
Ibanga Ekong¹, *, Joel Akilah²

¹Department of Community Health, University of Uyo, Uyo, Nigeria
²Department of Public Health, Federal Ministry of Health, Abuja, Nigeria

Abstract

This report, a review and synthesis of six zonal reports, was aimed at examining trends, over an eight-month period, in identifying malaria vectors, their behavioural pattern and susceptibility status to insecticides across the six ecological zones in Nigeria. Review of project reports and Key Informant interviews revealed that monthly entomological surveys of adult mosquitoes using pyrethrum spray collection (PSC) and the CDC human-baited light trap stationed both indoors and outdoors overnight for three to four consecutive days each month generally from March to October 2014. Various relevant parameters, including global positioning system (GPS) locations of the sampling points were recorded in a total of randomly selected 32 houses per site. Samples were collected, identified morphologically and preserved for further laboratory analysis. Larval survey and insecticide resistance testing were conducted with the standard World Health Organization (WHO) procedures and susceptibility test kits using 2 to 3-day old adult Anopheles gambiae sensulacto reared from larval collections. The WHO susceptibility tests were accompanied with the CDC bottle assay tests and results analysed using World Health Organization Pesticides Evaluation Scheme (WHOPES) criteria. Results showed that Anopheles gambiae sensulacto was the dominant malaria vector of each monthly collection either in the PSC or CDC light trap collection indoors or outdoors. The mosquito indoor and outdoor activities were remarkable, showing possible malaria transmission either inside or outside the house. The Anopheles population was highly susceptible to bendiocarb and propoxur insecticides but resistant in variable degrees to primiphos-methyl, permethrin, alpha cypermethrin, deltamethrin, lambdacyhalothrin and DDT.

Keywords

Entomological, Surveillance, Malaria, Nigeria

1. Introduction

Malaria has remained a most important global health challenge, with Nigeria carrying the biggest burden, contributing 25% of the 219 million malaria cases and 19% of the 435,000 malaria deaths globally (WMR 2018) [1]. With the high endemicity of malaria in Nigeria, in addition to the thousands of lives lost annually, it also results in massive economic losses.

Through the coordination of the Global Malaria Programme, many other countries have progressed to pre-elimination, and even elimination, however, for some others, including Nigeria, the scourge of malaria remains a major, unresolved problem in tropical Africa. Vector control being a widely used malaria control strategy is based on the relationship between Anopheles mosquitoes, the arthropod vector and malaria transmission. For an effective malaria vector control which requires proper planning and implementation, there needs to be adequate entomological information as regards identification of major malaria vectors,

* Corresponding author
E-mail address: hangeky@yahoo.com (I. Ekong)
determining the intensity of transmission, inoculation rate and determination of insecticide resistance. Hence, the Federal Ministry of Health through the National Malaria Elimination Program (NMEP) in collaboration with the African Indoor Residual Spraying (AIRS) Nigeria project established malaria vector surveillance site in each of the six geopolitical zones in Nigeria. Kirkasamma LGA of Jigawa State, in the Sahel and Sudan Savanna, represents the North Western zone; Karbang Tumbi Village in Shendam LGA, Plateau State, in the Northern Guinea Savanna, represents the North Central Zone; Nasarawa Eggon and Doma LGAs of Nassarawa State in the Derived Savanna, also represents the North-Central zone; Amechi-Idodo in Nkanu East LGA, Enugu State in the Guinea Savanna, represents the South Western zone; and Oduoha- Emuoha in Emuoha LGA of Rivers State in the Forest which represents Mangrove Forest, represents the South Southern zone. Figures 1 and 2 show types of vegetations surrounding dwelling places.

The sentinel sites were established to: i) identify malaria vectors in the sentinel sites, ii) establish vector density, distribution and seasonality, iii) monitor vector feeding period in the sentinel sites and iv) determine the susceptibility of Anopheles species to the recommended four classes of insecticides for malaria control.

Taking into cognizance research on entomological parameters of malaria transmission, a sentinel site can be a designated place, locality or community where information relevant to transmission of malaria is collected systematically, over a period. Using standard protocols, baseline malariometric indices such as the identity of the malaria vector species, their distribution in time and space in the area, density in respect to human habitation, biting and resting behaviour, man – biting rate and the rate of malaria transmission is continuously generated [2]. Concurrently, data on the types of breeding sites of Anopheline vector species, their geographical location, productivity and ecological characteristics is recorded. Similarly, environmental factors and anthropogenic activities which influence malaria transmission are also recorded alongside the entomological parameters.

Site selection is often guided by high values of entomological indices for malaria transmission and corresponding cases of severe malaria.

