Larvicidal Effects of *Hyptis suaveolens* and *Chenopodium ambrosoides* on Anopheles Mosquito Larvae

A.Dawet., A. G.Ikani and D. P. Yakubu

Abstract- Anopheles mosquitoes which are vectors of many disease pathogens pose a great threat to global health and attention on control is spreading from adult to include developmental stages. Laboratory experiments were conducted to evaluate the larvicidal activity of ethanolic extracts of leaves of two plants. H. suaveolens and C. ambrosoides at various concentrations ranging from 0, 300, 350, 400 and 500 mg/l against 3rd and 4th instar larvae of the Anopheles mosquito. Mortality was recorded from 3, 6 and 24 hours after administration of the plant extracts and the LC 50 value was obtained at 400mg/l and 450 mg/l respectively. The larvicidal effect of each plant extract was compared with the synthetic insecticides (Rambo). The two tested plant extracts in their different concentrations have shown larvicidal effect on Anopheles mosquito larvae. At 300 mg/l, mortality was 12.22 % and 11.11 % for H. suaveolens and C. ambrosoides respectively. While at 500 mg/l, H. suaveolens caused 90.00 % mortality and C. ambrosoides caused 84.44 % mortality rate. Statistical analysis shown that, there was highly significant difference ($p<0.05$) in the mean mortality of Anopheles species larvae across concentration after 3, 6 and 24 hours exposure time using H. suaveolens and C. ambrosoides respectively. While there was no significant difference ($p>0.05$) in the mean mortality of Anopheles species larvae between extracts of both plant species after 3, 6 and 24 hours exposure time respectively. The results indicate that H. suaveolens extract had the highest larvicidal effect against Anopheles species larvae with the lowest LC 50 of 354.81 mg/l and 24 hours upper and lower confidence s limit of 585.44 and 236.54 followed by C. ambrosoides extrac with highest LC 50 of 389.05 mg/l and 24 hours upper and lower confidences limit of 825 and 345 respectively. This study suggests that, the leaf extracts of the two plant species should be considered as promising larvicides against Anopheles mosquito larvae.

Keywords: Insecticidal plants, Hyptis suaveolens, Chenopodium ambrosoides, Anopheles larvae.

INTRODUCTION

Mosquitoes are of immense importance because they are able to host and transmit various diseases pathogens species of virus, protozoa and nematodes, till today; they pose a threat to health [1]. In Jos, Plateau State, Nigeria [2] reported on the abundance and intense breeding activity of public health importance mosquitoes of potential filariasis vectors: *Culex quinquefasciatus*, *Cx. decens*, *Cx. vittatus*; as well as the principal malaria carrier-*Anopheles gambiae* S.L. An association of brain tumor incidence and malaria suggests that *Anopheles* might transmit a virus or other agent that could cause brain tumor [3]. According to the released in December 2013, there were about 207 million cases in 2012 (with an uncertainty range of 135 million to 287 million) and an estimated 627000 deaths (with an uncertainty range of 473000 to 789000) [4].

Studies on the economic burden that malaria places in Nigeria revealed that 12.0 % of Gross Domestic Product (GDP) representing about N881 millions is lost annually due to malaria [5]. Mosquito born-diseases are prevalent almost in all Africa countries and infecting over millions of people every year globally but less in developed countries. They act as a vector for most of the life threatening diseases like malaria, filariaisis, yellow fever, denque fever, encephalitis, and virus infection e.t.c., in almost all tropical and subtropical countries and other part of the world.

To prevent proliferation of mosquito born-diseases and to improve quality of environment and public health, control is essential. The control of mosquitoes has received some attention based on the study of ecology and distribution with field trials of pesticides aimed at controlling and eradicating it. The major tools in mosquitoes control operation are the application of synthetic insecticides such as organochlorine and organophosphate compounds. The microencapsulated formulation of pirimiphos methyl (Actellic 300CS) is a highly effective and appropriate insecticide for indoor residual spraying (IRS) use in Zanzibar as it showed a relatively prolonged residual activity compared to other products used for the same purpose. The insecticide extends the residual effect of IRS thereby making it possible to effectively protect communities with a single annual spray round reducing overall costs. Thus, the encapsulated formulation of pirimiphos-methyl proved to be a useful alternative to other insecticides for resistance management plans in Zanzibar [6]. In a study on the resistance status, [7] reported that wild populations of *Culex quinquefasciatus* have developed resistance against pyrethroids, organochlorine and carbamate. Studies by [8] also reported that pyrethroid and DDT resistance was widespread in malaria vector in Benin and there is a correlation between the resistance degree of a mosquito to permethrin and DDT and the time that this mosquito takes to react to these products. This situation of

resistance may seriously jeopardize the efficacy of Insecticide Residual Spray (IRS) and Long-Lasting Insecticide nets (LLINs) on which, most African countries including Benin, rely to reduce malaria transmission. However, due to lack of novel insecticides, high cost of synthetic insecticides, concern for environmental sustainability, harmful effect on human health and other non-target populations, high rate of biodegradable nature, high rate of biological magnification through ecosystem, and increasing insecticides resistances on a global scale [9], [10].

