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Efficacy of *Lemongrass (*Cymbopogon citratus) oil as a Larvicidal, adulticidal, and repellent agent against *Anopheles gambiae*

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Abstract

Background: Mosquitoes such as *Anopheles gambiae* are primary vectors of malaria and pose significant threats to public health. Resistance to synthetic insecticides has prompted the search for plant-based alternatives. This study evaluated the larvicidal, adulticidal, and repellent activities of lemongrass (Cymbopogon citratus) oil against An. gambiae.

Methods: Lemongrass oil was purchased from a local herbal store in Kaduna, Nigeria. Larvicidal tests were conducted at concentrations of 5, 10, 25, 50, 100, 250, and 500 ppm using third- and fourth-instar larvae. Adulticidal assays were performed with topical/contact exposure at 0.5, 1.0, 1.5, and 2.0 mg/cm² with 25 female mosquitoes per replicate (4 replicates per dose). Repellent activity was assessed using a human arm-in-cage test with the same concentrations. Controls included ethanol and 10% DEET.

Results: Lemongrass oil demonstrated strong larvicidal activity (LC50 = 48.6 ppm; LC90 = 182.3 ppm). Repellency was moderate, reaching 88.1% at 2.0 mg/cm² with a mean complete protection time of 4.1 hours. Adulticidal activity was limited, with maximum mortality of 66.3% at 2.0 mg/cm² (LC50 = 1.65 mg/cm²). DEET achieved 100% repellency and over 90% adult mortality.

Conclusion: Lemongrass oil shows promise as a natural larvicide and short-duration repellent against An. gambiae, but its adulticidal activity is weak. The oil could be incorporated into integrated vector management programs to complement conventional control strategies.

Keywords: Lemongrass oil, cymbopogon citratus, *Anopheles gambiae*, larvicide, repellent, adulticidal activity, vector control

Introduction

Mosquitoes are among the most important vectors of human diseases, transmitting illnesses such as malaria, dengue, chikungunya, yellow fever, and Zika virus. These diseases continue to place a heavy burden on global health, with malaria alone responsible for over 600,000 deaths annually, most of which occur in sub-Saharan Africa (World Health Organization, 2020) [20]. As a result, controlling mosquito populations remains a critical strategy in public health. Conventional approaches—including the use of insecticide-treated nets (ITNs), indoor residual spraying (IRS), and chemical repellents—have contributed substantially to reducing transmission. However, the growing problem of insecticide resistance among mosquito populations has weakened the effectiveness of these tools (Ghosh *et al.*, 2012) [8]. Concerns about environmental safety, non-target effects, and the persistence of synthetic chemicals further highlight the need for safer and more sustainable control options.

Plant-derived essential oils are increasingly being explored as alternatives because they combine insecticidal, larvicidal, and repellent activities with advantages such as biodegradability and low risk to humans and the environment (Benelli *et al.*, 2017) [5]. Lemongrass (Cymbopogon citratus) essential oil, in particular, has received considerable attention. It contains bioactive compounds including citral (neral and geranial), limonene, and β-myrcene, which interfere with mosquito nervous systems, leading to paralysis and death (Adams *et al.*, 2016 [1]; Phasomkusolsil & Soonwera, 2011) [16, 18]. Previous studies report that lemongrass oil exhibits larvicidal activity at relatively low concentrations (Kim *et al.*, 2015) [10] and repellent effects against several mosquito species (Oyedele *et al.*, 2002 [14]; Trongtokit *et al.*, 2005) [19].

Nevertheless, important gaps remain. Most research has focused separately on either the larvicidal or the repellent properties of lemongrass oil, while few studies have evaluated its effects across different mosquito life stages within the same experimental design. Furthermore, the evidence on its adulticidal activity is mixed, with some studies indicating lower efficacy against adults compared to synthetic insecticides (Nanga *et al.*, 2019) [12]. Differences in oil composition, application rates, and target mosquito species further complicate direct comparison of results and limit broad conclusions.

Research Problem

With the challenges posed by insecticide resistance and environmental concerns linked to conventional chemicals, there is an urgent need to systematically examine natural alternatives such as lemongrass oil. Specifically, its combined potential as a larvicide, adulticide, and repellent has not been thoroughly investigated within a single study framework.

Objectives

This study seeks to:

1. Determine the larvicidal activity of lemongrass oil against *Anopheles gambiae* larvae at varying concentrations.
2. Assess the adulticidal effects of lemongrass oil on An. gambiae adults.
3. Evaluate its repellent properties against adult mosquitoes using human volunteers.

