World Journal of Advanced Research and Reviews
eISSN: 2581-9615 CODEN (USA): WJARAI
Cross Ref DOI: 10.30574/wjarr
Journal homepage: https://wjarr.com/

(RESEARCH ARTICLE)

Check for updates

Repellent activities of ethanolic extract of *Artemisia annua* (Asteraceae) and *Carica papaya* linn (Caricaceae) leaves against *Culex quinquefasciatus* mosquitoes in Lokossa district in south-western Benin, West Africa

Nazaire Aïzoun [1,*], Sylvestre Codjia [1], Eloi Honvoh [1] and Daniel Chougourou [2]

[1] *Department of Sciences and Agricultural Techniques, Normal High School of Technical Teaching (ENSET) of Lokossa, National University of Sciences, Technologies, Engineering and Mathematics (UNSTIM) of Abomey, P. O. Box 133 Lokossa Cotonou, Benin*
[2] *Department of Environment Genius, Polytechnic School of Abomey-Calavi (EPAC), University of Abomey-Calavi(UAC), Cotonou, Benin.*

World Journal of Advanced Research and Reviews, 2025, 26(01), 3127-3134

Publication history: Received on 05March 2025; revised on 14 April 2025; accepted on 16 April 2025

Article DOI: https://doi.org/10.30574/wjarr.2025.26.1.1187

Abstract

The use of chemical insecticides causes important damages to environment and human health and there is a need to search for alternative solutions. The current study was aimed to evaluate the repellent activities of ethanolic extract of *Artemisia annua* and *Carica papaya* linn leaves against *Culex quinquefasciatus* adult mosquitoes in lymphatic filariasis vector control in Lokossa district in south-western Republic of Benin. Larvae of *Culex quinquefasciatus* mosquitoes were collected from breeding sites using the dipping method from March to July 2024 during the great rainy season in Lokossa district of Mono 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/cm^2, 2mg/cm^2, 3mg/cm^2, 4mg/cm^2 and 5mg/cm^2. The results showed that the leaves of both trees had repellent effects against *Culex quinquefasciatus* adult mosquitoes. However, the repellent effect of *Carica papaya* linn leaves was higher than that of *Artemisia annua* leaves. Both ethanolic extracts of *Artemisia annua* and *Carica papaya* linn leaves are found to be effective in the repellency against the bites of *Culex quinquefasciatus* adult mosquitoes.

Keywords: *Artemisia annua; Carica papaya* linn; *Culex quinquefasciatus*; Lymphatic filariasis; Benin

1. Introduction

Mosquitoes have become a major public health concern due to the blood feeding females of different species transmitting malaria causing protozoa, elephantiasis causing filarial worms, and dengue fever, yellow fever, Rift valley fever, West Nile fever causing viruses [1].

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 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.

*Corresponding author: Nazaire Aïzoun

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 [2-3].

Contrastingly, conventional insecticides with single active ingredient and plant-based insecticides which comprise of plant materials act efficiently on both behavioural and physiological processes of insects which give little chance of pests developing resistance to such constituents [4]. The insecticidal properties of plants remain the subject of intense research with an explosion of research over the past decade.

The potential of plants as insecticide, repellent or fumigant sources against mosquitoes and other pest and vectors insects is well known [5-7]. 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 [8-9].So, the transmission of disease can be interrupted by controlling the vector using various methods including botanical phytochemicals with mosquitocidal potential and repellent activities [10]. Various products with plant based insecticidal and repellent properties are widely in use in control of mosquitoes and protection from mosquito bites [11].

Very few researches were published on the use of essential oils in the repellency of *Culex quinquefasciatus* adult 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 extract of *Artemisia annua* and *Carica papaya* linn leaves against *Culex quinquefasciatus* adult mosquitoes in lymphatic filariasis vector control in Lokossa district in south-western Republic of Benin.

