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REPELLENT ACTIVITIES OF ETHANOLIC EXTRACT OF HYPTIS SUAVEOLENS LINN (LAMIACEAE) AND AZADIRACHTA INDICA (MELIACEAE) LEAVES AGAINST CULEX QUINQUEFASCIATUS MOSQUITOES IN LOKOSSA DISTRICT IN SOUTH-WESTERN BENIN, WEST AFRICA

Entomology

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ABSTRACT

Introduction: 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. **Objectives:** The current study was aimed to evaluate the repellent activities of ethanolic extract of Hyptis suaveolens Linn and Azadirachta indica leaves against Culex quinquefasciatus adult mosquitoes in lymphatic filariasis vector control in Lokossa district in south-western Republic of Benin. **Methodology:** 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/cm2, 2mg/cm2, 3mg/cm2, 4mg/cm2 and 5mg/cm2. **Results:** The results showed that the leaves of both plants had repellent effects against Culex quinquefasciatus adult mosquitoes. However, the repellent effect of Azadirachta indica leaves was higher than that of Hyptis suaveolens Linn leaves. **Conclusion:** Both ethanolic extracts of Hyptis suaveolens Linn and Azadirachta indica leaves are found to be effective in the repellency against the bites of Culex quinquefasciatus adult mosquitoes.

KEYWORDS

Hyptis suaveolens Linn, Azadirachta indica, Culex quinquefasciatus, lymphatic filariasis, Benin

INTRODUCTION

Mosquitoes are the most important vectors of certain human infections and diseases. Mosquito-borne diseases such as malaria, yellow fever, filariasis and dengue contribute significantly to mortality in most tropical countries. Culex quinquefasciatus Say known as the southern house mosquito, is a subtropical mosquito belonging to the complex Culex pipiens, present in the Americas, Australia, Asia, Africa, Middle East and New Zealand. In addition to the considerable discomfort caused by the nocturnal biting behavior, Culex quinquefasciatus is the main vector of several pathogens, especially including the nematode Wulchereria bancrofti (agent of bancroftian filariasis) and the West Nile virus [1,2].

The major problems associated with the use of chemicals for the control of pests including mosquitoes include: the development of resistance to the chemicals, issues around the residues in animals and the environment and their undesirable side effects [3].

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.

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 [4,5].

The most economical method of controlling mosquitoes is by eradicating their breeding sites using environmentally friendly botanicals. And so, the search for alternative vector control measure using botanicals that do not produce adverse effects on the ecosystem remains a top priority for scientists today[6]. For this reason, searching for alternative therapies in handling of mosquitoes and some infectious vectors is necessary especially from eco-friendly and readily available phytochemicals.

The potential of plants as insecticide, repellent or fumigant sources against mosquitoes and other pest and vectors insects is well known [7,8,9]. 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 [10,11].

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.

Objectives

To evaluate the repellent activities of ethanolic extract of Hyptis suaveolens Linn and Azadirachta indica leaves against Culex quinquefasciatus adult mosquitoes in lymphatic filariasis vector control in Lokossa district in south-western Republic of Benin.

MATERIAL AND METHODS

Study Area

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 260 km^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. These factors were important to evaluate the repellent activities of ethanolic extract of Hyptis suaveolens Linn and Azadirachta indica 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.

Figure 1: Map of Republic of Benin Showing Lokossa District Surveyed

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

Collection of the Plant Leaves

The leaves of Hyptis suaveolens Linn and Azadirachta indica were collected in their predilection areas in Lokossa district.

Figure 3: Tree of Hyptis Suaveolens

Figure 4: Tree of Azadirachta Indica

Plant Leaves Extraction

To extract Hyptis suaveolens Linn and Azadirachta indica 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 Hyptis suaveolens Linn and Azadirachta indica leaves separately.

Performing of Repellent Activities Test of Hyptis Suaveolens Linn and Azadirachta Indica Leaves

A batch of twenty five (25) Culex quinquefasciatus 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/cm^2, 2mg/cm^2, 3mg/cm^2, 4mg/cm^2 and 5 mg/cm^2 of ethanolic extract of Hyptis suaveolens Linn and Azadirachta indica 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.