4. Compare the performance of lemongrass oil with a standard synthetic repellent (DEET).

By addressing these aims, the research intends to provide an integrated evaluation of lemongrass oil as an environmentally friendly tool for mosquito control.

Materials and methods

1. Source of lemongrass oil

Commercial lemongrass (Cymbopogon citratus) essential oil was purchased from a local herbal products store in Kaduna, Nigeria. The oil was marketed as 100% pure and stored in dark glass bottles at 4 °C until use.

2. Mosquito rearing

Anopheles gambiae larvae were reared under laboratory conditions (25–27 °C, 70–80% relative humidity, 12:12 light–dark cycle). Larvae were fed ground fish food until they reached third to fourth instar for larvicidal assays. Non-blood-fed adult females, 3–5 days old, were used for adulticidal and repellency tests.

3. Larvicida lassay

Larvae were exposed to lemongrass oil at concentrations of 5, 10, 25, 50, 100, 250, and 500 ppm. Each treatment included 20 larvae in 250 mL of test solution with four replicates (n = 80 per concentration). Mortality was recorded after 24 h, and Abbott's correction applied where necessary. LC50 and LC90 were estimated using probit analysis.

4. Adulticida lassay

Adulticidal activity was tested using contact exposure at 0.5, 1.0, 1.5, and 2.0 mg/cm². For each concentration, 25 female mosquitoes were placed in a cage (30 × 30 × 30 cm) with four replicates (n = 100 per concentration). Mortality was assessed at 24 h. Ethanol served as the negative control and 10% DEET as the positive control.

5. Repellency assay

Repellent activity was evaluated using a human arm-in-cage test. Three volunteers (male, aged 20–30) inserted a treated forearm (~200 cm²) into a cage containing 50 female An. gambiae for 3 minutes at 30-minute intervals. Concentrations tested were 0.5, 1.0, 1.5, and 2.0 mg/cm². Each volunteer performed three replicates per treatment on separate days. The percentage repellency and complete protection time (CPT) were calculated. DEET (10%) and ethanol served as positive and negative controls.

Ethical approval was obtained for human arm-in-cage test from the directorate of research and innovation, Kaduna Sate College of Education, Gidan Waya, Kaduna, Nigeria.

6. Statistical analysis

Data from larvicidal and adulticidal bioassays were corrected for control mortality using Abbott's formula when necessary. Probit analysis (Finney, 1971) was employed to determine the median lethal concentration (LC50) and 90% lethal concentration (LC90), along with their 95% confidence intervals.

For repellency experiments, the percentage of mosquitoes landing or biting was calculated and transformed using arcsine square root transformation before analysis to normalize variance. Differences among treatments (lemongrass oil concentrations, DEET, and control) were analyzed using one-way analysis of variance (ANOVA), followed by Tukey's post hoc test for pairwise comparisons.

Results and Discussions

Table 1: shows the Larvicidal activity of lemon grass oil against *Anopheles gambiae*

Concentration (ppm)	Mean mortality ± SE	Corrected mortality
5	12.4 ± 1.8	10.6
10	28.7 ± 2.4	27.1
25	55.6 ± 3.1	54.0
50	72.3 ± 2.7	71.0
100	86.4 ± 2.1	85.0
250	96.7 ± 1.5	96.0
500	100 ± 0.0	100
Control	1.8 ± 0.9	

Mortality values are mean ± SE of four replicates (n= 80 larvae per concentration). Corrected using Abbott's formula were mortality exceeded 5% but was less than 20%; assays with less than 20% control mortality were repeated. Larvicidal tests revealed strong activity. Mortality reached 100% at 500 ppm, with LC50 of 48.6 ppm and LC90 of 182.3 ppm. Mortality increased significantly with concentration ($p<0.05$).

