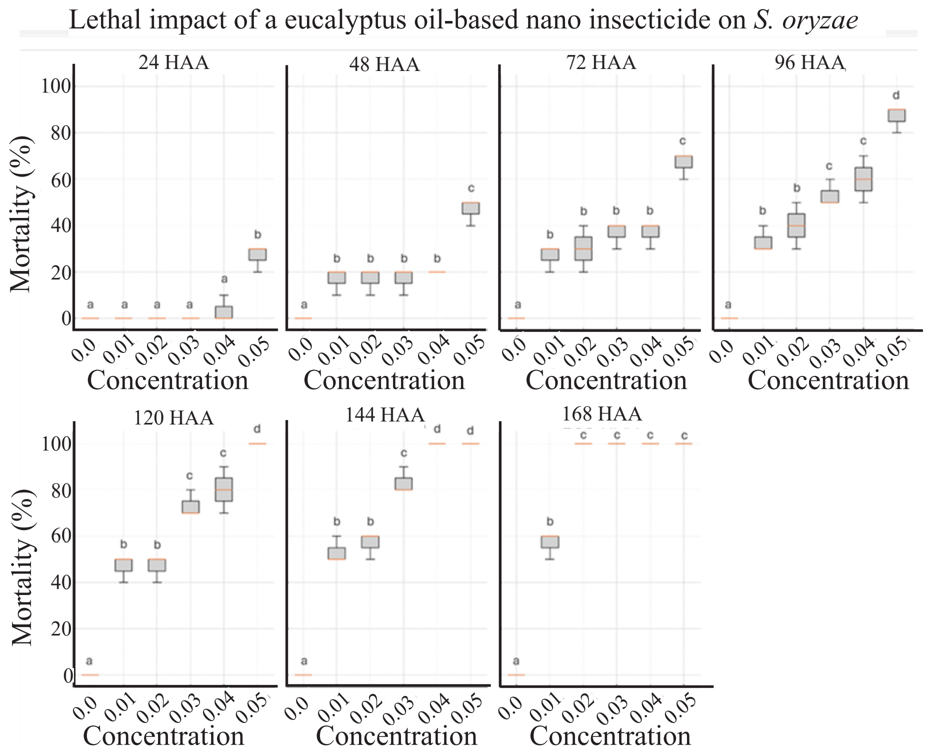

Figure 3. Mortality of *S. oryzae* after treatment with a mint oil-based nano-insecticide. HAA = Hours after application. Different letters above the boxplots indicate statistically significant differences among treatments at the same time point, as determined by the DMRT post hoc test at a 95% confidence level.