2. Material and methods

2.1. Study area

Figure 1 Map of Republic of Benin showing Lokossa District surveyed

The study area is located in Republic of Benin (West Africa) and includes the department of Mono. Mono department is located in the south-western Benin and the study was carried out more precisely in Lokossa district (Figure 1). The southern borders of this district are Athiémé and Houéyogbé districts. The northern border is Dogbo district. The eastern border is Bopa district and the western border is Togo republic. Lokossa district covered 260km^2. 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 *Artemisia annua* and *Carica papaya* linn leaves against *Culex quinquefasciatus* adult mosquitoes in lymphatic filariasis vector control in Lokossa district in south-western Republic of Benin. Mono 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 March to July 2024 during the great rainy season in Lokossa district of Mono 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 Lokossa district

2.3. Collection of the plant leaves

The leaves of *Artemisia annua* and *Carica papaya* linn were collected in their predilection areas in Lokossa district.

Figure 3 Farm of *Artemisia annua*

Figure 4 Tree of *Carica papaya*

2.4. Plant leaves extraction

To extract *Artemisia annua* and *Carica papaya* linn 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 leave 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/cm^2, 2mg/cm^2, 3mg/cm^2, 4mg/cm^2 and 5 mg/cm^2. These concentrations were then used in the performing of repellent activities test of ethanolic extract of *Artemisia annua* and *Carica papaya* linn leaves separately.

2.5. Performing of repellent activities test of *Artemisia annua* and *Carica papaya* linn 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 Lokossa 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/cm^2, 2mg/cm^2, 3mg/cm^2, 4mg/cm^2 and 5 mg/cm^2 of ethanolic extract of *Artemisia annua* and *Carica papaya* linn 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 *Artemisia annua* and *Carica papaya* linn 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 *Artemisia annua* and *Carica papaya* linn leaves

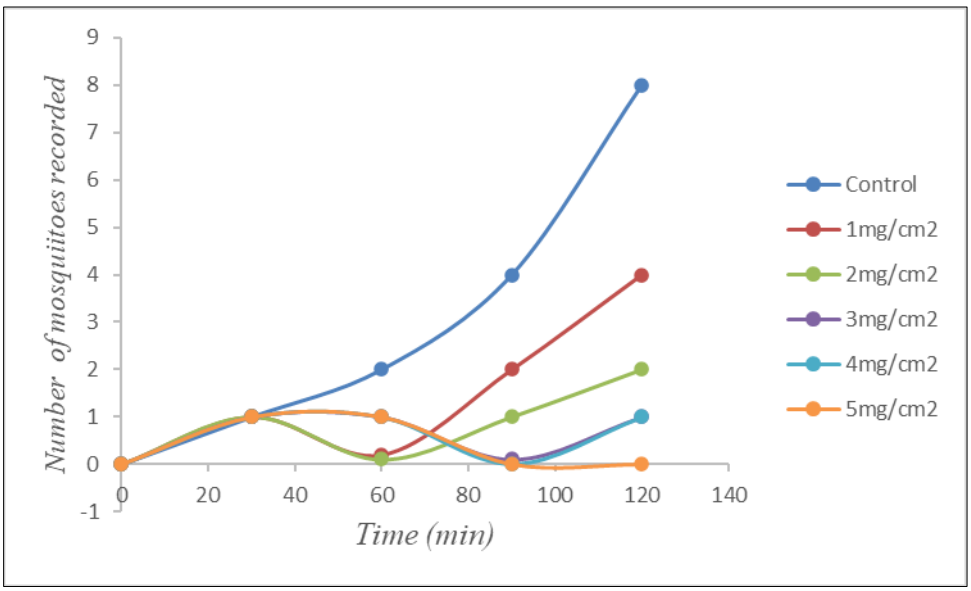

Figure 5 Repellent activity of *Artemisia annua* leaves on *Culex quinquefasciatus* female adult mosquitoes

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 *Artemisia annua* 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.

In similar way, 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 *Carica papaya* linn leaves (test hand) with all tested concentrations. The repellent activities were detected after fifty (50) minutes with all the concentrations tested of 1mg/cm², 2mg/cm², 3mg/cm², 4mg/cm² and 5mg/cm².

