

Repellent activities of ethanolic extract of *Cymbopogon citratus* (Poaceae) and *Ocimum basilicum* L. (Lamiaceae) leaves against *Culex quinquefasciatus* mosquitoes (Diptera: Culicidae) in Dogbo district in south-western Benin, West Africa

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Abstract

The alteration of mosquito life cycle using larvicides is key in the control of diseases transmitted by mosquitoes. The residual and environmental effect of some of these synthetic larvicides has given an opportunity to search for a potent larvicide as an alternate control measure against mosquitoes. The current study was aimed to evaluate the repellent activities of ethanolic extract of *Cymbopogon citratus* (Poaceae) and *Ocimum basilicum* L. (Lamiaceae) leaves against *Culex quinquefasciatus* adult mosquitoes in lymphatic filariasis vector control in Dogbo district in south-western Republic of Benin. Larvae of *Culex quinquefasciatus* mosquitoes were collected from breeding sites using the dipping method from September to November 2023 during the small rainy season in Dogbo district of Couffo department. Then, these larvae were reared in insectary until became adult mosquitoes. Repellent activities of ethanolic extract of both plant leaves were measured on adult mosquitoes with different concentrations of 1mg/cm², 2mg/cm², 3mg/cm², 4mg/cm² and 5mg/cm². The results showed that the leaves of both plant had repellent effects against *Culex quinquefasciatus* adult mosquitoes. However, the repellent effect of *Cymbopogon citratus* leaves was higher than that of *Ocimum basilicum* L. leaves. Both ethanolic extracts of *Cymbopogon citratus* and *Ocimum basilicum* L leaves are found to be effective in the repellency against the bites of *Culex quinquefasciatus* adult mosquitoes.

Keywords: *Cymbopogon citratus*; *Ocimum basilicum* L; *Culex quinquefasciatus*; Lymphatic filariasis; Benin

1. Introduction

Mosquitoes are one of the most important insect pests that affect the health and well-being of humans and domestic animals worldwide. They can cause a variety of health problems due to their ability to transmit viruses and other disease-causing pathogens even in the arid regions. Female mosquitoes require a blood meal for egg production, and they can produce a painful bite as they feed. While feeding, they can transmit to humans and other animals, viruses that are responsible causes of brain inflammation (encephalitis) [1], dengue fever [2], yellow fever [3], and protozoan parasites such as malaria and filariasis [4]. The most susceptible to the effects of these mosquito-borne pathogens are children and the elderly. However, in some instances life-threatening illness and/or permanent debilitation can occur in infected human hosts of any age [5].

Prevention of mosquito bites is one of the main strategies to control or minimize incidence of many vector-borne diseases, such as filariasis, transmitted to human through mosquito bites. The use of insect repellants can provide

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practical and economical means of preventing mosquito-borne diseases. It is not important only for local people in disease risk areas especially in tropical countries, but also for travelers who are vulnerable to diseases spread by mosquito vectors when they visit and seek leisure away from their home countries.

The adverse environmental effects from chemical insecticides, the spread of resistance to insecticide in the main vector species of human diseases could jeopardize these control tools. These vector species are becoming more and more resistant to synthetic insecticides [6-7].

Due to the detrimental effects of synthetic products used for decades to reduce the transmission of mosquito-borne diseases, coupled with current resistance problems that have made it difficult to eliminate the vector, there is a growing interest in exploring plant-derived products to control mosquito populations and prevent their bite. Therefore, it is crucial to develop control strategies that are safer, more effective, and more sustainable compared to traditional chemical interventions.

The potential of plants as insecticide, repellent or fumigant sources against mosquitoes and other pest and vectors insects is well known [8-10]. Several ethnobotanical studies in East and West Africa have shown that human populations use local plants, mostly aromatic plants, to protect themselves against mosquito bites [11-12].

Very few researches were published on the use of essential oils in *Culex quinquefasciatus* adult repellency in Benin. Therefore, there is a need to carry out new researches for this purpose.

The goal of this study was to evaluate the repellent activities of ethanolic extracts of *Cymbopogon citratus* and *Ocimum basilicum* L leaves against *Culex quinquefasciatus* adult mosquitoes in lymphatic filariasis vector control in Dogbo district in south-western Republic of Benin.