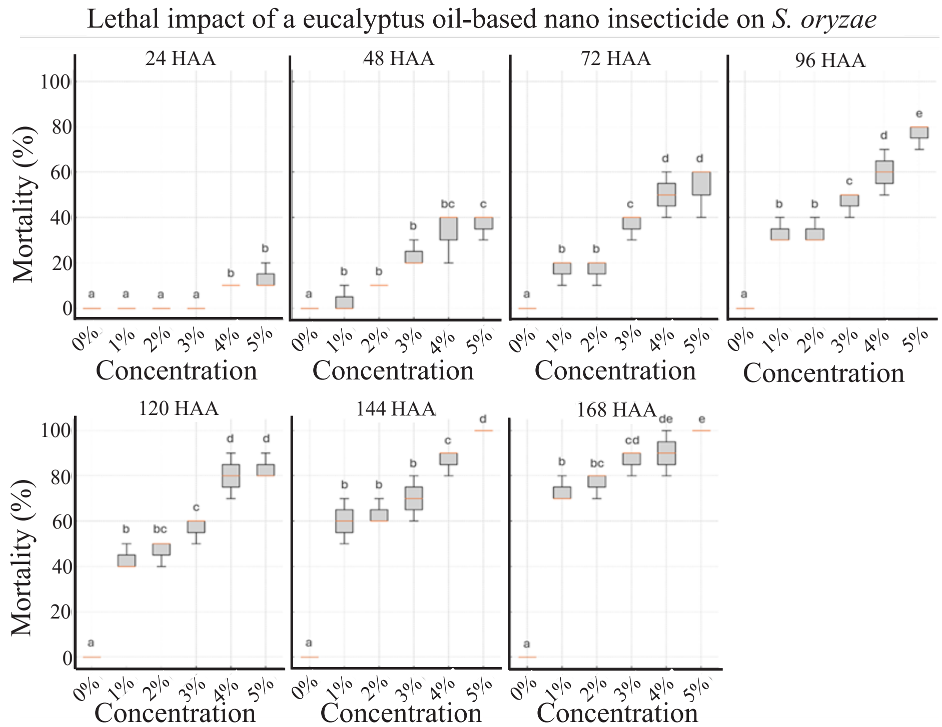

Figure 4. Mortality of *S. oryzae* following treatment with a lemongrass oil-based nano-insecticide. HAA = Hours after application. Different letters above the boxplots indicate statistically significant differences among treatments at the same time point, as determined by the DMRT post hoc test at a 95% confidence level.

pest control. The gradual yet significant increase in mortality over time indicates that lemongrass oil can substantially reduce pest populations with prolonged exposure. The high mortality rates observed with lemongrass and mint oils suggest their strong potential for use in sustainable pest management strategies.

Effect of Oil Type and Concentration on the Mortality of *S. oryzae*. Table 1 presents a detailed analysis of the interaction between oil type and concentration on the mortality rate of *S. oryzae* after 168 hours of application. Both factors significantly influenced mortality rates, with higher concentrations generally resulting in greater mortality across all oil types. At a 0% concentration, none of the oils affected *S. oryzae*, resulting in 0% mortality. However, at 1%, a notable increase was observed, with lemongrass oil showing a significantly higher mortality rate (73.33%) compared to eucalyptus, basil, and mint oils, which ranged between 53.33% and 56.66%.

The interaction between oil type and concentration becomes more pronounced at higher concentrations. At 2%, mint oil achieved 100% mortality, significantly higher than the 56.66% observed for both eucalyptus and basil oils, and higher than the 76.66% for lemongrass oil. This trend continued at 3% and 4%, where mint oil consistently produced 100% mortality, while lemongrass, eucalyptus, and basil oils showed incremental increases. At 5%, lemongrass oil reached 100% mortality, whereas eucalyptus and basil oils peaked at 90% and 83.33%, respectively. The DMRT statistical grouping indicated that lemongrass and mint oils were significantly more effective at higher concentrations than eucalyptus and basil oils. These results demonstrate a clear interaction effect between oil type and concentration on the mortality of *S. oryzae*.

The insecticidal properties of essential oils derived from plants such as mint, lemongrass, basil, and eucalyptus can be attributed to their disruption of insects' physiological and biochemical processes. Mint oil, for instance, contains menthol, a monoterpene known for its neurotoxic effects on insects (Gad et al., 2022). Menthol inhibits acetylcholinesterase (AChE), an enzyme essential for nerve function, by preventing the breakdown of acetylcholine, a neurotransmitter. This inhibition causes continuous nerve stimulation, leading to paralysis and death, which explains the high mortality observed shortly after application (Rants'o et al., 2022; Topal, 2019). Additionally, menthol disrupts the octopaminergic system, which regulates vital functions such as movement and feeding. Interference with octopamine receptors leads to loss of coordination, impaired feeding behavior, and eventual death, further enhancing the efficacy of mint oil as an insecticide (Hikal et al., 2017; Zeni et al., 2021).

Table 1. Effect of different essential oil types and concentrations on *S. oryzae* mortality 168 hours after application

Concentration (%)	Mortality (%)			
	Lemongrass oil	Eucalyptus oil	Basil oil	Mint oil
0	0.00 ± 0.00 (a)	0.00 ± 0.00 (a)	0.00 ± 0.00 (a)	0.00 ± 0.00 (a)
	A	A	A	A
1	73.33 ± 0.58 (b)	53.33 ± 0.58 (b)	53.33 ± 0.58 (b)	56.66 ± 0.58 (b)
	B	B	B	B
2	76.66 ± 0.58 (bc)	56.66 ± 1.00 (bc)	56.66 ± 0.58 (b)	100.00 ± 0.00 (c)
	C	C	C	C
3	86.66 ± 0.58 (cd)	66.66 ± 0.58 (c)	66.66 ± 0.58 (c)	100.00 ± 0.00 (c)
	D	D	D	D
4	90.00 ± 1.00 (de)	73.33 ± 0.58 (d)	76.66 ± 0.58 (d)	100.00 ± 0.00 (c)
	E	E	E	E
5	100.00 ± 0.00 (e)	90.00 ± 1.00 (e)	83.33 ± 0.58 (d)	100.00 ± 0.00 (c)
	F	F	F	F