An effective alternative approaches is to explore the floral biodiversity and enter the field of using safer insecticides of botanical origin as a simple and suitable method of mosquito control, unlike synthetic insecticides which are based on a single active ingredient. [11] reviewed the current state of knowledge on larvicidal plant species extraction process, growth and reproduction inhibiting phytochemicals, botanical ovicides, synergistic, additive and antogonistic joint action effects of mixture, residual capacity, effects on non-target organisms, resistance and screening methologises and discussed some promising advances made in phytochemical research.

H. suaveolens; (family lamiaceae) is a common occurring herb found mainly amongst the tall savannah grasses in the middle belt and eastern part of Nigeria. It possesses a characteristic irritating smell which disseminates in the air. This quality character led to its widespread used as a mosquito repellent in some parts of Nigeria. One such place is Ala community of Idah Local Government Area of Kogi state where the smoke emanancting from the burned leaves is used by the inhabitant to derive the lot of mosquitoes. *C. ambrosoides* (family Chenopodiacea) is used by inhabitant of Jos Plateau State, Nigeria to treat cough and dysentery. It is common herb found mainly among the savannah tall tree grasses that possesses a characteristic irritating smell which disseminate in the air. More importantly, the leaves are burnt by the local inhabitant to

generate smoke to ward-off housing haunting mosquitoes. This mosquito repellence quality prompted the investigation of the insecticidal potentials of these plants against *Anopheles* laevae.

MATERIALS AND METHODS

Collection of plant samples.

Leaves *of H. suaveolens* were collected from Ala farming Area of Idah Local Government Area, Kogi State while *C. ambrosoides* leaves were collected from senior staff quarter of the University of Jos, Plateau State, Nigeria. The leaves of both plants were first authenticated in the Department of Pharmacognosy, University of Jos where the study was conducted. The leaves were then air-dried under shed at room temperature and pounded using mortar and pistle.

Extraction of plant materials.

70g each of *H. suaveolens* and *C. ambrosoides* powdered was weight and soaked separately in 300ml of 70 % Ethanol. These were allowed to stand overnight and slackened for 3 hours. The content of each was filtered and the filtrate was evaporated to dryness on a water bath. The extract was transferred into sample container and preserved desiccators. The percentage (%) yield was determined and crude extract were stored in a refrigerator. Prior to use they were prepared into the desired series of solution: 0, 300,350, 400,450 and 500 mg/l

Phytochemical screening

The ethanolic crude extracts were screened for their phytochemical constituents and extract were evaluated for the presence of alkaloids, tannins, saponins, flavonoids, carbohydrate, steroids, anthraquinones, cardiac glycosides using the methods of [12], [13].

Standard insecticide

The synthetic insecticide used was a commercial Rambo (emulsifiable concentrate) containing active ingredient of the residual 0.50 %, Dichlovos, 0.2 0 % permethrin, and 0.15

% transflurin. Test solution were formulated by dissolving 2ml of the liquid material in 100ml of distilled water and mixed vigorously.

Collection, identification and rearing of mosquito larvae

Larvae of *Anopheles* mosquitoes were collected from a pools of stagnant water near the Department of Physics Bauchi Road Campus, back of Village student hostel of the University of Jos and around Police barracks, Division 'A', Police Headquarter Jos, Plateau State following the method described by [14]. A container containing any amount of water was considered as wet container and the wet container containing any number of immatures (larvae, pupae or both) was considered as positive container. The immatures were collected by using different immature collecting materials like pipettes, dipper, strainer depending upon the type and size of breeding source. The collected immatures were kept in plastic containers labeled with the code of breeding source. The collected larvae and emerged adults (Some larvae were allowed to moult into adults' mosquitoes for proper identification by an entomologist) were pinned and identified under microscope to separate them according to species by using the standard taxonomic keys [15]. The larvae of *Anopheles* mosquito were differentiated and kept in a separate container. Anopheline larvae have abdominal palmate hairs, tergal plate and are surface feeder. The larvae were kept in a plastic container in netted enclosure in the Undergraduate Research Laboratory, Department of Zoology, University of Jos which had sufficient air and light and were fed with whole ban wheat biscuits and yeast and the water was changed daily. They were monitored under laboratory condition.

Bioassay

Twenty one laboratory test beakers (250ml) were divided into seven groups each with three replicate and kept on a clean sterilised laboratory bench for each plant and the

controls. The arrangements were also labelled 1, 2, 3,4,5,6, and 7. 100ml of the test extracts at 300, 350, 400, 450 and 500 mg/l concentrations respectively was each transferred into separate laboratory plastic test beakers labelled as 2, 3, 4, 5 and 6 respectively. Water was used as control (Group 1) and the synthetic insecticides; (Rambo) was prepared and used as standard control (Group 7) at a rate of 500mg/l. All concentrations of the two extracts and the standard insecticide were evaluated for mosquito larvicidal activity. 30 of 3rd and 4rd instar larvae were placed in each beaker. Observations were made over 3, 6, and 24 hours after the test subject were introduced into distilled water to noticed recovery. A recovery time of 5-minutes was allowed. The larval mortality after 3, 6, and 24 hours was calculated and mean mortalities evaluated. The LC_{50} value were determined and larvae were counted as dead when they were not coming to the surface for respiration even after touching the beaker and were probe insensitive.