2. Material and methods

2.1. Study area



Figure 1 Map of Republic of Benin showing Dogbo District surveyed

The study area is located in Republic of Benin (West Africa) and includes the department of Couffo. Couffo department is located in the south-western Benin and the study was carried out more precisely in Dogbo district (Figure 1). The southern borders of this district are Lokossa and Bopa districts. The northern border is Djakotomey district. The eastern border is Lalo district and the western border of Dogbo district is Togo republic. Dogbo district covered 475 km² and belongs to geographic region of ADJA. The choice of the study site took into account the economic activities of populations, their usual protection practices against mosquito bites and peasant practices to control farming pests. We took these factors into account to evaluate the repellent activities of ethanolic extract of *Cymbopogon citratus* and *Ocimum basilicum* L leaves against *Culex quinquefasciatus* adult mosquitoes in lymphatic filariasis vector control in Dogbo district in south-western Republic of Benin. Couffo has a climate with four seasons, two rainy seasons (March to July and August to November) and two dry seasons (November to March and July to August). The temperature ranges from 25 to 30°C with the annual mean rainfall between 900 and 1100 mm.

2.2. Mosquito sampling

Culex quinquefasciatus mosquitoes were collected from September to November 2023 during the small rainy season in Dogbo district of Couffo department. Larvae were collected from breeding sites using the dipping method and kept in labeled bottles (Figure 2). The samples were then carried out to the insectary of Laboratory of Pluridisciplinary Researches of Technical Teaching (LaRPET) in Department of Sciences and Agricultural Techniques of Normal High School of Technical Teaching (ENSET) located in Dogbo district.



Figure 2 A breeding site of *Culex quinquefasciatus* larvae surveyed in Dogbo district

2.3. Collection of the plant leaves

The leaves of *Cymbopogon citratus* and *Ocimum basilicum* L were collected in their predilection areas in Dogbo district.



Figure 3 Plant of *Ocimum basilicum* L.



Figure 4 Plant of *Cymbopogon citratus*

2.4. Plant leaves extraction

To extract *Cymbopogon citratus* and *Ocimum basilicum* L leaves, we collected fresh green leaves of both plants and we washed them with tap water. The leaves were dried outside of the laboratory at ambient temperature in a class room for a period of 3 days. Then, the dried leaves were crushed or grounded into powder with an electronic mix and a weight of 100grammes of the leaf powder of each plant were extracted with 250 milliliters of ethanol for a period of 48 hours at temperature of 25°C. Each extract was then filtered with the aid of Whatman No. 1 filter paper. The mixture were dried and stored in some labeled bottles before being used in the preparing of concentrations of 1mg/cm², 2mg/cm², 3mg/cm², 4mg/cm² and 5 mg/cm². These concentrations were then used in the performing of repellent activities test of ethanolic extract of *Cymbopogon citratus* and *Ocimum basilicum* L leaves separately.

2.5. Performing of repellent activities test of *Cymbopogon citratus* and *Ocimum basilicum* Leaves

A batch of twenty five (25) adult mosquito *Culex quinquefasciatus* aged 3 days old was put in a mosquito cage. These mosquitoes were obtained after collection from the breeding sites and rearing of larvae and pupae from Dogbo district in the insectary of the Department of Sciences and Agricultural Techniques of Normal High School of Technical Teaching (ENSET). A volunteer whom before-arm is without trace of pomade, perfumed soap and so on, the day of the test was used. The left before-arm of this volunteer (handful closed) was coated with ethanol and served as control whereas his right before-arm was coated with 1mg/cm², 2mg/cm², 3mg/cm², 4mg/cm² and 5 mg/cm² of ethanolic extract of *Cymbopogon citratus* and *Ocimum basilicum* L leaves separately. The test was performed at night in laboratory (LaRPET) between 8 and 10 pm with each of prepared concentrations. The before-arm control and test were introduced simultaneously in the mosquito cage. The test was repeated during five (05) consecutive night in the same conditions of temperature (25+/-2) °C and relative humidity (80+/-2) % in laboratory (LaRPET). Both before-arm control and test were exposed to mosquitoes during one minute and retired after one minute to be introduced after four minutes of rest time. That was repeated during two hours which correspond to this test duration. The number of mosquitoes which went towards the before-arm control and test were registered just before the volunteer moving his before-arms in order to avoid that these mosquitoes took their meals on his body.