The project sought to perform the following:

1. Adult mosquito sampling using pyrethrum spray collection (PSC) method to sample 32 structures per site per month;
2. Sampling two structures per site for 3 nights per month using human – baited CDC Light Trap method. Changing the cups hourly to evaluate behavioural biting and resting pattern of the Anopheline vectors both indoor and outdoor;
3. Morphological identification, processing and preservation of specimens of the Anopheline mosquitoes;
4. To conduct Larval Sampling monthly at the sentinel site; and
5. To conduct insecticide susceptibility test using the WHO Bioassay Test Kits and the CDC Bottle methods.

This report is aimed at synthesizing reports from six ecological zones with the aim to compare malaria transmission mechanics, thereby giving a wholistic picture of entomological surveillance for malaria in Nigeria.

2. Methodology

2.1. Study Area

2.1.1. Rivers State

The study was conducted in a malaria endemic community, Oduoha-Emuoha in Emuoha Local Government Area (LGA) of Rivers State. It is located in the Southern South region of Nigeria. It represents a rural ecotonal community with characteristics of both fresh and salt water environment. The community is inproximity with the new Calabar river which empties into the Bonny Estuaries.

The rural dwellers are traditional Ikwerre-speaking people of the Rivers State. The natives depend mainly on farming as their income. Other economic activities include fishing and trading. The community is located in the upland region of Rivers State and is characterized by tropical rain forest. The topography is flat with pockets of forest stream, which also serve as source of drinking water. The climate is characterized by two distinct seasons, the wet and dry seasons, the former taking place from April to October and dry season between November and March.

Their dwellings are mostly modern houses (cemented and partially cemented, painted and unpainted) with iron roofing sheets, only a few dwell in traditional houses (mud or wooden) with thatched or zinc roofs.

They live in clusters and are mostly Christians; a few are idol worshippers.

2.1.2. Plateau State

The study was carried out in the malaria endemic community of Karbang Tumbi Village, a large agrarian community in Shendam LGA of Plateau State, North Central Nigeria. It is surrounded by a swamp where different species of mosquitoes breed all year round. The community consists of 99% traditional houses (mud wall) and 1% modern houses (block wall).
2.1.3. Lagos State
Epe LGA is at the outskirts of Lagos metropolis in South Western Nigeria. The climate is characteristic of the forest zone. The rainy season is from April to October and dry season from November to March. The mean annual rainfall is 2000mm with a mean relative humidity of 78%. The mean temperature is 24°C during the wet and 30°C during the dry season. The study community, which is endemic for malaria, flanks the extension of the Lagos Lagoon along the Epe-Ijebu ode road (approximately 15km from Epe town) with an approximate population of 500 people. The people are mainly of the Yoruba ethnic group involved in trading and fishing. Housing structures consist of both traditional houses (15%: mud wall with thatched roof) and modern houses (>80%: brick houses with corrugated iron roof).

2.1.4. Jigawa State
Kirikasamma LGA, a malaria endemic community, is drained by Rivers Hadejia, Jamaare and Komadugu which form part of the Lake Chad basin system. The annual rainfall ranges about 400-480mm with a mean annual temperature of about 27°C but with temporal fluctuations. The vegetation consists of various types of heat-resistant tree variation predominantly of the Acacia species (Neem and Baobab trees) all scattered across wide expanse of grasslands. Sporadic shelterbelts to prevent the impending desert encroachment are also found. The main economic activities in which the people are engaged in include irrigation farming, fishing animal rearing and potash mining.

2.1.5. Enugu State
Amechi- Idodo is largely an agrarian community. The inhabitants produce rice, yam, cocoyam, cassava, oil palm, palm wine. Other occupations common in the area aside farming include: fishing, hunting and trading. Average educational status of the community is seemingly low.

2.1.6. Nasarawa State
Nasarawa Eggon and Doma LGAs are agrarian in nature. The state is made up of plain lands and hills.

Figure 2. Typical vegetation surrounding dwelling places [north] (credit to M.G.).

2.2. Study Strategy

2.2.1. Advocacy and Sensitization
Prior to the commencement of the studies, advocacy and sensitization visits were carried to the community gate keepers, notably the traditional rulers of the study community to seek support, ethical consent and cooperation from relevant stakeholders of the proposed malaria vector surveillance studies. Community members were also educated on the principle and objectives of the sentinel site and community spokesperson engaged in the house selection process for the PSC. Monthly collection was preceded with health education information led by the LGA Malaria Focal Persons. Though, due to cultural factors in the northern zones, house-wives were trained to collect mosquitoes by PSC at the household level. This innovation of using the house-wives in the project was found to be culturally compatible and acceptable to the people.