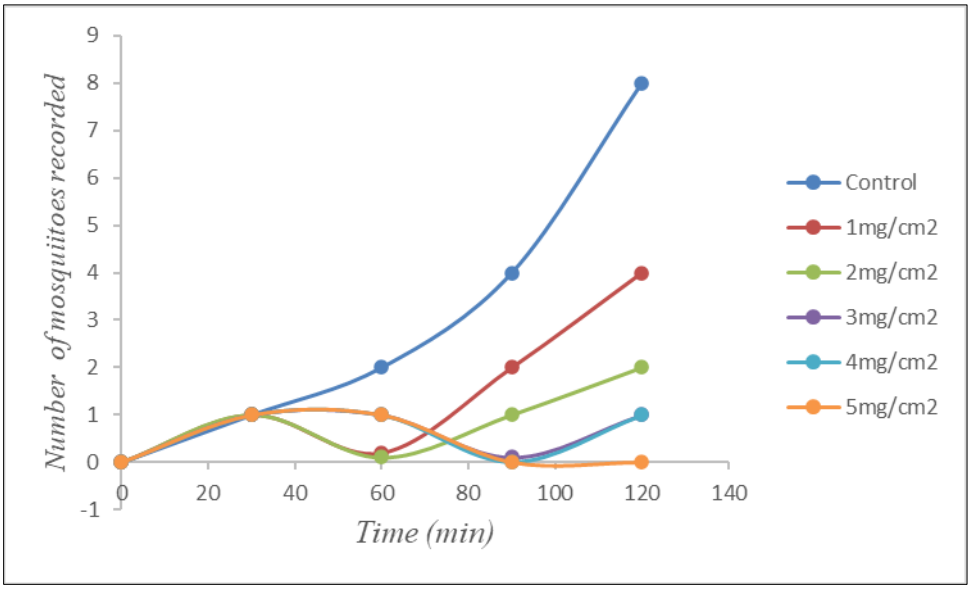

Figure 6 Repellent activity of *Carica papaya* linn leaves on *Culex quinquefasciatus* female adult mosquitoes

The table 1 showed the advantages and disadvantages of the use of ethanolic extract of *Artemisia annua* and *Carica papaya* L. leaves.

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

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

4. Discussion

Mosquitoes continue to transmit diseases despite the use of several methods of control such as treated bed nets, aerosols and repellent creams. Mosquito biting cause stress and medical problems to school going children. So, mosquitoes play a predominant role in the transmission of many diseases. Repeated use of synthetic insecticides for mosquito control has disrupted natural biological control systems. One such possibility is the use of botanicals which are readily biodegradable, nontoxic and show broad spectrum target specific activity.

Although the most common mosquito repellent currently available on the market, DEET (N,N-diethyl-3-methylbenzamide) has shown excellent protection from mosquito bites [12-14], there were reports of toxicity problems after application of DEET, range from mild effects such as contact urticaria [15] and skin eruption [16], to severe reactions, such as toxic encephalopathy [17-19]. 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 *Artemisia annua* leaves (test hand) with all tested concentrations. The repellent activities were detected after one hour with all the concentrations tested of 1mg/cm^2, 2mg/cm^2, 3mg/cm^2, 4mg/cm^2 and 5mg/cm^2. These results showed that the ethanolic extract of *Artemisia annua* leaves had repellent activities against *Culex quinquefasciatus* female adult mosquitoes. Our results corroborated with those obtained by Mutua *et al* [20] who had carried out an experimental ecological study to evaluate the effectiveness of two months old potted *Artemisia annua* plants for mosquito repellence in boarding schools' dormitories of Mwea Rice Irrigation Scheme, Kenya. For that, two months old potted live *Artemisia annua* plants were hung next to doors and windows of dormitories and mosquito including *Culex quinquefasciatus* Say collection for four months as per WHO protocol was done. Their study concluded that live *Artemisia annua* reduced mosquito density in dormitories of boarding schools. Further there was reduction of mosquito density in neighbouring dormitories, as far as 50 metres where *Artemisia annua* was not introduced. Their study recommends that *Artemisia annua* would be very important for implementing vector control strategies to prevent man-vector contact for the prevention of vector borne diseases. Another study was carried out by Mohan [21] to study the growth inhibitory nature of *Artemisia annua* extract against *Culex quinquefasciatus* (Say) with good results. In their study, Petroleum ether (Pee), carbon tetrachloride (Cte) and methanol extract (Mee) of *Artemisia annua* were screened for their efficacy against *Culex quinquefasciatus* larvae. Pee of *A. annua,* Mee of *A. annua* and Cte of *A. Annua* were found effective in descending order after 24 and 48 hours of treatment. *Pee of A. annua*, the most potent extract with LC of 78.2 ppm was selected to study its influence on the development and metamorphosis of the culicine mosquito. The extract significantly affected the hatching, larval development, pupal transformation and also lengthened the larval and pupal periods. Growth index was remarkably reduced. Treated culicine eggs, larvae and pupae showed deformities including disruption of the body wall, distorted alimentary canal, damaged tracheal network and arrested histogenesis. The extract has remarkable effect on the metamorphosis and high larvicidal potential, hence, can be used as an effective alternative to the existing synthetic pesticides for the control of *Cx. quinquefasciatus*.