2.6. Statistical analysis

Correlation between different concentrations of ethanolic extract of *Cymbopogon citratus* and *Ocimum basilicum* L leaves and duration of protection against *Culex quinquefasciatus* mosquitoes was established. The significance of the relationship at 5% level was determined using student t-test statistical application.

3. Results

3.1. Evaluation of repellent activity of *Cymbopogon citratus* leaves

The analysis of figure 5 showed that *Culex quinquefasciatus* female adult mosquitoes preferred go towards the hand coated with ethanol (control hand) than the hand coated with ethanolic extract of *Cymbopogon citratus* leaves (test hand) with all tested concentrations. The repellent activities were detected after forty five (45) minutes with all the concentrations tested of 1mg/cm², 2mg/cm², 3mg/cm², 4mg/cm² and 5mg/cm².

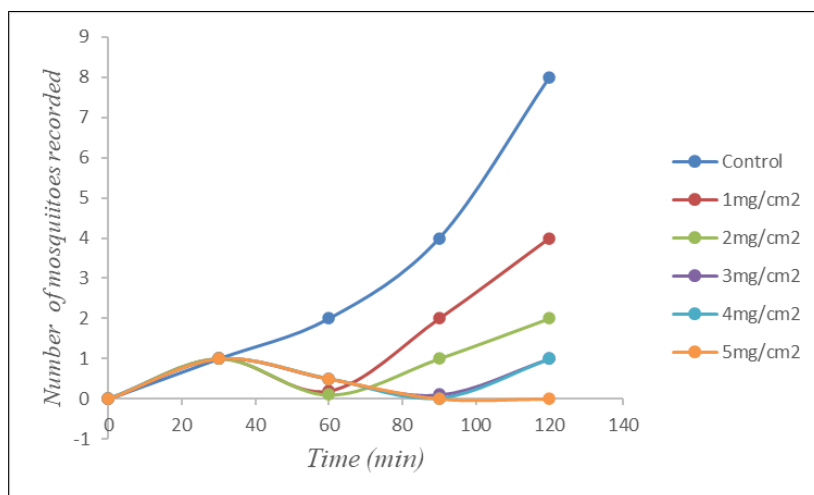


Figure 5 Repellent activity of *Cymbopogon citratus* leaves on *Culex quinquefasciatus* female adult mosquitoes.

3.2. Evaluation of repellent activity of *Ocimum basilicum* L. leaves

The analysis of figure 6 showed that *Culex quinquefasciatus* female adult mosquitoes also preferred go towards the hand coated with ethanol (control hand) than the hand coated with ethanolic extract of *Ocimum basilicum* L. leaves (test hand) with all tested concentrations. The repellent activities were detected after one hour with all the concentrations tested of 1mg/cm², 2mg/cm², 3mg/cm², 4mg/cm² and 5mg/cm².

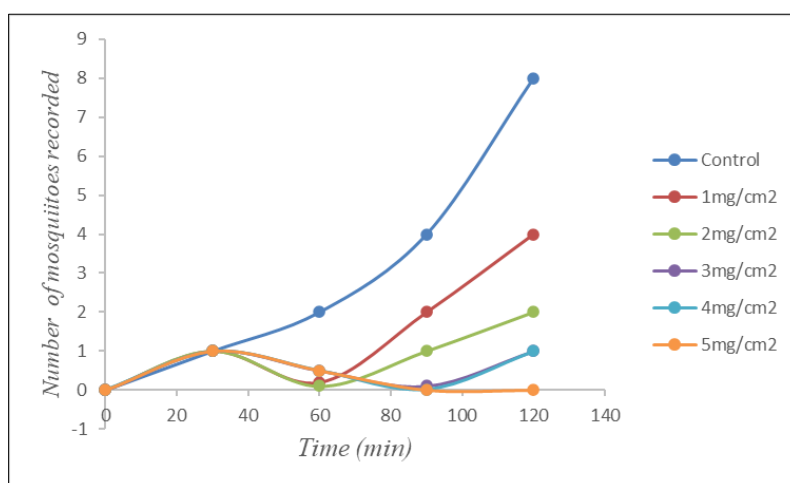


Figure 6 Repellent activity of *Ocimum basilicum* L. leaves on *Culex quinquefasciatus* female adult mosquitoes

The table 1 showed the advantages and disadvantages of the use of ethanolic extract of *Cymbopogon citratus* and *Ocimum basilicum* L. leaves.