RESULTS

Evaluation of Repellent Activity of Hyptis Suaveolens Linn and Azadirachta Indica

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 Hyptis suaveolens Linn leaves (test hand) with all tested concentrations. The repellent activities were detected after forty (40) minutes with all the concentrations tested of 1mg/cm^2, 2mg/cm^2, 3mg/cm^2, 4 mg/cm^2 and 5mg/cm^2.

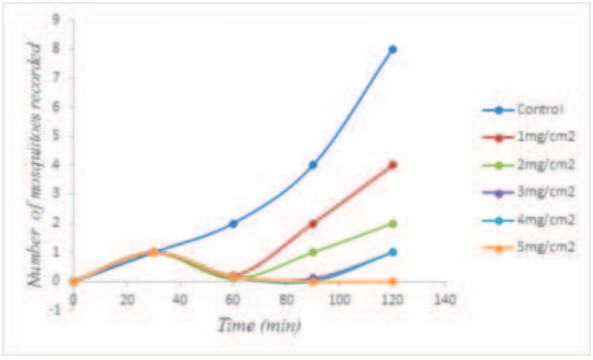

Figure 5: Repellent Activity of Hyptis Suaveolens Linn Leaves on Culex Quinquefasciatus Female Adult Mosquitoes

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 Azadirachta indica leaves (test hand) with all tested concentrations. The repellent activities were detected after thirty five (35) minutes with all the concentrations tested of 1mg/cm^2, 2mg/cm^2, 3mg/cm^2, 4mg/cm^2 and5mg/cm^2.

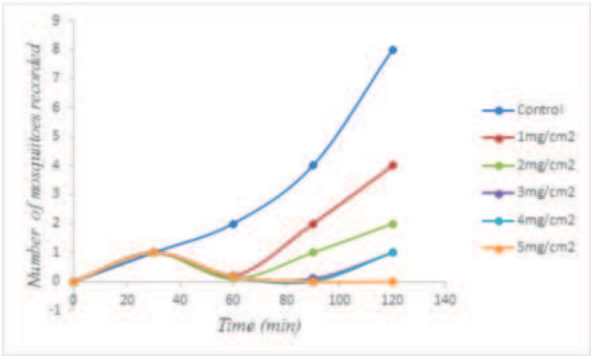

Figure 6: Repellent Activity of Azadirachta Indica Leaves on Culex Quinquefasciatus Female Adult Mosquitoes

The table 1 showed the advantages and disadvantages of the use of ethanolic extract of Hyptis suaveolens Linn and Azadirachta indica leaves.

Table 1: Advantages and Disadvantages of the Use of Ethanolic Extract of Plant Leaves

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

DISCUSSION

Mosquito vector control is facing a threat due to the emergence of resistance to synthetic insecticides. Safe and eco-friendly insecticides of botanical origin may serve as suitable alternative biocontrol techniques in the future. So, mosquito-borne diseases have intruded the globe since immemorial time. The present scenario for commanding the mosquitoes is aimed at application of target and stage-specific, cost-effective and biodegradable phytoproducts. Plant extracts are safer for non-target organisms including man. Plant based formulations would be more feasible environmental products with proven potential as insecticide.

