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Repellent activities of ethanolic extract of *Artemisia annua* (Asteraceae) and *Carica papaya* linn (Caricaceae) leaves against *Aedes aegypti* mosquitoes in Lokossa 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 *Artemisia annua* and *Carica papaya* linn leaves against *Aedes aegypti* adult mosquitoes in dengue vector control in Lokossa district in south-western Republic of Benin. Larvae of *Aedes aegypti* 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 *Aedes aegypti* 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 *Aedes aegypti* adult mosquitoes.

Keywords: *Artemisia annua*; *Carica papaya* linn; *Aedes aegypti*; dengue; Benin

1. Introduction

Mosquito-borne diseases are major threat to over 2 billion people in the tropics. Several mosquito species belonging to genera Anopheles, Culex, and Aedes are vectors for the pathogens of various diseases like malaria,f ilariasis, Japanese encephalitis, dengue, yellow fever, chikungunya, and so on… [1].

Aedes aegypti is a mosquito that belongs to the Order Diptera and Family Culicidae. This species is the main vector of Dengue and Dengue haemorrhagic fevers in the Americas and Asia [2].It is also incriminated in the transmission of Yellow fever in African urban and peri-urban areas. In some areas it is responsible for transmission of Chikungunya fever (India), Rift Valley fever and Nile encephalitis viruses (Eastern Africa), and Dengue and Dengue haemorrhagic fever in East Africa, Indonesia, Asia and Madagascar.

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 [3-4].

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].

Very few researches were published on the use of essential oils in *Aedes aegypti* 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 extract of *Artemisia annua* and *Carica papaya* linn leaves against *Aedes aegypti* adult mosquitoes in dengue 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 *Aedes aegypti* adult mosquitoes in dengue 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

Aedes aegypti 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. 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.

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 2 Farm of *Artemisia annua*

Figure 3 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 100 grammes 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/cm², 4mg/cm² and 5 mg/cm². 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) *Aedes aegypti* adult mosquito 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/cm², 2mg/cm², 3mg/cm², 4mg/cm²and 5 mg/cm² 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 *Aedes aegypti* 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

The analysis of figure 4 showed that *Aedes aegypti* 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 forty five (45) minutes with all the concentrations tested of 1mg/cm², 2mg/cm², 3mg/cm², 4mg/cm² and 5mg/cm².

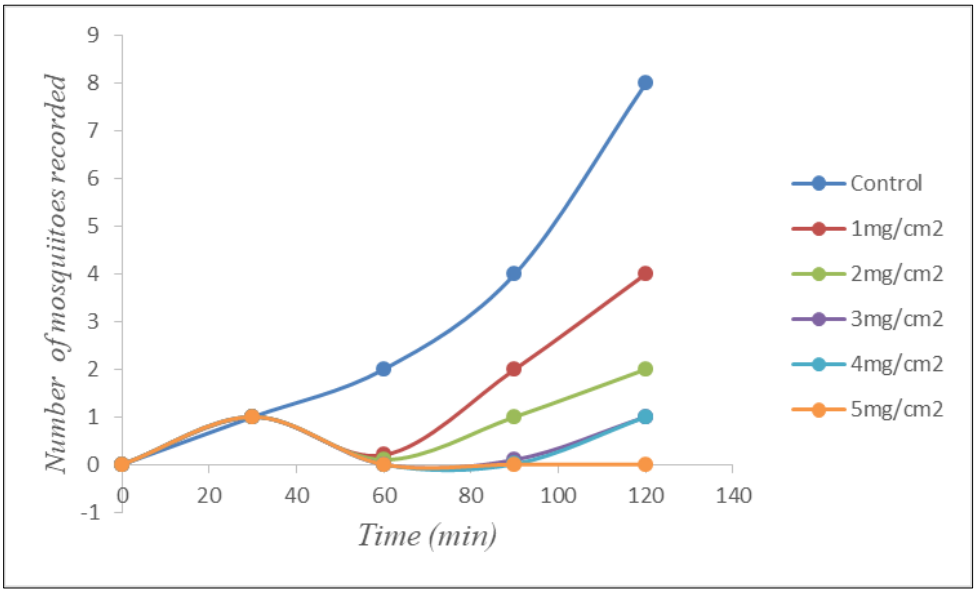

Figure 4 Repellent activity of *Artemisia annua* leaves on *Aedes aegypti* female adult mosquitoes

In similar way, the analysis of figure 5 showed that *Aedes aegypti* female adult mosquitoes preferred go towards the hand coated with ethanol (control hand) than the hand coated with ethanolic extract of *Carrica papaya* Linn leaves (test

hand) with all tested concentrations. The repellent activities were detected after thirty (30) minutes with all the concentrations tested of 1mg/cm², 2mg/cm², 3mg/cm², 4mg/cm² and 5mg/cm².