Statistical analysis

Data obtained was analyzed using R-console version 2.9.2. One way analysis of variance (ANOVA) was used to compare the mean mortality of *Anopheles* larvae across concentrations after each observed time of the larvicidal performance of the two plant species extracts. T-test was used to compare the mean mortality of *Anopheles* larvae between plants extracts after each time. Significant level was achieved if $P < 0.05$

RESULTS

Phytochemical screening of plant extracts

The result of phytochemical analysis of the *H. suaveolen*s, and *C. ambrosoides* investigated are summarized in Table1 below. The result shows that the plants contained chemical compounds such as alkaloids, tannins, saponins, flavonoids, carbohydrate, steroids, and cardiac glycosides. Both the two plant species extract had very high alkaloids and tannins content while anthraquinones was absent in both of extracts while steroids and cardiac glycosides were both absent in *C. ambrosoides*.

Mortality of *Anopheles* species larvae using *H. suaveolens* extract

The effects of the ethanol leaf extracts of *H. suaveolens* administered at the concentration of 0, 300, 350, 400,450, and 500 mg/l are as shown in figure 1. The percentage result indicated at the end of 24 hours of exposure showed that extract of plants had a toxic effect on the survival of *Anopheles* mosquito larvae. The mortality rate expressed in percentage was evident at all the levels of the test extract administered, though it was more pronounce and showed progression with the high concentration with time. A 100 % mortality of *An. gambiae* larvae was observed death after 6 and 24 hours respectively with the administration of 500mg/l of a standard instecticide (Rambo) in their breeding medium. In the control trials the larval mortality was 0 %. The mortality progressively increased with increase concentration of the extract with time. There was highly significant difference ($p<0.05$) in the mean mortality of *Anopheles* species larvae across concentrations after 3 hours, 6 hours and 24 hours exposure time respectively using *H. suaveolens* extract (after 3 hours: F_6 = 112.71, Adjusted R-squared = 0.971, P = 0.0001; after 6 hours: F_6 = 424.5, Adjusted R-squared = 0.9922, P = 0.0001; after 24 hours: F_6 = 988.93, Adjusted R-squared = 0.9966, P = 0.0001).

Mortality of *Anopheles* species larvae using *C. ambrosoides* extract

The mortality after 24 hours of exposure to *C. ambrosoides* showed that extract of plants had a toxic effect on the survival of *Anopheles* mosquito larvae. The percentage mortality was evident at all the levels of the test extract administered, though it was more pronounce and showed progression with the high concentration with time (Figure 2). A 100 % mortality of *An. gambiae* larvae was observed after 6 and 24 hours administration of 500mg/l of a standard insecticide (Rambo) while 0 % mortality was recorded in the control. The mortality rate of *Anopheles* larvae also progressively increased with increase concentration of the extract with time. There was highly significant difference ($p<0.05$) in the

mean mortality of *Anopheles* species larvae across concentrations after 3, 6 and 24 hours exposure time respectively using *C. ambrosoides* extract (after 3 hours: F_6 = 74.97, Adjusted R-squared = 0.9569, P = 0.0001; after 6 hours: F_6 = 381.4, Adjusted R-squared = 0.9913, P = 0.0001; after 24 hours: F_6 = 159.9, Adjusted R-squared = 0.9795, P = 0.0001).

Table1: phytochemical screening of *H. suaveolens* and *C. ambrosoides*.

Constituents	*H. suaveolens*	*C. ambrosoides*
Alkaloids	+++	+++
Tannins	+++	+++
Saponins	++	+
Flavonoids	++	+++
Carbohydrate	+	++
Steroids	+++	-
Anthraquinones	-	-
Cardiac glycosides	+++	-

Key: -Absent, +present, ++More present, +++Most present.

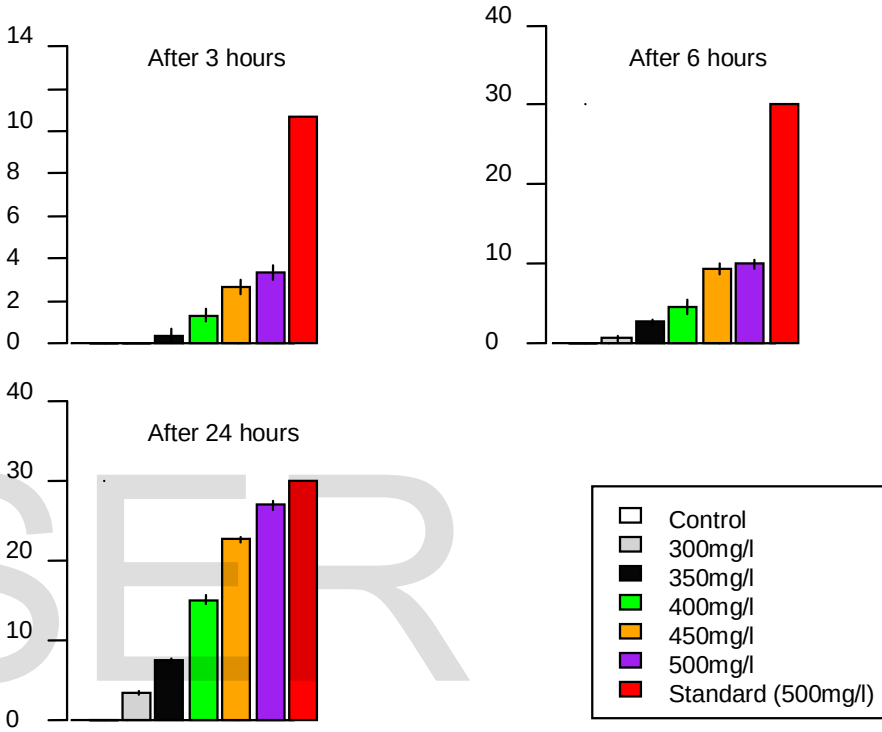

Figure1: Mortality of *Anopheles* species larvae exposure to *H. suaveolens* extract