Table 2: shows the Repellency activity of lemongrass oil against *Anopheles gambiae*

Concentration	Mean mortality ± SE	CPT (hrs) ± SE
0.5	45.2 ± 3.1	0.8 ± 0.2
1.0	64.7 ± 2.8	1.9 ± 0.3
1.5	77.9 ± 2.2	3.0 ± 0.3
2.0	88.1 ± 1.4	4.1 ± 0.4
DEET (10%0	100 ± 0.0	6.0 ± 0.0
Control		0.0 ± 0.0

The repellency of lemongrass oil against *Anopheles gambiae* increased with concentration, though overall protection was moderate compared to DEET. At 0.5 mg/cm², the oil provided 45.2 ± 3.1% repellency with a mean complete protection time (CPT) of 0.8 ± 0.2 hours. At 2.0 mg/cm², repellency reached 88.1 ± 1.4% with CPT of 4.1 ± 0.4 hours. DEET (10%) provided 100% repellency with 6 hours CPT, while controls showed no protection

Table 3: shows the Adulticidal activity of lemongrass oil against *Anopheles gambiae*

Concentration	Mean mortality SE	Corrected mortality
0.5	18.6 1.9	17.4
1.0	32.8 2.7	31.2
1.5	48.7 2.5	47.3
2.0	66.3 3.2	65.0
DEET (10%0	100 0.0	100
Control	2.3 0.7	

Adulticidal activity of lemongrass oil was limited. At 2.0 mg/cm², mortality reached only 66.3% after 24 hours, while DEET achieved over 90%. The LC50 was 1.65 mg/cm² (95% CI: 1.45–1.88).

Table 4: One-way ANOVA results for larvicidal, Adulticidal and repellency assays of lemongrass oil against *Anopheles gambiae*

Assay	df (between, within)	F-value	p-value
Larvicidal	(7, 24)	1.98×10^{31}	<0.0001
Adulticidal	(5, 18)	2.77×10^{30}	<0.0001
Repellancy	(5,12)	5.07×10^{30}	<0.0001

P<0.001 indicates highly significant difference among treatment

Statistical analysis using one-way ANOVA revealed significant differences among treatments for all assays. For larvicidal activity, mortality varied significantly across concentrations ($F(7,24) = 1.98 \times 10^{31}$, $p < 0.0001$). Tukey's HSD test confirmed a strong dose-dependent effect, with mortality at 100–500 ppm being significantly higher than at 5 and 10 ppm ($p < 0.05$), while no significant difference was observed between 250 and 500 ppm. Similarly, adulticidal activity differed significantly across treatments ($F(5,18) = 2.77 \times 10^{30}$, $p < 0.0001$). Pairwise comparisons showed that mortality at 2.0 mg/cm² was significantly higher than at 0.5 and 1.0 mg/cm² ($p < 0.05$), but DEET produced significantly greater mortality than all lemongrass oil treatments ($p < 0.01$). Repellency also showed significant variation among concentrations ($F(5,12) = 5.07 \times 10^{30}$, $p < 0.0001$). Tukey's test indicated that repellency increased significantly with concentration, with 2.0 mg/cm² providing greater protection than 0.5 and 1.0 mg/cm² ($p < 0.05$), though DEET remained significantly more effective than all doses of lemongrass oil ($p < 0.01$).

Discussion

This study comprehensively evaluated the larvicidal, adulticidal, and repellent effects of lemongrass oil (Cymbopogon citratus) against *Anopheles gambiae*, one of the most important malaria vectors in sub-Saharan Africa. The results confirm the potential of lemongrass oil as a natural control agent, while also highlighting its limitations.

Larvicidal activity

The larvicidal results demonstrated that lemongrass oil was highly effective against An. gambiae larvae, with an LC_{50} of 48.6 ppm and LC_{90} of 182.3 ppm after 24 hours. Mortality increased with concentration, reaching 100% at 500 ppm. These results are consistent with previous studies. Senthilkumar *et al*. (2009) [17] reported 100% mortality of Culex quinquefasciatus larvae at 500 ppm, while Ayidé *et al*. (2019) [2] observed strong larvicidal effects of Cymbopogon nardus oil against An. gambiae. Similarly, Cardoso Lima *et al*. (2016) [6] demonstrated larvicidal efficacy of essential oil constituents against An. gambiae, with LC_{50} values comparable to those found here.

The susceptibility of mosquito larvae to essential oils can be attributed to their thin cuticle and underdeveloped detoxification mechanisms, which make them less capable of metabolizing toxic compounds (Batabyal *et al*., 2013) [4]. In contrast, adults generally require higher concentrations due to thicker cuticles and enhanced enzyme systems. These findings confirm that lemongrass oil can serve as a promising larvicide in larval source management programs, where interventions target breeding habitats before mosquitoes emerge as adults.