Values followed by the same letter are not significantly different according to Duncan's Multiple Range Test (DMRT) at the 5% significance level. Uppercase letters indicate comparisons across rows, while lowercase letters indicate comparisons within columns.

Lemongrass oil owes its insecticidal properties mainly to its major components, citral (α-citral and β-citral), which induce oxidative stress in insects. This oxidative damage impairs cellular function, contributing to insect mortality over time (Ahmad et al., 2016; Moustafa et al., 2023). The volatile nature of lemongrass oil also allows it to interfere with the respiratory system of insects, as citral and other components can block spiracles, causing suffocation. This respiratory inhibition is particularly effective in nano-formulations, which ensure prolonged exposure and sustained release of active compounds (Sahu et al., 2021). Furthermore, components such as geranyl acetate inhibit detoxification enzymes including carboxylesterase (CarE) and glutathione S-transferase (GST), preventing the detoxification of toxic components and enhancing mortality (Awad et al., 2024; Moustafa et al., 2021).

Basil oil contains several bioactive compounds, primarily linalool and eugenol, which are responsible for its insecticidal activity. Similar to menthol, these compounds disrupt the insect nervous system by inhibiting AChE, causing continuous nerve stimulation, paralysis, and death (da Silva et al., 2020; Moretti et al., 2017). The aromatic properties of basil oil, mainly due to eugenol, also contribute to its repellent activity by interfering with olfactory receptors, deterring insects from treated areas and reducing feeding and reproduction. Additionally, basil oil can disrupt hormonal balance, particularly by affecting juvenile hormone levels, leading to developmental abnormalities and reduced reproductive capacity (Devrnja et al., 2024; Ismail EM & El-Gawad, 2018).

Eucalyptus oil, particularly from Eucalyptus globulus, contains eucalyptol (1,8-cineole) as its main active compound. Eucalyptol is highly volatile and effective as a fumigant, allowing it to diffuse through storage environments and act via inhalation (Arooj et al., 2023). This mode of action is advantageous for controlling pests in stored grain systems. Eucalyptol can block spiracles, causing suffocation, and this respiratory inhibition is enhanced when eucalyptus oil is used in nano-formulations that allow sustained release (Karabörklü & Ayvaz, 2023). Furthermore, eucalyptus oil inhibits detoxification enzymes such as GST, reducing the insect's ability to metabolize and neutralize toxic compounds, thereby increasing mortality (Pavlidi et al., 2018).

Repellency Effects of Botanical Nano Insecticides on *S. oryzae*. Based on Figure 5, which presents the effects of nano botanical insecticides from various essential oils on the repellency levels against *S. oryzae* at different concentrations and application times, significant differences were observed in the effectiveness of each oil. In the early hours post-application (1 and 3 HAA), all oils showed increasing repellency with rising concentrations. Nano insecticides derived from lemongrass and mint oils demonstrated a more consistent increase in repellency than those from basil and eucalyptus oils, particularly at lower concentrations.