Table 1 Advantages and disadvantages of the use of ethanolic extract of plant leaves

Advantages	Disadvantages
Plants as <i>Cymbopogon citratus</i> and <i>Ocimum basilicum</i> L. are cultivated in many regions in Benin country Ethanolic extract of <i>Cymbopogon citratus</i> and <i>Ocimum basilicum</i> L. leaves is miscible with water after mixture Ethanolic extract of <i>Cymbopogon citratus</i> and <i>Ocimum basilicum</i> L. leaves is a cheap and easy method for larval control in their breeding sites Mosquitoes may not develop resistance to ethanolic extract of <i>Cymbopogon citratus</i> and <i>Ocimum basilicum</i> L. leaves Ethanolic extract of <i>Cymbopogon citratus</i> and <i>Ocimum basilicum</i> L. Leaves is not toxic to most non-target organisms including mammals and fish.	Limited effectiveness of ethanolic extract of <i>Cymbopogon citratus</i> and <i>Ocimum basilicum</i> L. leaves in the presence of vegetation and floating debris (is the main disadvantage)

4. Discussion

Although the most common mosquito repellent currently available on the market, DEET (N,N-diethyl-3-methylbenzamide) has shown excellent protection from mosquito bites [13-15], there were reports of toxicity problems after application of DEET, range from mild effects such as contact urticaria [16] and skin eruption [17], to severe reactions, such as toxic encephalopathy [18-20]. To overcome these adverse effects, attempts to find and develop repellents derived from plant extracts is necessary. The development and use of locally available plants showing repellent activity avails an alternative strategy for the control or minimization of mosquito-borne diseases, especially in developing countries.

In the current study, *Culex quinquefasciatus* female adult mosquitoes preferred go towards the hand coated with ethanol (control hand) than the hand coated with ethanolic extract of *Cymbopogon citratus* leaves (test hand) with all tested concentrations. The repellent activities were detected after forty five (45) minutes with all the concentrations tested of 1mg/cm², 2mg/cm², 3mg/cm², 4mg/cm² and 5mg/cm². These results showed that the ethanolic extract of *Cymbopogon citratus* leaves had repellent activities against *Culex quinquefasciatus* female adult mosquitoes. The results obtained in the current study corroborated with those by Pushpanathan *et al.* [21] who had studied the larvicidal, ovicidal and repellent activities of *Cymbopogon citratus* Stapf (Graminae) essential oil against the filarial mosquito *Culex quinquefasciatus* (Say) (Diptera: Culicidae) with good results. In fact, their results showed that skin repellent test at 1, 2.5 and 5 mg/cm² concentration of *Cymbopogon citratus* gave 100% protection up to 3, 4 and 5 hours respectively. Also, Singh *et al.* [22] have recently carried out a study in Nigeria to determine insecticidal activities of essential oil extracts from lemon grass (*Cymbopogon citratus*) against adult stage of the mosquito *Anopheles* and *Culex* spp with good results. In fact, the greatest insecticidal activity 60% was exhibited at 4.5 % concentration with a 24hours exposure by *Cymbopogon citratus*.

