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Research Article

Mosquito Repellency and Toxicity Effects of *Hyptis suaveolens* (Leaves) Against Culex Mosquito

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ABSTRACT
Human pathogens that are transmitted by insects are a global problem, particularly those vectored by mosquitoes. Mosquitoes are the most important group of insect vectors able to transmit several diseases, including malaria, dengue and filariasis, especially in the local environment, hence the need to find safe, locally available and environmentally benign control measures. *Hyptis suaveolens* is an exotic plant that has high adaptability and tolerance of changes in abiotic factors, with a particularly high presence in the northern regions of Nigeria. This study aimed at analysing the repellency and toxicity effects of *Hyptis suaveolens* on *Culex* mosquitoes. Leaves of *Hyptis suaveolens* were obtained from Katsina-Mani Road. Using the prepared plant extract, phytochemical screening, repellency and toxicity tests were carried out. The result of phytochemical screening revealed the presence of alkaloids, flavonoids, glycosides, reducing sugar, steroids, saponins, and tannins. Total number of mosquitoes landed on the control arm was 15 (60%), followed by arm treated with 20% repellent cream, which was 3 (12%), while none (0%) landed on 40%, 60% and 100% arm treated with repellent cream. The toxicity of *Hyptis suaveolens* leaves against *Culex* mosquitoes was recorded at different concentrations, where the effect of *Hyptis suaveolens* leaf smoke was higher at higher concentrations and lower at lower concentrations. Further, 100% toxicity was obtained at 80 and 100% concentrations. This study revealed that an extract obtained from dried leaves of *Hyptis suaveolens* had the ability to repel adult *Culex* mosquitoes even at low concentration (40%).

Keywords: *Culex* mosquito; *Hyptis suaveolens;* Katsina State; Repellency; Toxicity

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INTRODUCTION

Vector-borne diseases globally are reported to account for more than 17% of infectious diseases (WHO, 2024), with the high incidence and mortality of more than one million people per year dying of malaria, dengue, African trypanosomiasis, leishmaniasis, Chagas diseases, yellow fever, Japanese encephalitis and onchocerciasis (WHO, 2024). Moreover, vector-borne diseases over the years have become emergent or reemergent in different areas, posing an immense threat to health and causing economic losses (Chala and Hamde, 2021). Such diseases can be caused by either parasites, bacteria or viruses which are transmitted by different insects of which the most important and common vectors are mosquitoes (Aedes, Anopheles, Culex), ticks, lice and flies (Ratcliffe *et al*., 2024).

Culex mosquitoes are cosmopolitan vectors found globally across tropical, subtropical, and temperate regions (Uttah *et al*., 2013), different researches across the globe have reported the *Culex* species as the widest spread mosquito species due to their high adaptability to feed on both humans and livestocks (Mohammed *et al*., 2021., Nchoutpouen *et al*., 2019), eventhough much attention and focus have been centered on *Anopheles* mosquitoes. Mosquitoes of the genus *Culex* are primary vectors for several high-consequence viral and parasitic zoonotic diseases

(Madhav *et al*., 2024), among which includes mosquito- borne viruses such as Sindbis virus, West Nile Virus, Equine encephalitis, St Louis encephalitis, Japanese encephalitis, Oro-pouche fever, Avian malaria, Lymphatic filariasis and Rift valley fever (RVF) (Madhav *et al*., 2024). Of the six different members of the *Culex pipiens* mosquitoes (*Cx. quinquifasciatus* Say, *Cx. pallens* Coquillet, *Cx. australicus* Dobrotworsky and Drummond, *Cx. pipiens* Linneaus, *Cx. globocoxitus* Dobrotworsky and *Cx. molestus* Forskll) (Farajollahi *et al*., 2011) *Cx. quinquefasciatus* are predominant in the urban environment notably in Africa including Nigeria (Mohammed *et al.,* 2021), as a result of the different suitable environmental conditions suitatable as a breeding site which were created by rapid unplanned urbanization (Nchoutpouen *et al*., 2019). Such high prevalence of culex mosquitoes were obsereved from different studies conducted across the country, in a study conducted in Niger state *Cx. quinquefasciatus* of all the mosquito species observed acount for 52% (Odoh *et al*., 2025), a prevalence of 98.5% was observed in Jos (Njunwa, K. 2015), 94% was observed in Jigawa state (Hamza *et al.*2024).