Comparison of the mean mortality of *Anopheles* species larvae between plant species extracts: Comparison on mean mortality of *Anopheles* species larvae after each time between plant species extracts shows that there was no significant difference statistically ($p>0.05$) in the mean mortality of *Anopheles* species larvae between extracts of plant species

after 3, 6 and 24 hours exposure time respectively (Figure 3) (after 3 hours: t = 0.315, df = 28, P = 0.7551; after 6 hours: t = -0.6688, df = 28, P = 0.5091; after 24 hours: t = -1.4116, df = 28, p-value = 0.1691). After three hours of administration, *C. ambrosoides* leaves extract recorded high mortality of larvae than *H. suaveolens.* As time progresses, the reverse was the case at 6 hours and 24 hours where *H. suaveolens* administration resulted to high mortality *Anopheles* larvae than *C. ambrosoides.* This is confirmed by the LC_{50} of 354.81 and 389.04 mg/l after 24 hours of the administration of *H. suaveolens* and *C. ambrosoides* respectively (Figure 4).

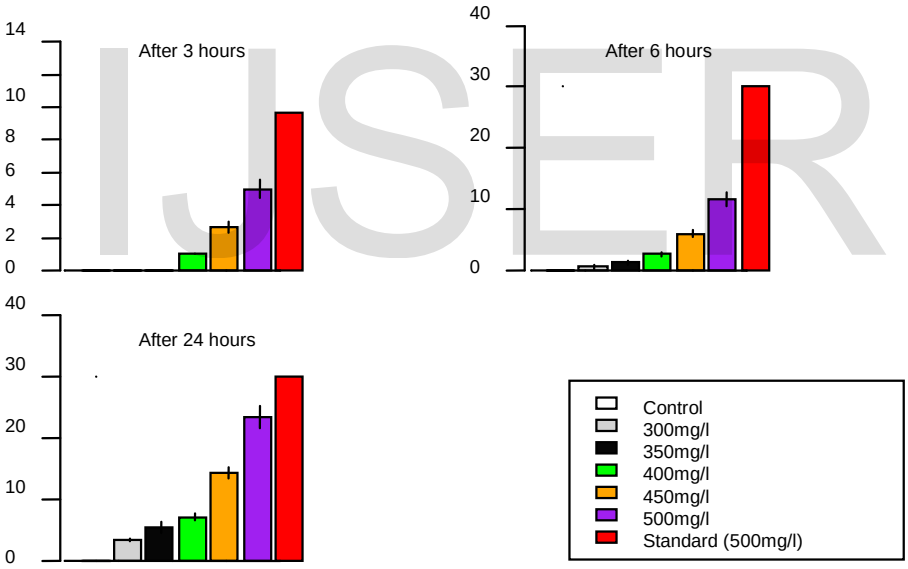

Figure 2: Mortality of *Anopheles* species larvae exposure to *C. ambrosoides* extract.

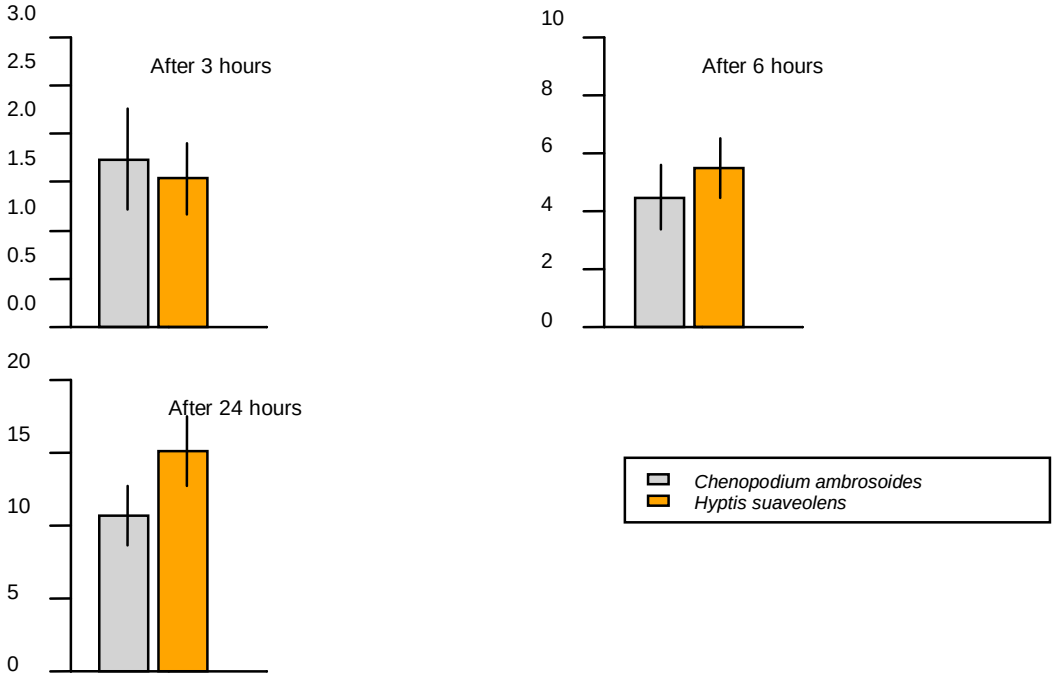

Figure 3: Mean mortality of *Anopheles* species larvae between plant species extracts