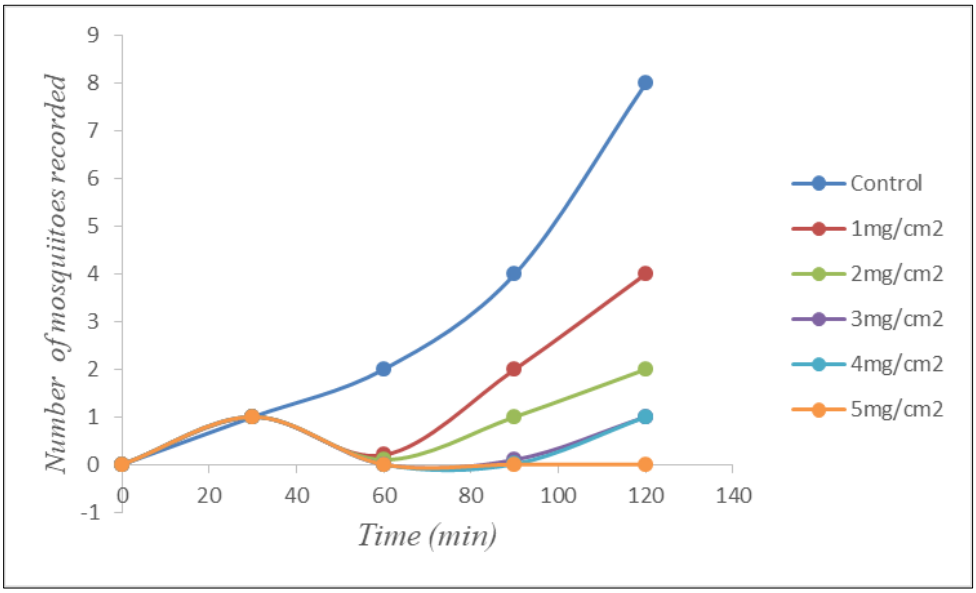

Figure 5 Repellent activity of *Carica papaya* Linn leaves on *Aedes aegypti* female adult mosquitoes

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 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.	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)

4. Discussion

Dengue is the most important vector-borne human disease caused by mosquito vector *Aedes aegypti*. Of the various strategies adopted for eliminating this disease, controlling of vector through herbs has been reckoned as one of the important measures for preventing this resurgence. So, the development of resistance to chemical insecticides among mosquito species has been considered as a setback in vector control. This has necessitated the need for a research and development of eco-friendly, biodegradable indigenous method for vector control.

Although the most common mosquito repellent currently available on the market, deet (N,N-diethyl-3 methylbenzamide) has shown excellent protectionfrom mosquito bites [10-12], there were reports of toxicity problems after application of DEET, range from mildeffects such as contact urticaria [13] and skin eruption [14], to severe reactions, such as toxic encephalopathy [15-17]. 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, *Aedes aegypti* 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 forty five (45) minutes with all the concentrations tested of 1mg/cm^2, 2mg/cm^2, 3mg/cm^2, 4 mg/cm^2 and 5mg/cm^2. These results showed that the ethanolic extract of *Artemisia annua* leaves had repellent activities against *Aedes aegypti* female adult mosquitoes. Our results corroborated with those obtained by Gaurav *et al* [18] who stated that *Artemisia annua* leaf extract has a strong larvicidal potential against *Aedes aegypti* L. by using chloroform solvent. It has a correlated value with the present study. In their study, *Artemisia annua* leaf chloroform extract when tried against larvae of *Aedes aegypti* has shown a strong larvicidal activity against this vector with respective LC50 and LC90 values of 0.67 and 5.84 ppm for *Aedes aegypti*. The respective marker compound purified from the individual fractions A, B and C, were Artemisinin, Arteannuin B and Artemisinic acid, as confirmed and characterized through FT-IR and NMR. Another study was carried out by Cheah *et al* [19] who had studied the effects of *Artemisia annua* (Asterales: Asteraceae) against *Aedes aegypti* (Diptera: Culicidae) with good results. In fact, their study focused on the larvicidal, oviposition, and ovicidal effects of a crude extract of *Artemisia annua* against *Aedes aegypti*. Dried cells of *Artemisia annua* from cell suspension cultures were extracted using hexane. The extract showed moderate larvicidal effects against mosquitoes. At 24 hours post treatment, the LC50 value for *Aedes aegypti* was recorded as 276.14 ppm. The percentage mortality of larvae was directly proportional to the tested concentration. The results indicated that the *Artemisia annua* extract showed concentration-dependent oviposition deterrent activity and had a strong deterrent effect. At 500 ppm, the percentage effective repellency was more than 85 % compared with the control group with oviposition activity index value of -0.94 for *Aedes aegypti*. *Artemisia annua* was found to be more effective against *Aedes aegypti*. Their study indicated that crude extract of *Artemisia annua* could be a potential alternative for use in vector management programs.