Although the most common mosquito repellent currently available on the market, DEET (N,N-diethyl-3-methylbenzamide) has shown excellent protection from mosquito bites [12,13,14], 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[17,18,19]. 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 Hyptis suaveolens Linn leaves (test hand) with all tested concentrations. The repellent activities were detected after forty (40) 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 Hyptis suaveolens Linn leaves had repellent activities against Culex quinquefasciatus female adult mosquitoes. Our results corroborated with those obtained by Ekesiobi and Igbodika [20] who had investigated the repellent and larvicidal activity of the ethanol extract of this herbs against the filarial vector, Culex quinquefasciatus 3rd instar larvae and adults respectively. The extract was screened chemically for phytochemicals. The extracts showed larvicidal and repellent activity with Lethal Concentration, LC 50, LC 90 and Effective Dose, ED 50 , ED 90 values 81.817mg/l, 166.704mg/g and 38.75mg/l , 258.925mg/l respectively. Alkaloids, flavonoids, Tannins, Saponins and Steroids were the phytochemicals present. Repellency and mortality were observed to significantly and positively correlated with the concentrations of the ethanol extracts ($p<0.05$). The present study revealed that an indigenous common weed, Hyptis suaveolens could be considered as potential natural larvicidal and repellent agent against mosquitoes. Another study carried out by Sakthivadivel et al [21] had studied the mosquito larvicidal activity of Hyptis suaveolens (L.) Poit (Lamiaceae) aerial extracts against the filarial vector Culex quinquefasciatus Say (Diptera: Culicidae) with good results. In fact, these authors had studied more precisely the larvicidal activity of crude petroleum ether, chloroform and acetone aerial extracts of Hyptis suaveolens against the filarial vector, Culex quinquefasciatus. Standard WHO protocols with minor modifications was adopted for the larvicidal bioassay. Larvicidal activity was evaluated at concentrations of 62.5, 125, 250, 500 and 1000 mg/L. Larval mortality was observed for 24 and 48 hours. Among the solvent extracts tested, acetone exhibited highest larvicidal activity and LC 50 values was 485.61 followed by petroleum ether and chloroform extract which were 493.44 and 625.97 mg/L after 24 hours. In the case of 48 hours, petroleum ether extract (LC 50 298.76 mg/L) was found to exhibit highest larvicidal activity followed by acetone (LC 50 344.03 mg/L) and chloroform (LC 50 429.50 mg/L). Further investigations are needed to explore the larvicidal activity of acetone and petroleum ether aerial extract of this plant against a wide range of mosquito species and also the active ingredient(s) of the extract responsible for larvicidal activity should be identified. Hyptis sauveolens (bush minth) belonging to the family, Lamiaceae is common and widely distributed in Republic of Benin.

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 Azadirachta indica leaves (test hand) with all tested concentrations. The repellent activities were detected after thirty five (35) 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 aAzadirachta indica leaves had repellent activities against Culex quinquefasciatus female adult mosquitoes. Our results corroborated with those obtained by Mandal [22] who had evaluated the repellent activity of Eucalyptus and Azadirachta indica (A. indica) seed oil against filarial mosquito Culex quinquefasciatus (Cx. quinquefasciatus) from Purulia district of the West Bengal state, India. The repellent activity of Eucalyptus and A. indica seed oils (using