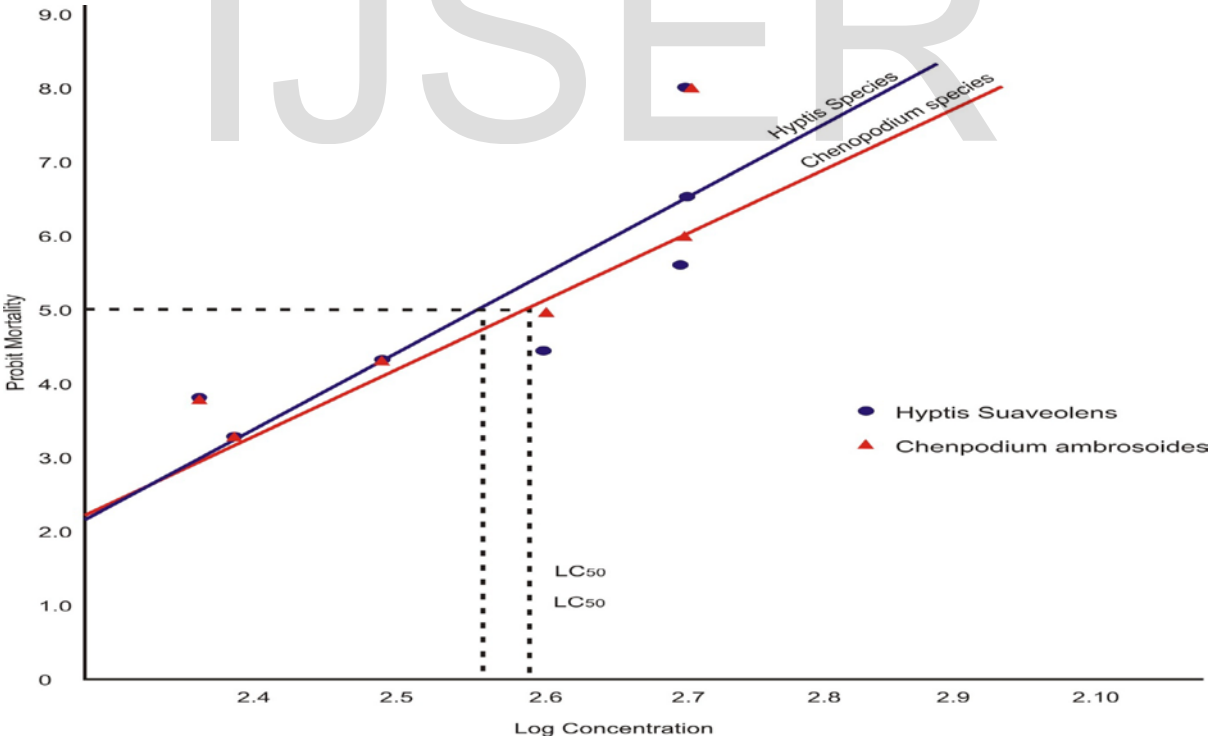

Figure 4: LC_{50} of *Anopheles* larvae after 24 hours of exposure to *H. suaveolens* and *C. ambrosoides*

(LC_{50}=354.81 mg/l and 389.04 mg/l respectively)

DISCUSSION

The presence of some bioactive components such as alkaloids, tannins, saponins, flavonoids and carbohydrate in both plants; steroids and cardiac glycosides in *H. suaveolens* while the absence of anthraquinones in both of the extracts is consistent [16] who in a study on the larvicidal activity of the methanolic leaf and stem/bark of *Jatropha curcas*, *Citrus grandis* and *Tinospora rumphii* against the dengue-vector, *Aedes aegypti* mosquito reported that the leaf and bark methanolic extracts of *Jatropha curcas* contains alkaloids, flavonoids and steroids while the leaf and bark/stem methanolic extracts of *Citrus grandis* and *Tinospora rumphii* are rich in alkaloids, saponins, tannins, flavonoids and steroids. These compounds are known to possess insecticidal and larvicidal activities of insects and other animals. It has been observed that insecticides of vegetable origin could be acutely toxic to various insects [17]. This toxicity has been linked with the potency of such extracts [18]. The phytochemical screenings of the ethanolic extract of *H. suaveolens* and *C. ambrosoides* leaves showed that the plants are rich in secondary metabolites bases which accounted for it toxicity. The presence of tannins in the plant may be the reasons why most animals do not graze on the plant and cardiac- glycoside is lethal to animals and insects when graze on the plant. Saponin is used in the manufacture of insecticides shampoos', and various drug preparation and synthesis of steroids hormone [19].