In the current study, *Culex quinquefasciatus* female adult mosquitoes also preferred go towards the hand coated with ethanol (control hand) than the hand coated with ethanolic extract of *Ocimum basilicum* L. leaves (test hand) with all tested concentrations. The repellent activities were detected after one hour with all the concentrations tested of 1mg/cm², 2mg/cm², 3mg/cm², 4mg/cm² and 5mg/cm². These results showed that the ethanolic extract of *Ocimum basilicum* L. leaves had also repellent activities against *Culex quinquefasciatus* female adult mosquitoes. The results obtained in the current study corroborated with those by Ramayanti *et al.* [23] who had determined the repellent potency of *Ocimum basilicum* against *Culex quinquefasciatus*. These authors showed that the extract of this plant for 6 hours at all concentrations were able to repel mosquitoes up to 92.51%. So, the extract of *Ocimum basilicum* was effective as a repellent against the *Culex quinquefasciatus*. The *Culex quinquefasciatus* mosquito is the main vector of filariasis and the various ways to control vector breeding is the use of insecticide from natural ingredients, one of which is basil containing essential oils and proven is effective as a repellent against the mosquito. Also, Baba *et al.* [24] had

shown that the essential oils of *Ocimum basilicum* leaf extracts have been confirmed to have potentials as mosquito repellent agents against *Culex quinquefasciatus*.

The use of ethanolic extract of both *Cymbopogon citratus* and *Ocimum basilicum* L. leaves presented many advantages and very few disadvantages. In fact, *Cymbopogon citratus* and *Ocimum basilicum* L. are cultivated in many regions in Benin country. Ethanolic extract of *Cymbopogon citratus* and *Ocimum basilicum* L. leaves is miscible with water after mixture. Ethanolic extract of *Cymbopogon citratus* and *Ocimum basilicum* L. leaves is a cheap and easy method for larval control in their breeding sites. Mosquitoes may not develop resistance to ethanolic extract of *Cymbopogon citratus* and *Ocimum basilicum* L. leaves. Ethanolic extract of *Cymbopogon citratus* and *Ocimum basilicum* L. leaves is not toxic to most non-target organisms including mammals and fish. However, limited effectiveness of ethanolic extract of *Cymbopogon citratus* and *Ocimum basilicum* L. leaves in the presence of vegetation and floating debris is the main disadvantage in its use.

5. Conclusion

Both ethanolic extracts of *Cymbopogon citratus* and *Ocimum basilicum* L. leaves have repellent activities against lymphatic filariasis vectors. They are found to be effective against the bites of *Culex quinquefasciatus* adult in the current study. However, *Cymbopogon citratus* extract has the highest repellent activities. More effort must be done in order to explore the potentiality of these plant parts available for botanical repellent preparing.

Compliance with ethical standards

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Disclosure of conflict of interest

There is no conflict of interest regarding the publication of this paper.

Statement of ethical approval

The study follows proper ethical procedures.

Statement of informed consent

Informed consent was obtained from all individual participants included in the study.

References

- [1] Meisch MV. The Dark Ricefield Mosquito, *Psorophora columbiae*. *Wing Beats*. 1994; 5(1):8.
- [2] Bennett KE, Olson KE, Munoz ML, Fernandez-Salas L, Farfan-Ale JA and Higgs S. Variation in Vector Competence for Dengue Virus among Twenty Four Collections of *Aedes aegypti* From Mexico and the United States. *Am J Trop Med Hyg*. 2002; 67(1):85-92.
- [3] Reiter P, Amador MA, Anderson RA and Clark GG. Short Report: Dispersal of *Aedes aegypti* in a Urban Area After Blood Feeding as Demonstrated by Rubidium-Marked Eggs. *Am J Trop Med Hyg*. 1995; 52:177-179.
- [4] Schoepke A, Steffen F and Norman G. Effectiveness of Personal Protection Measures Against Mosquito Bites for Malaria Prophylaxis in Travelers. *J Travel Med*. 1998; 5:188-92.
- [5] Service MW. Mosquito (Culicidae). In: Medical insect and arachnids, Lane, R.P. and R.W. Crosskey (Eds.). *Chapman and Hall, London*. 1993 ; pp: 120-240.
- [6] Badolo A, Traore A, Jones CM, Sanou A, Flood L, Guelbeogo WM, Ranson H, Sagnon NF. Three years of insecticide resistance monitoring in *Anopheles gambiae* in Burkina Faso: resistance on the rise ?. *Mala J*. 2012; 11: 232.
- [7] Toé KH, Jones CM, Sagnon N'F, Ismail HM, Dabiré RK, Ranson H. Increased pyrethroid resistance in malaria vectors and decreased bed net effectiveness, Burkina Faso. *Emerg Infect Dis*. 2014; 20(10): 1691-1696.