In the current study, *Aedes aegypti* female adult mosquitoes also preferred go towards the hand coated with ethanol (control hand) than the hand coated with ethanolic extract of *Carrica papaya* Linn leaves (test hand) with all tested concentrations. The repellent activities were detected after thirty (30) minutes with all the concentrations tested of 1mg/cm^2, 2mg/cm^2, 3mg/cm^2, 4 mg/cm^2 and 5mg/cm^2. These results showed that the ethanolic extract of *Carica papaya* Linn leaves had repellent activities against *Aedes aegypti* female adult mosquitoes. Our results corroborated with those obtained by Malathi *et al* [20] who had evaluated the mosquito larvicidal effect of *Carica Papaya* against *Aedes Aegypti* with good results. In their study, mosquito larvicidal efficacy and phytochemical screening of crude aqueous and ethanol extracts of the leaf, bark, root and seed of *Carica papaya* of family Caricaceae a common traditional medicinal plant were evaluated. Phytochemical screening of the selected parts' crude aqueous and ethanol extracts indicated the presence of alkaloids, carbohydrates, saponin, phenol, tannin, flavones, coumarins, anthocyanin and flavanoids which are known to possess medicinal and insect larvicidal properties. Larvicidal effect of 2nd and 4th instar larvae of mosquito species *Aedes aegypti* have been investigated for 24hours with 1mg/ml concentration of extracts of selected parts of *Carica papaya.* The results clearly indicated that there is a significant variation among the aqueous and ethanol extracts of the selected parts of the plant. The seed extracts are observed as effective larvicide than other selected parts of *Carica papaya*.

Dengue Hemorrhagic Fever DHF is caused by dengue virus is transmitted through the bite of the *Aedes aegypti* mosquito, which often occurs in various regions. Due to the mosquito-borne virus that causes the disease from live around us. Various prevention efforts have been done by using an electric mosquito repellent with papaya extract active ingredients as vegetable insecticide. Another study was carried out by Rasman *et al* [21] to determine the ability of papaya extract in controlling the *Aedes aegypti* mosquito. This type of research is an experiment, with the active ingredient papaya extract concentrate of 50%, 60%, and 70% in light weather conditions. Their results showed that papaya leaf extract, the first 15 minutes pulled all the mosquitoes in a cage and there is no death. At a concentration of 50% LC50 is reached after 45 minutes on average *Aedes aegypti* dead reached 11 tails. LC50 at 60% concentration is reached after 30 minutes on average *Aedes aegypti* dead reached 16 tails. LC50 at 70% concentration is reached after 30 minutes on average observation *Aedes aegypti* dead reached 17 tails. This mosquito smell the papaya extract containing bitter substances karpain an insecticide plant alkaloid that is not favored by mosquitoes. Alkoloid karpain that has the characteristics of safe neurotoxin when inhaled by humans. As an insect neurotoxin exposure to toxins is generally experiencing spasms and paralysis before death. Research papaya extract is able to control the mosquito *Aedes aegypti* with standard LC50. Concentration of 60% papaya extract more effective than a concentration of 50% and 70%. These authors recommended to people to take advantage of papaya leaf extract as an alternative vegetable insecticide.

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 dengue vectors. They are found to be effective against the bites of *Aedes aegypti* 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

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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] Service MW. Mosquitoes (Culicidae). In: Lane RP, Crosskey RW. Medical insects and arachnids. Chapman & Hall, London. 1993: 723-725.

[2] Lenhart AE, Walle M, Cedillo H, Kroeger A. Building better ovitraps for detecting *Aedes aegypti* oviposition Acta Trop. 2005; 96:56-59.

[3] 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.

[4] Toé KH, Jones CM, Sagnon N'F, Ismail HM, Dabiré RK, RansonH. Increased pyrethroid resistance in malaria vectors and decreased bed net effectiveness, Burkina Faso. Emerg Infect Dis. 2014; 20(10): 1691-1696.

[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] 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.

[11] 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.

[12] Thawara U, Tawatsin A, Chompoosri J. Laboratory and evaluation of the insect repellent 3535 (Ethyl butyl acetylaminopropionate) and deet against the insect mosquito vectors in Thailand. J Am Mosquito Control Assoc. 2001; 17: 190 -195.

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

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

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

[16] 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.

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

[18] Gaurav S, Himanshi K, Madhu C, Kaushal K, Veena A. Strong larvicidal potential of Artemisia annua leaf extract against malaria (*Anopheles stephensi* Liston) and dengue (*Aedes aegypti* L.) vectors and bioassay-driven isolation of the marker compounds. Parasitol Res. 2014; 113: 197-209.

[19] Cheah S-X, Tay J-W, Chan L-K, Jaal Z. Larvicidal, oviposition, and ovicidal effects of *Artemisia annua* (Asterales: Asteraceae) against *Aedes aegypti*, *Anopheles sinensis*, and *Culex quinquefasciatus* (Diptera: Culicidae). Parasitol Res. 2013; 9: 3275-3282.

[20] Malathi P, Vasugi SR. Evaluation of mosquito larvicidal effect of *Carica Papaya* against *Aedes Aegypti*. Int. J. Mosq. Res. 2015; 2(3): 21-24.

[21] Rasman, La Taha and Juhaerah. Ability of Papaya Leaf Extract (*Carica papaya*) in Controlling *Aedes Aegypti* Mosquito (Experimental Study). IJSBAR. 2015; 24(3):164-172.