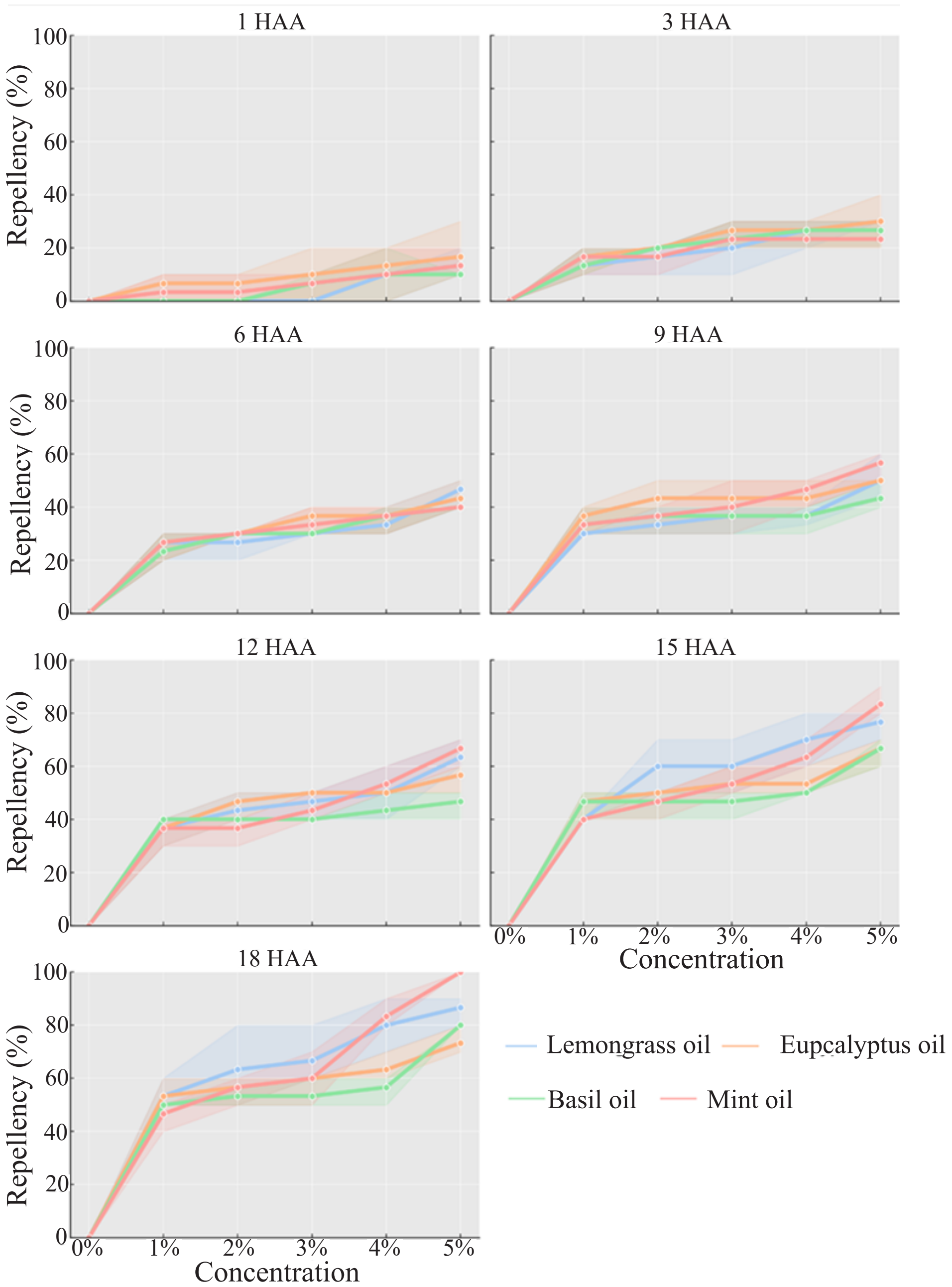

Figure 5. Repellency effect of nano-insecticide concentrations of lemongrass, eucalyptus, basil, and mint essential oils against *S. oryzae* at 1, 3, 6, 9, 12, 15, and 18 hours after application (HAA).

During the mid-observation period (6–12 HAA), the differences among oils became more pronounced. Nano-insecticides from mint oil exhibited a stable and generally higher increase in repellency compared to the others at the same concentrations. Lemongrass oil also showed promising results, though slightly lower than mint oil. Conversely, nano insecticides from basil and eucalyptus oils displayed fluctuating repellency levels, with occasional declines, suggesting their effects diminished more rapidly than those of lemongrass and mint oils.