Globally different techniques have been researched and are in use for the control of *culex* mosquitoes and other insect vectors among which includes the use of different biological strategies as reported by AL-Tememay *et al*., 2025., use of chemical methods (Shroff *et al*., 2020). Despite the different strategies employed in vector control, mosquito resistance to the different control measures have become a major global health challenge (Onen *et al*., 2023), which is driven by their biochemical, physiological, and behavioral adaptations. The resistance of culex mosquitoes is notably due to their adaptable and ubiquitous nature, which enables them to thrive in a wide spectrum of environments, from rural area, to agricultural landscapes to densely populated urban centers (Timileyin *et al*., 2025). Their larval habitats are equally diverse, encompassing natural water collections like streams and seasonal rain pools, as well as human-made sites such as rice paddies, clogged drainage gutters, and various artificial containers (Okiwelu, & Noutcha, 2012). Hence the global focus on new innovative ways such as the use of plant based bioinsecticides to control vectors (Şengül Demirak *et al.,* 2022).

The use of traditional repellents to repel or kill mosquitoes and other blood-sucking insects is common in rural communities in Africa (Shibeshi *et al.,* 2024). In Nigeria reports from different studies the likes of Gidado & Samaila, 2024; Okoh *et al.*2022 indicates rich local knowledge on the use of plant-based insecticide and repellent to manage disease transmitting and nuisance insects as well as crop pest. (Giday *et al., 20*18)

Hyptis suaveolens (bush mint) has long been known for its powerful repellent action on insects, and mosquitoes in particular. In developing countries, Lamiaceae have traditionally been used for their insecticidal and repellent properties against several insects' species (Conti *et al., 20*12). Most of them belong to the Hyptis genus that includes more than 400 species that grow in the tropical regions of the world, mainly in Africa and America and are highly aromatic plants (Benelli *et al., 20*12).

MATERIALS AND METHODS

Description of the study area

This study was conducted with in Katsina local government of Katsina state, where the mosquito larvae were collected at Dogon Rafi along Kofar Marusa area. Katsina State is located in the North-Western part of Nigeria, bordering Niger Republic to the North, Zamfara state to the South, Kano state to the East and Jigawa state to the West (El-Ladan *et al.,* 2014). The state covers a total area of 24, 192 km^2 spanning between longitude 10°33'59'' to 13°18'30''N and latitude 6°59'32'' to 9°00'0.1''E with an estimated population projection of 7.8 million people (NBS, 2017; Ladan, 2014).

Collection & preparation of plant sample.

Leaves of *Hyptis suaveolens* were obtained from Katsina-Mani Road, Katsina State. The plant was identified at the herbarium of the Biology Department, Umaru Musa Yar'Adua University Katsina with voucher number UMYUH2395.

Plant Sample Preparation and Extraction of plant extract for phytochemical screening.

The leaves of the test plant were rinsed with water to remove dirt and were spread out on a clean surface and allowed enough time to air-dry under shade at room temperature, the leaves were grinded into powder using mortar and pestle. The extraction of the plant extract was carried out using the quick Maceration method was carried according to the methods described by Fotsing *et al., 20*22.

Preparation of Pellets

Insecticidal pellets were prepared from the test leaf powder blended with pulverized wood shavings using Renew® a common commercial laundry starch following the method of Tawatsin *et al.* (2002) and Ajelara *et al.* (*20*18).

Qualitative Phytochemical Screening

Phytochemical screening of the plant extracts was carried out employing standard procedures and tests described by (Vishnoi *et al.,* 1979, Sofowora *et al.,* 1993, Trease *et al.* 1989; Aliyu & Aliyu, 2022), where the presence of alkaloids, flavonoids, glycosides, reducing sugars, saponins, steroids and tanins were measured.

Mosquito larvae collection & Breeding.

Culex mosquito larvae were identified and gotten from the drainage of Dogon rafi along Kofar marusa, Katsina Nigeria. Some of the larvae were allowed to pupate and transform into the adult mosquito.

The adult mosquitoes were fed on glucose solution for continuous maintenance of mosquito colony.

Test for repellency of *Hyptis suaveolens* crude extract against adult culex Mosquitoes:

The repellency test was carried out using the cage test of World Health Organization (WHO 2009). The cage test is the most common way of testing the effectiveness of mosquito repellency. Six cages (size: 35×35×35 cm per side) with a slot for inserting an arm were used as described by (WHO 2009).