2.2.2. Reconnaissance Visit/Mapping
The malaria focal person at Local Government Area, and recruited locals guided the team to conduct geographical survey of the area and suitable sites for the studies were identified. This was followed by GPS mapping and numbering of the houses. The house owners of the Identified structures were instructed to be around and make their houses available for the study. They were also instructed not to apply any insecticides two days to the sampling date. Anopheles breeding sites were also identified (figure 3), and geo referenced with a GPS. Suitable sites and structures for CDC light trap were also mapped. A total of 32 houses were randomly selected per site. Information on types of household interventions used against mosquito were collected using structure pretested questionnaires from household heads in houses selected.
2.2.3. Monthly Entomological Survey

Monthly entomological surveys of adult mosquitoes were collected in a total of 32 houses randomly selected using Pyrethrum spray collection (PSC) together with human-baited CDC trap indoors and outdoors for four consecutive days from March to October, 2014.

i. Adult female collections-psc

PSC of Adult Anopheline mosquitoes was done using standard collection protocols recommended by the Federal Ministry of Health and World Health Organization [2]. The mosquitoes were collected using spray sheet (room collection or pyrethrum knock down collection). At each house selected for PSC, geo-coordinates were measured; a local ID associated with the area was recorded alphanumerically. Prior to the procedure, members of each household were requested to remove food items, extinguish fires and remain outside for a while. White sheets were then spread on the floor of all sprayable rooms while doors and windows were shut. The eve perimeter was sprayed from inside and outside with pyrethrum. The inside spray men also sprayed the inside room, under tables, chairs. After that the spray men exited the room and closed the door. After 10 minutes, sheets were examined outside for adult Anopheles mosquitoes which were picked carefully with entomological forceps, see figure 4. Magnifying hand lens was used to differentiate between female anopheles and other mosquitoes using morphological characteristics. The female anopheles were preserved individually in a well labeled Eppendorf plastic vial half filled with silica gel and stuffed with white papers. The preserved mosquitoes were later sent to the reference laboratory for Polymerase Chain Reaction (PCR) analysis.

ii. Cdc light trapping

CDC light trapping was carried out in two (2) randomly selected houses for 3 nights each month according to the standard methods to determine the malaria vector feeding time and location. The CDC human baited light trap was placed close to the legs of a person sleeping under an untreated bed net, as an attractant, in both locations (indoors and outdoors) with collecting bags removed hourly and replaced by an empty one to determine feeding time of the vectors., see figure 5. The light traps collections were carried out from 6pm to 6am for three nights. The mosquitoes caught in each of the structures per hour were put in labeled ventilated paper cups, counted and sorted into genera Culex and Anopheles. The genus Anopheles was further identified into An. gambiacesesulacto, An. funestus group and other minor species. Morphological characteristics including the number and patterns of bands on wings, palps and legs were used to separate the species using methods described by Gillies and Coetzee [3], and later preserved in Eppendorf tubes for further processing.

Man-Biting Rate (MBR) is expressed as the number of bites a person receives from a specific vector species per night. This parameter can be directly estimated from CDC light trap catches using the formula below:

\[
MBR = \frac{\text{number of mosquitoes collected}}{\text{number of Traps} \times \text{number of nights}}
\]
2.2.4. Larva Collection

Before the commencement of the collection, basic training was given to the local personnel to be able to:

1. Differentiate between mosquito larvae and other larvae
2. Differentiate between Anopheles larvae and other mosquito larvae
3. Use the correct technique when approaching the area to be sampled and the
4. Appropriate dipping technique for the habitat to be sampled amongst others.

Field larval collections, which was geo-referenced, were carried out around the sentinel site by searching different types of larval breeding sites, e.g. various types of permanent water bodies, small water pools, or impoundments (see figure 6) using a white iron dipper (ladle) for larvae collection. The harvested larvae were counted from each water body and transported to the insectary. Morphological identification was carried out in situ using morphological keys [3] and species sorted out accordingly.

i. Rearing of larvae

Mosquito larvae were reared to adult stage under ambient laboratory environmental condition. The female Anopheles larvae were sorted out and put in suitable containers containing water, and precautions taken to prevent crawling insects from invading the larvae. See figure 7. The female mosquito larvae were fed with appropriate meals.

![Figure 7. Rearing of Larvae](Photo credit to E.N.)

ii. Rearing of adult mosquitoes

Adult mosquitoes that emerged were put into wooden rearing cages and fed with glucose solution (figure 8). Anopheles female mosquitoes that were two to three days old were aspirated out and used for susceptibility test.

![Figure 8. Rearing of Adult Anopheles Mosquitoes](Photo credit E.N.)