In the final observation period (15–18 HAA), nano-insecticides from mint oil consistently demonstrated the highest repellency levels among all oils tested against *S. oryzae*. Lemongrass oil also maintained strong repellency, although slightly lower than mint oil. Meanwhile, basil and eucalyptus oils showed sharper declines, particularly at lower concentrations. Overall, these data indicate that nano insecticides derived from mint and lemongrass oils exhibit stronger and more persistent repellency than those from basil and eucalyptus oils.

Table 2 presents the effects of oil type and concentration on the repellency of *S. oryzae* after 18

Table 2. Effect of different essential oil types and concentrations on the repellency of *S. oryzae* 18 hours after application

Concentration (%)	Repelency (%)			
	Lemongrass oil	Eucalyptus oil	Basil oil	Mint oil
0	0.00 ± 0.00 (a) A	0.00 ± 0.00 (a) A	0.00 ± 0.00 (a) A	0.00 ± 0.00 (a) A
1	50.33 ± 0.58 (b) B	50.33 ± 0.58 (b) B	50.00 ± 0.00 (b) B	40.67 ± 0.58 (b) B
2	60.33 ± 1.53 (bc) C	50.67 ± 0.58 (bc) C	50.33 ± 0.58 (b) C	50. 67 ± 0.58 (bc) C
3	60.67 ± 1.15 (bc) D	60.00 ± 0.00 (bc) D	50.33 ± 0.58 (b) D	60.00 ± 1.00 (c) D
4	80.00 ± 1.00 (cd) E	60.33 ± 0.58 (c) E	50.67 ± 0.58 (b) E	80.33 ± 0.58 (d) E
5	80.67 ± 0.58 (d) F	70.33 ± 0.58 (d) F	80.00 ± 0.00 (c) F	100.00 ± 0.00 (e) F

Values followed by the same letter are not significantly different according to Duncan's Multiple Range Test (DMRT) at the 5% significance level. Uppercase letters indicate comparisons across rows, while lowercase letters indicate comparisons within columns.

hours of application. At a 0% concentration, none of the oils showed repellency (0%). At 1%, the repellency rates were 50.33% (lemongrass), 50.33% (eucalyptus), 50.00% (basil), and 40.67% (mint). Increasing the concentration to 2% enhanced repellency to 60.33% (lemongrass), 50.67% (eucalyptus), 50.33% (basil), and 50.67% (mint). At 3%, the repellency rates were 60.67% (lemongrass), 60.00% (eucalyptus), 50.33% (basil), and 60.00% (mint). At 4%, lemongrass and mint oils showed marked increases to 80.00% and 80.33%, respectively, while eucalyptus and basil reached 60.33% and 50.67%. At the highest concentration (5%), repellency reached 80.67% (lemongrass), 70.33% (eucalyptus), 80.00% (basil), and 100% (mint), indicating a strong positive correlation between oil concentration and repellency for all oil types.

The heatmap (Figure 6) further illustrates the repellency effectiveness of various oil-based nano insecticides during rice storage. Eucalyptus oil at 5% demonstrated the highest repellency with a Class 5 index and 100% repellency rate. Lemongrass and mint oils also showed high repellency at higher concentrations—lemongrass achieving Class 4 at 3% and 5%, and mint reaching Class 5 at 5%. Basil oil exhibited lower repellency, with a maximum Class 4 index (80%) at 5%. Overall, higher concentrations yielded greater repellency, with eucalyptus and mint oils particularly effective at higher levels.

The significant repellency effects observed can be attributed to the specific modes of action of essential oils used in the nano insecticides. Essential oils contain volatile compounds that disrupt insect olfactory receptors, leading to olfactory confusion and deterrence (Jankowska et al., 2017). Monoterpenes and sesquiterpenes emit strong odors that repel insects, preventing them from approaching treated areas. For instance, menthol (in mint oil) and citral (in lemongrass oil) are especially effective in causing immediate and prolonged repellency (Devrnja et al., 2022).

Neurotoxic effects also contribute significantly to repellency. Essential oils inhibit acetylcholinesterase (AChE), leading to continuous nerve impulses and paralysis. Although this directly causes mortality, the initial neurotoxic response also induces disorientation and avoidance behavior, increasing repellency (Liu et al., 2021). Additionally, essential oils possess antifeedant properties that make treated surfaces unattractive, reducing feeding and reproduction. These compounds interfere with chemoreceptors involved in taste and smell, creating an unfavorable environment for insects (Jankowska et al., 2017).