Test for toxicity of *Hyptis suaveolens* pellet smoke against adult culex Mosquitoes

Twenty (two days old) adult culex were introduced into laboratory mosquito cages (35×35×35 cm) square cages. A 20% concentration pellet was ignited with lighter and mounted on a hanger inside the cage which was then covered to avoid the smoke escaping. The number of mosquitoes knocked down or dead was recorded at 30, 60, 90 and 120min of exposure. Mosquitoes found on the cage floor unable to fly but moving their legs, antennae or any body part were counted as knocked down. Similarly, mosquitoes that were knocked down and unable to move any body part upon gentle prodding of the abdomen with a fine brush brittle were counted as dead. The same procedure was used to set up similar experiments using 40, 60, 80 or 100% pellets, respectively. In each case control was set up using pellets without *H. suaveolens* leaf powder (Ajelara *et al., 20*18).

RESULTS

Phytochemical analyses of *Hyptis suaveolens*

The result of phytochemical screening of crude extract of dried leaves of *Hyptis suaveolens* is presented in Table 1. The analysis revealed the presence of alkaloids, flavonoids, glycosides, reducing sugar, steroids, saponins, and tannins.

Repellency effects of Hyptis suaveolens against culex mosquitoes.

The mosquito repellency test assessed how well *Hyptis suaveolens* prevented mosquito landings at different treatment concentrations as shown in Table 2. The total number of mosquitoes that landed varied significantly. On average, only 12% of mosquitoes landed across all treatments. The control group (0% concentration) had the highest mosquito landing rate at 60%, while no mosquito landed at concentrations of 40% and above.

Regression Analysis of mosquito repellency activity.

The results on Figure 2 showed a negative relationship between concentration and mosquito landings, meaning that as concentration increases, mosquito landing decreases. The regression model explained 56% of the variation in mosquito landings ($R^2 = 0.560$), which indicates a moderate effect.

The regression coefficient for concentration was -0.48, meaning that for every 10% increase in extract concentration, mosquito landings decreased by 4.8%. However, the p-value was 0.087, which is not statistically significant at the 5% level ($p < 0.05$).

Toxicity analysis of *Hyptis suaveolens against culex mosquitoes*

After 120 minutes of exposure 48% of adult culex mosquito can fly, 32% can move from one place to another and 20% can move some parts of their body while 0% mortality was recorded in control cage. While in 20% concentration of dried leaves of *Hyptis suaveolens* in 1gram of pulverized wood powder 12% of mortality rate was recorded, 52% mortality rate was recorded at 40% concentration, 88% mortality rate at 60% concentration and 100% mortality rate at 80% and 100% concentration of dried leaves of *Hyptis suaveolens* dried leaves respectively (Table 3).

Regression analysis of mosquito toxicity activity.

The results on Figure 3 showed a strong positive relationship between concentration and mosquito death rates. The model explained 92% of the variation in mosquito mortality ($R^2 = 0.920$), meaning that treatment concentration is a very strong predictor of mosquito death.

The regression coefficient for concentration was 1.14, indicating that for every 10% increase in concentration, mosquito mortality increased by 11.4%. The p-value was 0.002, which is highly significant ($p < 0.05$). This confirms that increasing the concentration of *Hyptis suaveolens* extract has a statistically significant effect on mosquito mortality.

Table 1: Phytochemical compound of the leaves of *Hyptis suaveolens*

S/N	Phytochemical compound	Status
1	Alkaloids	Present
2	Flavonoids	Present
3	Glycosides	Present
4	Steroids	Present
5	Reducing sugar	Present
6	Saponins	Present
7	Tannins	Present

Table 2: mosquito repellency of *Hyptis suaveolens*

Treatment	No. of culex spp. Landed (%)	Time interval (minutes)
Control arm	15 (60%)	30
20% concentration	3 (12%)	30
40% concentration	0 (0%)	30
60% concentration	0 (0%)	30
80% concentration	0 (0%)	30
100% concentration	0 (0%)	30

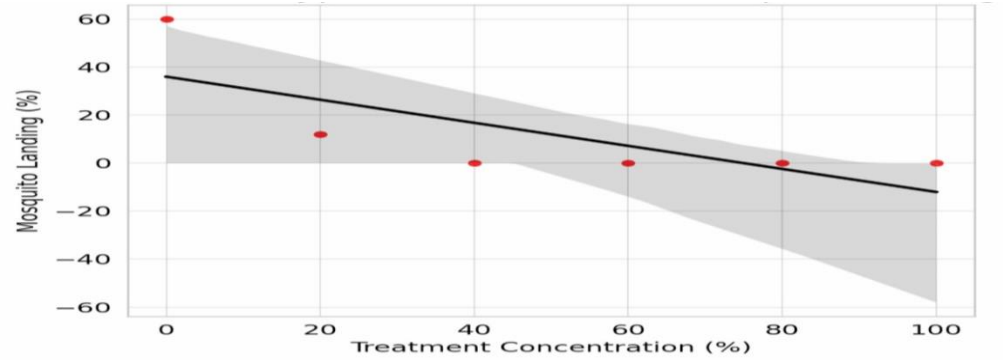

Fig. 2. Effect of *Hyptis suaveolens* on Mosquito Repellency showing that as concentration increases, mosquito landings decrease