Active ingredient/ compounds may cause death by acting as neurotoxins or respiratory toxins by inhibiting the flow of nervous impulses resulting in the accumulation of the enzyme acetylcholinesterase at the post synaptic membrane and decrease in oxygen uptake ultimately resulting in death. The insects feed on these secondary metabolites potentially encountered toxic substances with relatively non-specific effects on a wide range of molecular target. These targets range from proteins enzymes, receptors, signalling, molecules, ion-channels and structural

proteins, nucleic acids, bio membranes, and other cellular compound [20]. This in turn, affects insects physiology in many different ways and at various receptor sites, the principal of which is abnormally in the nervous system (such as in neurotransmitter synthesis, storage, release, binding, and re-uptake, receptor activation and function, enzymes involved in signal transduction pathway). Rattan [20] reviewed the mechanism of action of plant secondary metabolites on insect body and documented several physiology disruptions such as inhibition of acetylcholinsterase by essential oils, GABA-gated chloride channel by thymol, sodium and potassium ion exchange disruption by pyrethrin and inhibition of cellular respiration by rotenone. The most important activity is the inhibition of acetylcholinerase activity (AchE) as it is a key enzyme responsible for terminating the nerve impulse transmission through synaptic pathway.

The leaf extracts of *H. suaveolens* and *C. ambrosoides* administered at different concentrations after 24 hours revealed that they have good degree of toxicity on Anopheles mosquito larvae. It was observed that development and growth processes such as pupation were inhibited as it only occurred in the control were no extract was added. These results are comparable to earlier results of [21] using *Artemisia annua* extract against *Culex* mosquito larvae, [22] using the Neem *aradirachta indica* extract against *Pipiens* larvae and [23] using aqueous extract of leaf of *Striga hermonthica* and *Mitracarpus scaber* against *Culex quinquefasciatus* larvae. There was no striking difference in the larvicidal effect of either plant extract on the *Anopheles* mosquito larvae. *H. suaveolens* extract at 500mg/l had mortality up to 90.00 % of *Anopheles* mosquito larvae while the same dose of *C. ambrosoides* extract recorded up to 84.44 % of the same species of mosquito larvae. However, noticeable and not surprisingly, the larval mortality was more pronounced with the use of the synthetic chemical insecticide (Rambo) which caused 100 % of Anopheles mosquito

larvae at 500mg/l. The high mortality reported in this study is in agreement with [24] who reported that the ethanolic extracts of *Acalypha gaumeri*, *Annona squamosa*, *Carlowrightia myriantha*, *Petiveria alliaceae* and *Trichilia arborea* (collected from different localities of the Yucatan peninsula, Mexico) at concentration of 10 mg mL-1 caused high mortality (95 to 100%) on *B. tabaci* eggs. Similarly, mortality caused by aqueous extracts of these plants ranged from 98 to 100% at concentration of 3% w/v. No significant differences on mortality within the same type of extract and the chemical insecticide imidacloprid were observed. The overall mortality of *An. arabiensis* in DEET sprayed huts (82%) was significantly higher than lambdacyhalothrin (76%, P = 0.043) and not statistically different to pirimiphos methyl (86%, P = 0.204). Mortality rates of *Cx. quinquefasciatus* in all sprayed huts were much lower than those recorded for *An. arabiensis*. However, mortality rates associated with all sprayed huts were significantly greater than the control (P < 0.001) [25]. Studies showed that larvae are more susceptible to insecticides than adults. Boussaada et al. [26] in a study carried out on sixteen aromatic plants extracts from three species belonging to the Asteraceae family: (*Mantisalca duriaei*, *Rhaponticum acaule*, and *Scorzonera undulata*,) obtained by using organic solvents of increasing polarity. They were tested for insect growth inhibition, contact toxicity and antifeedant activity against adults and larvae of confused flour beetle *Tribolium confusum* du Val (Coleoptera Tenebrionidae). For all extracts, mortality was higher for larvae than adults. It reached respectively 83%, 77% by using petroleum ether and methanol extracts of *R. acaule*, suggest that *M. duriaei* and *R. acaule* may be used in grain storage against insect pests. However, the results of this study is at variance with [27] recorded slightly low mortality (58 %) of *Tribolium castaneum* caused by the *Peganum harmala* extract at 100mg/ml during the 10 days after treatment. At the same time, mortality rates for the extracts from *Aristolochia baetica*, *A juga iva* and *Raphanus.*

raphanistrum (100mg/ml each) reached 34, 31 and 26% respectively.

The present study suggests that half of the population of the experimental *An. gambiae* larvae were killed using 400mg/l of *H. suaveolens* extract and *C. ambrosoides* extracts. Studies by [28] showed that oil from *Lantana camara* leaves produced repellence action against the bees, *Apis melifera* and the horsefly, *Tabanus* species. The mosquitocidal activity of *Lantana camara* has been demonstrated on the larval forms in the laboratory [29]. Studies by [30] showed that this plant produced 90-100% mortality of larvae as well as adults of *Aedes aegypti* and *Cx. quinquefasciatus*. The results of *Lantana* leaves extract also proved that they have larvicidal properties against *An. gambiae*. These finding agree with [31] who reported that leaf extract of *Lantana camara* showed larvicidal activity against *Cx. quinquefasciatus* and *Aedes allbopicus*. Another study by [32] showed the effect of the root barks of *Lantana viburnoides* species against late 3rd or 4th instar larvae of *Ano. gambiae*. They reported that extracts could serve as a source of larvicidal for managing various mosquito habitats in the field. Similarly, the presence of saponins, tannins, cardiac-glycosides and alkaloids in *H. suaveolens and C. ambrosoides* may serves as an indicator for the plant's mosquito larvicidal properties.