- [8] Lucia A, Juan LW, Zerba EN, Harrand L, Marcó M, Masuh HM. Validation of models to estimate the fumigant and larvicidal activity of Eucalyptus essential oils against *Aedes aegypti* (Diptera:Culicidae). *Parasitol Res.* 2012; 110: 1675-1686.
- [9] Regnault-Roger C, Vincent C, Arnason JT. Essential oils in insect control: low-risk products in a high-stakes world. *Ann Rev Entomology.* 2012; 57: 405-424.
- [10] Dias CN, Moraes DFC. Essential oils and their compounds as *Aedes aegypti* L. (Diptera:Culicidae) larvicides: review. *Parasitol Res.* 2014; 113: 565-592.
- [11] Palsson K, Jaeson TGT. Plant products used as mosquito repellents in Guinea Bissau, West Africa. *Acta Trop.* 1999; 39-52.
- [12] Seyoum A, Kabiru EW, Lwande W, Killeen GF, Hassanali A, Knols BGJ. Repellent of live plants against *Anopheles gambiae* from human baits in semi-field experimental huts. *Am J Trop Med Hyg.* 2002; 67(2): 191-195.
- [13] Yap HH. Effectiveness of soap formulations containing deet and permethria as personal protection against outdoor mosquitoes in Malaysia. *J Am Mosquito Control Assoc.* 1986; 2: 63-67.
- [14] Walker TW, Robert LL, Copeland RA. Field evaluation of arthropod repellents, Deet and piperidine compound against *Anopheles arabiensis* in Western Kenya. *J Am Mosquito Control Assoc.* 1996; 12:172-176.
- [15] Thawara U, Tawatsin A, Chompoosri J. Laboratory and evaluation of the insect repellent 3535 (Ethyl butylacetylaminopropionate) and deet against the insect mosquito vectors in Thailand. *J Am Mosquito Control Assoc.* 2001; 17: 190 -195.
- [16] Malbach HI, Johnson HL. Contact urticaria syndrome to diethyl-toluamide (immediate-type hypersensitivity). *Arch Derma.* 1975; 111: 726-730.
- [17] Reuveni YH, Yagupsky P. Diethyl toluamide containing insect repellent: adverse effects in worldwide use. *Arch Derma.* 1982; 118: 582-583.
- [18] Zadikoff CM. Toxic encephalopathy associated with the use of insect repellent. *J Pediat.* 1979; 95: 140-142.
- [19] Roland EH, Jan JE, Rigg JM. Toxic encephalopathy in a child after a brief exposure to insect repellents. *Can Med Assoc J.* 1985; 132: 155-156.
- [20] Edwards DL, Johnson CE. Insect-repellent-induced toxic encephalopathy in a child. *Chin Pharm.* 1987; 6: 496-498.
- [21] Pushpanathan T, Jebanesan A and Govindarajan M. Larvicidal, ovicidal and repellent activities of *Cymbopogon citratus* Stapf (Graminae) essential oil against the filarial mosquito *Culex quinquefasciatus* (Say) (Diptera: Culicidae). *Trop. Biomed.* 2006; 23(2):208-212.
- [22] Singh K, Abdulhammed NA, Singh J, Norouzi R and Khoshkhoo A. Insecticidal efficacy of lemon grass (*Cymbopogon citratus*) and orange peel (*Citrus sinensis*) on mosquitoes. *J. Vet. Res. Adv.* 2024; 06(01): 36-40.
- [23] Ramayanti I, Hartanti MD & Rahman RA. Repellent Potency of N-Hexane Extract Leaf and Stem *Ocimum basilicum* against *Culex quinquefasciatus*. *Jurnal Lahan Suboptimal: JLSO.* 2022; 11(1), 42-50.
- [24] Baba G, Lawal A, Sharif H. Mosquito repellent activity and phytochemical characterization of essential oils from *Striga hermonthica*, *Hyptis spicigera* and *Ocimum basilicum* leaf extracts. *Br.J.Pharmacol.* 2012; 3(10):43-48.