Table 3: Toxicity of *Hyptis suaveolens* leaves smoke against culex mosquito

Treatment	No. of *Culex* spp. Exposed	Mortality Rate	Time Interval (Minutes)
Control	15	0%	120
20% concentration	25	12%	120
40% concentration	25	51%	120
60% concentration	25	88%	120
80% concentration	25	100%	120
100% concentration	25	100%	120

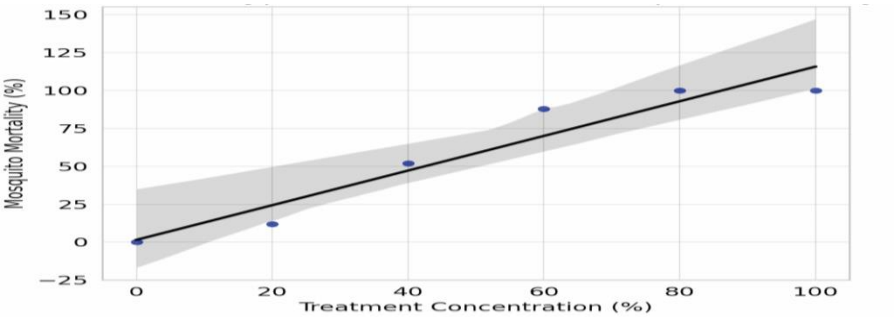

Fig 3. Effect of *Hyptis suaveolens* on Mosquito Mortality (Toxicity Test)

$y = 1.1429x + 1.5238$
$R^2 = 0.9197$

DISCUSSION

Different studies the likes of (Sánchez *et al., 20*20, Panigrahi *et al., 20*22) reported that *Hyptis suaveolens* belonging to family Lamiaceae serves as a rich source of different medicinally important phytochemicals such as tannins, saponins, phenols, flavonoids, terpenoids, alkaloids, and sterols as was observed from the present study. Over the years different researches from around the world shows how many of these phytochemical compounds have antioxidative, anti-inflammatory, antispasmodic, anti-septic, anti-cancer, anti-ulcer, antimicrobial, antibacterial, antiviral, antifungal activity (Mishra *et al., 20*21., Gayatri *et al., 20*21., Gaikwaid *et al., 20*23). The toxicity analysis from the present study shows that the mortality rate of adult culex mosquito is directly proportional to the percentage of *Hyptis suaveolens* dried leaves in the pulverized wood powder. The present study agrees with findings of Ayange *et al., 20*15. Who worked on the Effect of dried leaves extract of *Hyptis suaveolens* on various stages of mosquito development in Benue State, Nigeria. The lower concentration (50 mg/ml), of the extract that shows much lower percentage of mortality rate in the present study is similar as what was reported by Ajelara *et al., 20*18. who worked on the Toxicity of Smoke from Pelletized Leaf Powder and Essential Oil of *Hyptis suaveolens* Against Anopheles gambiae.

At the beginning of the repellency test, mosquitos were observed to perch on the volunteer hand within three minutes, which shows that the volunteer does not naturally repel mosquito as reported by WHO 2009. Equally effect of pellets and plant-based coils have shown to be similar in their mode of action; this have been demonstrated by Tawatsin *et al., 20*02; Ajelara *et al.*2018.

The toxicity analysis from the present study shows that the mortality rate of adult culex mosquito is directly proportional to the percentage of *Hyptis suaveolens* dried leaves in the pulverized wood powder. The present study agrees with findings of Ayange *et al., 20*15. Who worked on the Effect of dried leaves extract of *Hyptis suaveolens* on various stages of mosquito development in Benue State, Nigeria. The lower concentration (50 mg/ml), of the extract that shows much lower percentage of mortality rate in the present study is similar as what was reported by Ajelara *et al., 20*18. who worked on the Toxicity of Smoke from Pelletized Leaf Powder and Essential Oil of *Hyptis suaveolens* Against Anopheles gambiae.

CONCLUSION

The presence of the different phytochemicals obtained from this study such as alkaloids, flavonoids, saponins, reducing sugar, glycosides, steroids and tannins shows that *Hyptis suaveolens* besides having antibacterial, antimicrobial, anti-inflammatory effects as reported by other researchers, it also shows insects repellency effects.

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