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 *Carica papaya* linn leaves (test hand) with all tested concentrations. The repellent activities were detected after fifty (50) minutes with all the concentrations tested of 1mg/cm^2, 2mg/cm^2, 3mg/cm^2, 4mg/cm^2 and 5mg/cm^2. These results showed that the ethanolic extract of *Carica papaya* linn leaves had repellent activities against *Culex quinquefasciatus* female adult mosquitoes. Our results corroborated with those obtained by Rawani *et al* [22] who had studied the aliphatic amide from seeds of *Carica papaya* as mosquito larvicide, pupicide, adulticide, repellent and smoke toxicant with good results. In their study, the crude and solvent extracts of seed extract *of Carica papaya* was investigated for anti-mosquito potential, including larvicidal, pupicidal, adulticidal, smoke toxicity and repellent activities against *Culex quinquefasciatus*, the vector of filaria. The mortality rate of 3rd larval instars of *Cx. quinquefasciatus* at 0.5% concentration was significantly higher ($p<0.05$) than the mortality rates at 0.1%, 0.2%, 0.3% and 0.4% concentrations of crude extract. Among the solvent extracts, the petroleum ether extract showed the highest mortality at 100 ppm with LC 50 and LC 90 values of 31.16 ppm and 341.86 ppm against *Cx. quinquefasciatus*. In testing for pupicidal activity, this plant extract exhibited a slightly pupicidal potency with LC 50 values of 86.53 ppm against *Cx. quinquefasciatus*. It showed repellency against the adult females *Cx. quinquefasciatus* with 78 % protection. It also provided biting protection time of 4 hours against *Cx. quinquefasciatus*. In adulticidal activity there is 70% death of adult mosquito against *Cx. quinquefasciatus* after 72 hours. The smoke toxicity test showed that out of 200 adult mosquitoes, 190 adult mosquitoes of *Cx. quinquefasciatus* dropped down at the floor after 5 hours of smoke. One toxic compounds was detected having R f =0.853 (80% mortality in 24 hours for

Cx. quinquefasciatus). IR analysis provided preliminary information about the polyhydroxy aliphatic amide nature of the active ingredient. Another study carried out by Anuskha and Dhivya [23] had evaluated the ovicidal activity of extracts of *Carica papaya* leaf for their toxicity against the medicinally important vector mosquitoes (*Culex quinquefasciatus*).The ovicidal activity was observed after 48 hours of exposure. Maximum egg mortality was observed in chloroform extract followed by ethanol and petroleum ether extracts. Phytochemical screening of the leaf extracts showed the presence of bioactive compounds such as alkaloids, tannins, phenols, flavonoids, sterols, terpenoids, saponins, anthroquinones, proteins and quinones. These authors concluded that their investigation leaded the path of exploration of *C. papaya* for eradication of selected important human vector mosquito, thereby, gaining a real momentum to include this plant product for intense mosquito control programme.