coconut oil base) against Cx. quinquefasciatus mosquito were evaluated in indoor conditions. Three concentrations, 0%, 50% and 100% (v/v) of both the agents were considered in their study. The protection percentage was determined, and the protection time was recorded. The test oils showed excellent repellent action against Cx. quinquefasciatus. The A. indica seed oil provided 90.26% and 88.83% protection, and the Eucalyptus oil 93.37% and 92.04%, at concentrations 50% and 100% (v/v), respectively, with the protection time up to 240 min. There was no bite within 120 min and 180 min, respectively, due to the action of Eucalyptus and A. indica seed oil, and thus 100% protection from the bite of Cx. quinquefasciatus mosquito was achieved. Their study clearly demonstrates the potential of Eucalyptus and A. indica seed oils as topical repellents against Cx. quinquefasciatus, the mosquito vector of filariasis. Another study carried out by Chatterjee et al [23] had evaluated the neem-based products as potential eco-friendly mosquito (including Cx. quinquefasciatus) control agents over conventional eco-toxic chemical pesticides with good results. In fact, mosquitoes cause serious health hazards for millions of people across the globe by acting as vectors of deadly communicable diseases like malaria, filariasis, dengue and yellow fever. Use of conventional chemical insecticides to control mosquito vectors has led to the development of biological resistance in them along with adverse environmental consequences. In this light, the recent years have witnessed enormous efforts of researchers to develop eco-friendly and cost-effective alternatives with special emphasis on plant-derived mosquitocidal compounds. Neem oil, derived from neem seeds (Azadirachta indica A. Juss, Meliaceae), has been proved to be an excellent candidate against a wide range of vectors of medical and veterinary importance including mosquitoes. It is environment-friendly, and target-specific at the same time. The active ingredients of neem oil include limonoids like azadirachtin A, nimbin, salannin and numerous other substances that are still waiting to be discovered. Of these, azadirachtin has been shown to be very effective and is mainly responsible for its toxic effects. The quality of the neem oil depends on its azadirachtin content which, in turn, depends on its manufacturing process. Neem oil can be used directly or as nanoemulsions or nanoparticles or even in the form of effervescent tablets. When added to natural breeding habitat waters they exert their mosquitocidal effects by acting as ovicides, larvicides, pupicides and/or oviposition repellents. The effects are generated by impairing the physiological pathways of the immature stages of mosquitoes or directly by causing physical deformities that impede their development. Neem oil when used directly has certain disadvantages mainly related to its disintegration under atmospheric conditions rendering it ineffective. However, many of its formulations have been reported to remain stable under environmental conditions retaining its efficiency for a long time. Similarly, neem seed cake has also been found to be effective against the mosquito vectors. The greatest advantage is that the target species do not develop resistance against neem-based products mainly because of the innumerable number of chemicals present in neem and their combinations. This makes neem-based products highly potential yet unexplored candidates of mosquito control agents. The current review helps to elucidate the roles of neem oil and its various derivatives on mosquito vectors of public health concern.

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

CONCLUSION

Both ethanolic extracts of Hyptis suaveolens Linn and Azadirachta indica 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, the repellent effect of Azadirachta indica leaves was higher than that of Hyptis suaveolens Linn leaves. More effort must be done in order to explore the potentiality of these plant parts available for botanical repellent preparing.

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Conflict of Interest

We declare that there is no conflict of interest regarding the publication of this article.