Nano-formulation further enhances the stability and persistence of essential oils, allowing sustained release of active compounds over time. Nanoparticles adhere to surfaces and release volatile components gradually, resulting in long-lasting repellency (Palermo et al., 2021). This explains the strong and persistent repellency observed, particularly for mint and lemongrass oils, whose key compounds—menthol and citral—are known for their potent olfactory and

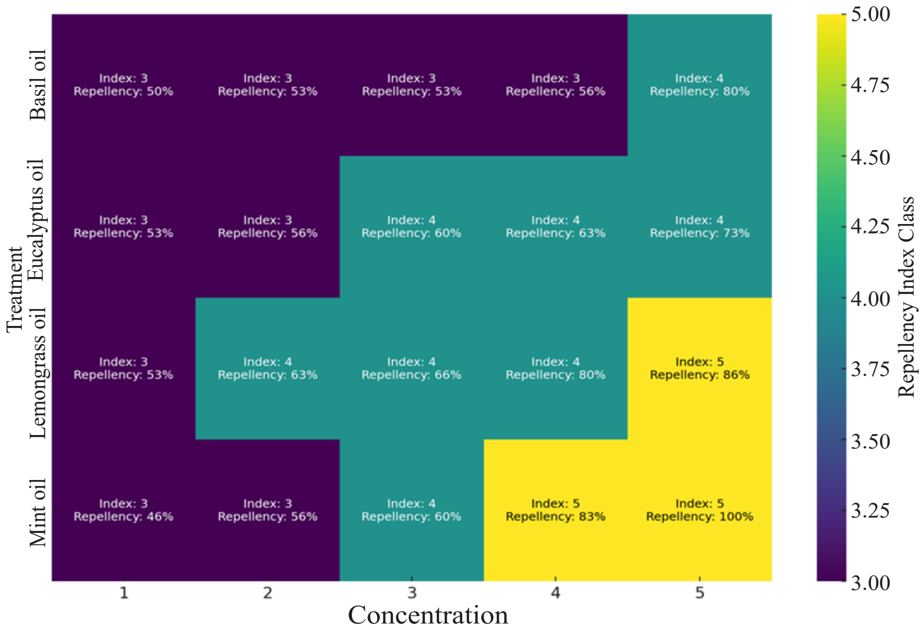

Figure 6. Repellency index categories of various essential oil-based nano-insecticides against *S. oryzae.*

neurotoxic properties (Abdelgaleil et al., 2021; Zhang et al., 2022).

While nano-insecticides demonstrated strong repellency, further research is needed to assess their long-term stability under varying environmental conditions. Factors such as temperature, light exposure, and oxygen may alter nanoparticle size or degrade active compounds, reducing efficacy (Modafferi et al., 2024). Likewise, differences in storage conditions, humidity, dust, and grain composition could influence formulation persistence and distribution. Testing performance under diverse environmental scenarios remains essential to ensure consistent repellency and mortality outcomes (Jasrotia et al., 2022; Zannat et al., 2021).

CONCLUSION

This study demonstrates that nano-insecticides derived from mint and lemongrass essential oils are highly effective against *S. oryzae* in stored rice, achieving 100% mortality at a 5% concentration within 168 hours, along with excellent repellency. Eucalyptus and basil oils were less effective. These findings support the potential of essential oil-based nano-insecticides as sustainable and eco-friendly alternatives to synthetic pesticides, offering a promising solution for protecting stored rice. Future research should explore large-scale applications of these nano-insecticides in actual storage facilities, including their compatibility with existing storage practices, long-term stability, and potential effects on rice quality.

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AUTHORS' CONTRIBUTIONS

MH, AF, and APP performed the experiments and wrote the manuscript. NTH and SW designed the research and prepared the materials. FKA and MA co-authored the manuscript and conducted statistical analyses. All authors contributed to discussions, data interpretation, and manuscript revision. All authors have read and approved the final version of the manuscript.

COMPETING INTEREST

The authors declare no known competing financial interests or personal relationships that could have influenced the work reported in this paper.

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