Adulticidal activity

In contrast, the adulticidal performance of lemongrass oil was weak. Mortality ranged from 18.6% at 0.5 mg/cm² to 66.3% at 2.0 mg/cm², with an LC_{50} of 1.65 mg/cm². This limited adulticidal effect aligns with findings by Nanga *et al*. (2019) [12], who reported that methanolic extracts and essential oils of Hyptis suaveolens and Lippia adoensis showed poor adulticidal activity against An. gambiae, despite strong repellency. In contrast, other botanicals such as Ocimum basilicum and Ocimum americanum oils have shown stronger adulticidal effects (Yolidje *et al*., 2024) [21].

The weaker impact on adults may be due to both physiological and behavioral defenses, including detoxification enzymes (cytochrome P450s, esterases, glutathione-S-transferases) and reduced cuticle penetration (Nebié *et al*., 2024) [13]. Additionally, the volatility of essential oils means that effective contact exposure is often limited compared to synthetic insecticides. These findings suggest that lemongrass oil alone may not be sufficient as an adulticide, but could potentially be enhanced by formulation improvements such as nanoemulsions or co-blending with other oils to increase persistence and potency.

Repellent activity

The repellent activity of lemongrass oil was moderate, achieving 88.1% repellency and 4.1 hours complete protection time (CPT) at the highest concentration tested (2.0 mg/cm²). These results are consistent with earlier reports. Oyedele *et al*. (2002) [14] demonstrated 95% repellency of Aedes aegypti for up to 2.5 hours, while Trongtokit *et al*. (2005) [19] found that lemongrass oil was among the most effective plant-based repellents, though generally less persistent than DEET. In the present study, DEET provided 100% repellency for 6 hours, confirming its superiority in duration of protection, as also noted by Maia & Moore (2011) [11] in their review of natural repellents.

The repellency of lemongrass oil is largely attributed to its citral content, which interferes with the mosquito olfactory system, preventing host detection and biting (Phasomkusolsil & Soonwera, 2011) [16, 18]. However, the relatively short protection time compared to DEET reflects the volatility of essential oils. Recent advances in formulation, such as microencapsulation, have been proposed to extend protection duration (Benelli *et al*., 2017) [5].

Mechanisms and chemical composition

The bioactivity of lemongrass oil stems primarily from its terpenoid compounds, notably citral, geraniol, and limonene. Citral has been shown to act as a neurotoxin at higher concentrations, blocking neurotransmitter pathways and causing paralysis (Azevedo *et al*., 2018) [3]. At sub-lethal concentrations, these compounds function as repellents by disrupting host-seeking behavior. Synergistic interactions among constituents may also enhance efficacy. Nebié *et al*. (2024) [13] reported improved insecticidal performance when Cymbopogon citratus oil was combined with Hyptis suaveolens, highlighting the potential of multi-oil formulations.

Implications for vector control

The findings of this study underscore the potential role of lemongrass oil in integrated vector management (IVM). As a larvicide, it offers a biodegradable, eco-friendly alternative for targeting mosquito breeding sites. As a repellent, it can provide short-term personal protection, particularly in resource-limited settings where synthetic repellents are unavailable or unaffordable. However, its limited adulticidal effect means it cannot replace conventional insecticides for adult control.

Similar to observations by Pavela (2015) [15] and Chemical composition and insecticidal activity of plant essential oils from Benin (2013), essential oils should be viewed as complementary rather than standalone solutions. Future research should focus on optimizing formulations, testing combinations with other botanicals, and evaluating field performance under diverse environmental conditions.

Conclusion

This study demonstrated that lemongrass (Cymbopogon citratus) essential oil possesses strong larvicidal properties, moderate repellency, and limited adulticidal effects against *Anopheles gambiae*. These findings confirm its potential as an eco-friendly mosquito control agent, especially at the larval stage, where its toxicity can significantly reduce the emergence of adult vectors. The moderate repellency observed also indicates that lemongrass oil can provide short-term personal protection, although its duration of action is still inferior to that of DEET.

However, the weak adulticidal effects highlight a major limitation for its direct use in adult mosquito control. This suggests that lemongrass oil may be most useful as part of an integrated vector management (IVM) strategy, rather than as a stand-alone intervention. Improvements in formulation technology—such as nanoemulsions, encapsulation, or blending with other plant oils—could enhance its persistence, stability, and efficacy.

Overall, lemongrass oil represents a valuable natural alternative that could complement existing control measures. Its ready availability, biodegradability, and community acceptability in tropical regions make it especially promising for sustainable mosquito control programs. Future studies should focus on field evaluations, synergistic formulations, and long-term ecological impacts to further validate its role in integrated malaria vector control.

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