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REFERENCES

1. Brasil. (2011). Guia de vigilância do Culex quinquefasciatus. Ministério da Saúde, Secretaria de Vigilância em Saúde, Departamento de Vigilância Epidemiológica. Série A. Normas e Manuais Técnicos 1ª Edição, 76 pages.
2. Lima-Camara TN. (2016). Emerging arboviruses and public health challenges in Brazil. Rev Saude Publica, 50:36. https://doi.org/10.1590/S1518-8787.2016050006791.
3. Peter RJ, Van den Bossche P, Penzhorn BL, Sharp B. (2005). Tick, fly and mosquito control-lessons from the past, solutions for the future. Veterinary Parasitology, 132, 205-215. https://doi.org/10.1016/j.vetpar.2005.07.004.
4. Badolo A, Traore A, Jones CM, Sanou A, Flood L, Guelbeogo WM, Ranson H, Sagnon NF. (2012). Three years of insecticide resistance monitoring in Anopheles gambiae in Burkina Faso: resistance on the rise ?. Malaria Journal, 11, 232. https://doi.org/10.1186/1475-2875-11-232.
5. Toé KH, Jones CM, Sagnon N'F, Ismail HM, Dabiré RK, Ranson H. (2014). Increased pyrethroid resistance in malaria vectors and decreased bed net effectiveness, Burkina Faso. Emerging Infectious Deseases, 20(10), 1691-1696. https://doi.org/10.3201/eid2010.140619.
6. World Health Organization. (2013). Test procedures for insecticide resistance monitoring in malaria vector mosquitoes. ISBN 978 924 150515 4. Retrieved 21 June 2019. Pp 18–20.
7. Lucia A, Juan LW, Zerba EN, Harrand L, Marcó M, Masuh HM. (2012). Validation of models to estimate the fumigant and larvicidal activity of Eucalyptus essential oils against Aedes aegypti (Diptera:Culicidae). Parasitology Research, 110 (5), 1675-1686. https://doi.org/10.1007/s00436-011-2685-9.
8. Regnault-Roger C, Vincent C, Arnason JT. (2012).Essential oils in insect control: low-risk products in a high-stakes world. Annual Rewiew of Entomology, 57, 405-424. https://doi.org/10.1146/annurev-ento-120710-100554.
9. Dias CN, Moraes DFC. (2014). Essential oils and their compounds as Aedes aegypti L.(Diptera:Culicidae) larvicides: review. Parasitology Research 113, 565-592. https://doi.org/10.1007/s00436-013-3687-6.
10. Palsson K, Jaeson TGT. (1999). Plant products used as mosquito repellents in Guinea Bissau, West Africa. Acta Tropica, 72 (1), 39-52. https://doi.org/10.1016/s0001-706x(98)00083-7.
11. Seyoum A, Kabiru EW, Lwande W, Killeen GF, Hassanali A, Knols BGJ. (2002). Repellent of live plants against Anopheles gambiae from human baits in semi-field experimental huts. American Journal of Tropical Medicine and Hygiene, 67(2), 191-195. https://doi.org/10.4269/ajtmh.2002.67.191.
12. Yap HH. (1986). Effectiveness of soap formulations containing deet and permethria as personal protection against outdoor mosquitoes in Malaysia. Journal of American Mosquitoes Control Association, 2(1), 63-67.
13. Walker TW, Robert LL, Copeland RA. (1996). Field evaluation of arthropod repellents, Deet and piperidine compound against Anopheles arabiensis in Western Kenya. Journal of American mosquito Control Association, 12, 172-176.
14. Thawara U, Tawatsin A, Chompoosri J. (2001). Laboratory and evaluation of the insect repellent 3535 (Ethyl butylacetylaminopropionate) and deet against the insect mosquito vectors in Thailand. Journal of American Mosquito Control Association, 17, 190 -195.
15. Malbach HI, Johnson HL. (1975). Contact urticaria syndrome to diethyl-toluamide (immediate-type hypersensitivity). Arch Derma, 111(6), 726-730. https://doi.org/10.1001/archderm.111.6.726.
16. Reuveni YH, Yagupsky P. (1982). Diethyl toluamide containing insect repellent: adverse effects in worldwide use. Arch Derma, 118 (8), 582-583.
17. Zadikoff CM. (1979). Toxic encephalopathy associated with the use of insect repellent. Journal of Pediatrics, 95(1), 140-142. https://doi.org/10.1016/s0022-3476(79)80109-2.
18. Roland EH, Jan JE, Rigg JM. (1985) .Toxic encephalopathy in a child after a brief exposure to insect repellents. Canadian Medical Association, 132(2), 155-156.
19. Edwards DL, Johnson CE. (1987). Insect-repellent-induced toxic encephalopathy in a child. China Pharma, 6(6), 496-498.
20. Ekesiobi AO and Igbodika MC. (2015). Evaluation of repellent and larvicidal activity of Hyptis suaveolens against filarial vector, Culex quinquefasciatus. American Academic & Scholarly Research Journal, 7(6), 78-88.
21. Sakthivadivel M., Gunasekaran P., Sivakumar M., Arivoli S., Raveen R. and Tennyson S. (2015). Mosquito larvicidal activity of Hyptis suaveolens (L.) Poit (Lamiaceae) aerial extracts against the filarial vector Culex quinquefasciatus Say (Diptera: Culicidae). Journal of Medicinal Plants Studies, 3(4): 1-5.
22. Mandal S. (2011). Repellent activity of Eucalyptus and Azadirachta indica seed oil against the filarial mosquito Culex quinquefasciatus Say (Diptera: Culicidae) in India. Asian Pacific Journal of Tropical Biomedicine, 1(1), S109-S112.
23. Chatterjee S, Bag S, Biswal D, Paria DS, Bandyopadhyay R, Sarkar B, Mandal A, Dangar TK. (2023). Neem-based products as potential eco-friendly mosquito control agents over conventional eco-toxic chemical pesticides-A review. Acta Tropica, 240, 106858. https://doi.org/10.1016/j.actatropica.2023.106858.