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

5. Conclusion

Both ethanolic extracts of *Artemisia annua* and *Carica papaya* linn leaves have repellent activities against vectors of lymphatic filariasis*.* They are found to be effective against the bites of *Culex quinquefasciatus* adult in the current study. However, the repellent effect of *Carica papaya* linn leaves was higher than that of *Artemisia annua* leaves. More effort must be done in order to explore the potentiality of these plant parts available for botanical repellent preparing.

Compliance with ethical standards

Acknowledgments

The authors would like to thank people from Lokossa district surveyed who had helped us in mosquito collection.

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] Lundström JO, Schäfer ML, Hesson JC, BlomgrenE, Lindström A, Wahlqvist P *etal*. The geographic distribution of mosquito species in Sweden. *JEMCA*. 2013;31:21-35

[2] 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 ?. *Malar J*. 2012; 11: 232.

[3] 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.

[4] Ghosh A, Chowdhury N, Chandra G. Plant extracts as potential mosquito larvicides. *Indian J. Med. Res*. 2012; 135: 581-598.

[5] 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.

[6] Regnault-Roger C,Vincent C, Arnason JT. Essential oils in insect control: low-risk products in a high-stakes world. *Ann Rev Entomol*. 2012; 57: 405-424.

[7] Dias CN, Moraes DFC. Essential oils and their compounds as *Aedes aegypti* L.(Diptera:Culicidae) larvicides: review. *Parasitol Res*. 2014; 113: 565-592.

[8] Palsson K, Jaeson TGT. Plant products used as mosquito repellents in Guinea Bissau, West Africa. *Acta Trop*. 1999; 39-52.

[9] 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

[10] Govindarajan M. Larvicidal efficacy of Ficus benghalensis L. plant leaf extract against *Culex quinquefasciatus* Say, *Aedes aegypti* L. and *Anopheles stephensi* L.(Diptera:Culicidae).*Eur Rev Med Pharmacol Sci*.2010;14(2):107-111.

[11] Bhargava S, Agrawal DD, Agrawal OP. Repellent Activity of Essential Oil and Leaf extract of Lantana camara L. In Laboratory condition. *IJTAS*. 2013; 5(1):170-174.

[12] 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.

[13] 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.

[14] 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.

[15] Malbach HI, Johnson HL. Contact urticaria syndrome to diethyl-toluamide (immediate-type hypersensitivity). *Arch Derma.* 1975; 111: 726-730.

[16] Reuveni YH, Yagupsky P. Diethyl toluamide containing insect repellent: adverse effects in worldwide use. *Arch Derma*. 1982; 118: 582-583.

[17] Zadikoff CM.Toxic encephalopathy associated with the use of insect repellent. *J Pediat*.1979; 95: 140-142.

[18] 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.

[19] Edwards DL, Johnson CE. Insect-repellent-induced toxic encephalopathy in a child. *Chin Pharm*. 1987; 6: 496-498.

[20] Mutua MT, Kamindu GN and Wanjohi KE. Ecologically sustainable mosquito repellence using live *Artemisia annua* Linnaeus plants in boarding schools of MWEA rice irrigation scheme, Kenya. *Int. J. Mosq. Res.* 2021; 8(2): 48-53.

[21] Mohan L. Growth Inhibitory Nature of *Artemisia annua* Extract against *Culex quinquefasciatus* (Say). *J Asia-Pac Entomol*. 2006 ; 9(4) :389-395.

[22] Rawani A., Ghosh A., Laskar S., Chandra G. Aliphatic Amide from Seeds of *Carica papaya* as Mosquito Larvicide, Pupicide, Adulticide, Repellent and Smoke Toxicant. *J.Mosq. Res*. 2012, 2(2):8-18.

[23] Anuskha D.U. and Dhivya R. Preliminary phytochemical profiling and ovicidal potential of *Carica papaya* leaf extracts against the filarial vector *Culex quinquefasciatus* (Diptera: Culicidae). *Int. J. Mosq. Res.* 2017; 4(3): 01-08.