Today, environment safety is considered to be of paramount importance. An insecticide does not need to cause high mortality on target organisms in order to be acceptable but should be eco-friendly in nature. Chemical compounds from plant may serves as these are relatively safe, inexpensive and readily available in many parts of the world. Several plants are used in traditional medicines for the mosquito larvicidal activities in many parts of the world. According to [33], screening of locally available medicinal plants for mosquito control would generate local employment, reduce dependence on expensive and imported products and stimulate local effects to enhance the public health system.

The ethno-pharmacological approaches used in the search of new bioactive toxins from plants appear to be predictive compared to the random screening approach. The recently developed new isolation techniques and chemical characterization through different types of spectroscopy and chromatography together with new pharmacological testing have led to an interest in plants as source of new larvicidal compounds. Synergistic approaches such as application of mosquito predators with botanical blends and microbial pesticides will provide a better effect in reducing the vector population and the magnitude of epidemiology.

Bio-pesticides are found to be safe for usage without phototoxic properties and do not have a trailing side effects in the environment [34]. It is therefore recommended that *H. suaveolens* and *C. ambrosoides* be tried as a component in the synthesis of bio-pesticides of plant origin to be used against the developmental stage of the mosquito. Further investigation on the larvicidal properties of these traditional plants would be desirable as they may serve as cheaper alternative to the chemical properties and eco-friendly against non-target organisms.

REFERENCES

[1] Iwuala, M.O.E., and Ejezie, G. Control of Arthropod vector of Diseases in Nigeria; A review. *Bullectin of Animal Health and Production in Africa* 1978, Pp, 202-205.

[2] Anyanwu, G. I. and Iwuala, M.O.E. Mosquitoes breeding survey: range and distribution of species in peri-urban Jos area, central Nigeria. *Journal of Pest, Disease and Vector Management* 1995, **1(1)**: 39 – 45.

[3] Lehrer, S. "*Anopheles* mosquito transmissionof brain tumor". *Medical Hypotheses* 2010, **74**(1): 167-168.

[4] World Health Organization. Malaria. 2014*http://www.who.int/mediacentre/fact sheets/fs094/en/* , Accedssed 20/02/2014. At 01.43pm.

[5] Jimoh, A., Sofola, O., Petu,A., and Okorosobo T. Quantifying the economic burden of malaria in Nigeria using the willingness to pay approach. Cost Effectiveness and Resource Allocation 2007, **5(6)**. http://www.resource-allocation.com/ content/5/1/6. Accessed 03/04/2013 At 11.41 am.

[6] Haji, K.A., Thawer, N.G., Khatib, B.O., Mcha**,** J.H., Rashid, A., Ali**,** A.S., Jones, C., Bagi, J., Magesa, S.M., Ramsan**,** M.M., Garimo, I., Greer, G., Richard Reithinger, R. and Jeremiah M. Ngondi, J.M. Efficacy, persistence and vector susceptibility to pirimiphos-methyl (Actellic® 300CS) insecticide for indoor residual spraying in Zanzibar.2015, http://www.parasitesandvectors.com/cont

ent/8/1/628 Accessed 10/12/2015. At 12.47 pm.

[7] Yadouléton, A., Badirou, K., Agbanrin, R., Jöst, H., Roseline Attolou, R., Srinivasan, R., Gil Padonou, G. and Akogbéto, M. Insecticide resistance status in *Culex quinquefasciatus* in Benin. *Parasites & Vectors* 2015 http://dx.doi.org/10.1186/s13071-015-0638-3 Accessed 17/12/2015 At 01. 47pm.

[8] Aïzoun, N., Azondekon, R. and Akogbéto, M. Establishment of the correlations between resistance level to permethrin and DDT and knocked-down time in two *Anopheles gambiae* sensu lato populations from the Sudano Guinean area in the central part of Benin, West Africa. *International Journal Current Microbiology and Applied Sciences* 2014, **3(10)**: 878-884.

[9] Brown, A. W. Insecticide resistance in mosquitoes: a pragmatic review. *Journal of American for Mosquito control Association* 1986, **21(12)**: 23-40.

[10]. Russell T .L., Kay B .H., and Skilleter G .A. Environmental effects of mosquito insecticides on salt marsh invertebrate fauna. *Aquatic Biology* 2009, **6**: 77-90.

[11]. Shaalan, E .A .S., Canyonb D., Younesc, M.W. F., Abdelwahaba, H. and Mansoura, A .H. A review of botanical phytochemicals with mosquitocidal potential. *Environmental International* 2005, **3(11)**: 49-66.

[12] Harborne, J. B. and Baxter, H. Phytochemical Dictionary. A handbook of bioactive compounds from plants. Taylor and Francis Limited London Washinton D.C. London 1993, Pp. 324-689.

[13] Trease, G. E., and Evans, W. C. Pharmacognosy. English language Book society. Bailliere TindalLondon. 12th edition 1983,Pp 374-435, 715-726.

[14] Bhat, M. A. and Krishnamoorthy, K. Entomological investigation and distribution of *Aedes* mosquitoes in Tirunelveli, Tamil Nadu, India. *International Journal of Current Microbiology and Applied Sciences* 2014, **3(10)**: 253-260

[15] Tyagi, B.K., Munirathinam, A., Krishnamoorthy, R. and Venkatesh, A. A field based handbook on Identification keys to mosquitoes of public health importance in India. CRME Madurai 2012.

[16] Gutierrez, P. M., Antepuesto, A. N., Eugenio, B. A. L. and Santos, M. F. L. Larvicidal activity of selected plant extracts against the dengue vector *Aedes aegypti* mosquito**.** *International Research Journal of Biological Sciences* 2014, **3(4)**: 23-32.

[17] Bowers, W. S., Ohta, T., Clecra, J .S. and Marsella, P .A. Discovery of insect anti-juvenile hormones in plants: plants yield a potential fourth generation insecticide. *Science* 1976, **19(3)**: 542-547.

[18] Busvine, J. A. Insects and hygiene; The biology and control of insect pests of Medical and Domestic importance. 3rd Edn. Methuen and Co. Ltd. 1980, Pp.568.

[19] Ukwu, D. E. The potentials of *Ocimum gratissumum* ,*Penrgularia extensa,* and *Tetrapleura* as species and flawing agent. *Nigerian Agricultural Journal* 2003, **34**:143-144.

[20] Rattan R .S. Mechanism of action of insecticidal secondary metabolites of plants origin. *Crop protection* 2010, **29(9)**:13-20.

[21] Yohanna, J.A., Mafuyai, H. B. and Akinyombo, A. O. The larvicidal effects of leaf extract of *Artemisia annua* (sweet wood-worm) on *Culex* mosquito larvae:

Nigeria Journal of parasitology 2012, **33(2)**: 217-220.

[22] El-Bokl, M .M. Latent toxicity of azadirachtin treatment on *Culex pipiens* (Diptera: Culicidae). *Journal for Egyptician Academic Society for Environmental Development* 2003, **3**: 63-74.

[23] Abdullahi, K., Abubakar, M .G., Umar, R .A., Gwarzo, M. S., Mohammad, M. and Ibrahim, H .M. The larvicidal efficacy of aqueous extracts of *Striga hermonthica (delile) Benth* and *Mitracarpus scaber* (*Zucc*) on *Culex quinquefasciatus* larvae. *Nigerian journal of parasitology* 2011, **35(1)**: 105-108.

[24] Cruz-Estrada. A., Gamboa-Angulo. M., Rocío Borges-Argáez, R. and Ruiz-Sánchez, E. Insecticidal effects of plant extracts on immature whitefly *Bemisia tabaci* Genn. (Hemiptera: Aleyroideae). *Electronic Journal of Biotechnology* 2014, http://www.ejbiotechnology.info DOI: 10.2225/vol16-issue1-fulltext-6. Accessed 15/12/2015 at 11.40am

[25] Kitau, J., Oxborough, R., Matowo, J., Mosha, F., Magesa, S. M. and Rowland, M. Indoor residual spraying with microencapsulated DEET repellent (N, N-diethyl-m toluamide) for control of *Anopheles arabiensis* and *Culex quinquefasciatus. Parasites & Vectors* 2014, http://www.parasitesandvectors.com/content/7/1/446. Accessed 02/10/2014 at 1. 23pm

[26] Boussaada, O., Kamel, M. B. H., Ammar, S., Haouas, D., Mighri, Z. and Helal, A. N. Insecticidal activity of some Asteraceae plant extracts against *Tribolium confusum, Bulletin of Insectology* 2008, **61 (2)**: 283-289.

[27] Jbilou, R., Ennabili, A. and Sayah, F. Insecticidal activity of four medicinal plant extracts against *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae). African *Journal of Biotechnology* 2006, **5 (10)**: 936-940.

[28] Attri, B .S. and Singth, N. A note on the biological activity of the oils of *Lantana camara* L. *Indian Journal Entomology* 1978, **39**: 384-385.

[29] Yohanna, J. A., Mafuyai, H. B. and Akinyombo, A. O. The larvicidal effects of leaf extract of *Artemisia annua* (sweet wood-worm) on *Culex* mosquito larvae: *Nigeria Journal of parasitology* 2012, **33(2)**: 217-220.

[30] Anyanwu, G. I. And Uloko, J. I. Evaluation of insecticidal effects of *Lantana camara* (Verbanaceae) on mosquito adults and larvae. West *African Jornal of Pharmacol*. Drug Resource 1997, **13(2)**:23-26.

[31] Nath, D.R., Bhuyan, M., and Goswami S. Botanicals as mosquito larvicides. *Defence Science Journal* 2006, **56**: 507-511

[32] Innocent, E.C., Joseph, C., Gikonyo, N.K., Moshi, M.J., Nkunya M.H.H., and Hassanali, A Mosquito larvicidal constituents from *Lantana viburnoides* sp *viburnoides var kisi* (A.rich) verdc (verbenaceae). *Jorunal Vector Borne Disease* 2008, **45**: 240-244.

[33] Bowers, W. S., Sener, B., Evans, P. H., and Erdogan, I. Activity of Turkish medicinal plants against mosquitoes *Aedes aegypti* and *Anopheles gambiae. Insect Science* 1995, **16(3)**: 39-42.

[34] Meshnick, S. R. Artermisinin; Mechanisms of action resistance and toxicity. *International Journal of Parasitology* 2003, **32**: 1655-1660.

University of Jos, Faculty of Natural Sciences, Department of Zoology, P.M.B. 2084, Jos, Plateau State, Nigeria.
Correspondence: thonydawet@gmail.com