2.2.5. Susceptibility Tests

Insecticide susceptibility tests were carried out using the standard WHO protocol (WHO, 1998), and CDC bottle Bio Assay.

i. WHO susceptibility test procedure

WHO Insecticide susceptibility test kits and impregnated paperswere used for this test. Two to three day old non blood-fed adult female anopheles mosquitoes collected around the sentinel site were tested. Mosquitoes from the
different breeding sites were pooled so that the mosquitoes tested were fully representative of the vector population in the sentinel area. Batches of 20-25 mosquitoes were exposed to test papers impregnated with insecticides as listed in table 1. Control experiments with the same batch (20-25) of mosquitoes from the site were carried out, in this case, the mosquitoes were exposed to untreated papers impregnated with mineral oils. Experimental set-up was placed on a platform surrounded with water to prevent crawling insects from eating up the mosquitoes. The knockdown effect of each insecticide was recorded every 10 minutes over the one hour exposure period. After the exposure, mosquitoes were then transferred to a recovery tube and provided with 10% glucose solution. Final mortality was recorded 24 hours post-exposure. If the mortality in the control group was over 5%, but less than 20%, correction of the mortality was made by applying the Abbot formula. When mortality in the control was over 20%, the tests were discarded.

The mosquitoes used for the tests were preserved individually in Eppendorff tubes, labelled appropriately for identification and further analysis (see figure 9).

Table 1. Names and classes of the 8 insecticides tested for susceptibility of Anopheles spp. Mosquitoes.

<table>
<thead>
<tr>
<th>S/No</th>
<th>Class of Insecticide</th>
<th>Name of Insecticide</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Pyrethroid insecticides</td>
<td>alpha-cypermethrin (0.4%), deltamethrin (0.05%), lambda-cyhalothrin (0.05%) and permethrin (0.75%)</td>
</tr>
<tr>
<td>2</td>
<td>Organophosphate insecticide</td>
<td>preminiophos-methyl (0.25%)</td>
</tr>
<tr>
<td>3</td>
<td>Carbamate insecticides</td>
<td>propoxur (0.05%) and Bendiocarb (0.13%)</td>
</tr>
<tr>
<td>4</td>
<td>Organochlorine insecticide</td>
<td>dichloro diphenyl trichloroethane (DDT) 4%</td>
</tr>
</tbody>
</table>

Figure 9. WHO Susceptibility Test Demonstration (Credit to E.N.).

Figure 10. Experimental Set-up of CDC Bio-Assay Bottles (credit to E.N.).

ii. CDC bottle bioassay

The CDC bioassay measures the rate of kill of members of a population at a given dose of insecticide. Mosquitoes which survive after the exposure period are assumed to be resistant.

The CDC test bottles were rinsed with detergents and air dried for 2 hours to achieve complete dryness. The bottles were lined up with their lids open. See figure 10. A batch of 20-25 female anopheles mosquitoes (1-3 days old) were collected with an aspirator from the rearing cages and introduced into each of the five bottles. Of the five bottles, four were coated with appropriate insecticide while one served as control. The bottles were labelled with the name and concentration (µg/bottle) of the insecticide to be coated. The date of the experiment was also labeled on the lid and the bottle.

Serial dilutions of each of the insecticides (Permethrin, Lambda-cyhalothrin, Alpha-cypermethrin, Deltamethrin, Bendiocarb, Premiphos-Methyl and DDT) were prepared by adding 1ml of the stock solution (49ml acetone + 1ml insecticide) to the treatment bottles. The control bottle contained 1ml of acetone only. Upon adding the solution, the bottles were swirled gently to ensure coating of inside surfaces. This process was continued long enough to allow the solvent to evaporate completely. The bottles were left open without their lids on in an undisturbed clean mat overnight to ensure that they are well dried. They were then covered with a cloth to protect them from the light.

2.2.6. Statistical Analysis

i. Rivers state

Microsoft excel statistical package was used to summarize the data. Minitop software was used for data analysis. The statistical variations between the mean number mosquitoes sampled in the 32 structures using PSC across the months were determined using one-way analysis of variance (ANOVA). Test was used to separate the means. Comparison between the indoor and outdoor behaviours was carried out using two ways ANOVA. TURKEY method was also used to separate the means: Sigma plot was used to draw the graphs of the variation of the number of mosquitoes sampled indoors and outdoors across the month and sampling hours. The same methods were applicable for the analysis of variations in the
temperature and relative humidity over the periods. Correlation between the density of mosquitoes sampled indoor and outdoor with the mean temperature and relative humidity over the study period was analyzed using regression test.

**ii. Plateau state**

Data obtained was analyzed using R Console version 2.9.2. Two sample t-test was used to compare the mean number of Anopheles species caught between points for each month.

**iii. Lagos state**

The data were entered into a Microsoft Access database and subsequently analyzed using SPSS. 12.1, STATA.

**iv. Jigawa state**

Statistical analytic method not stated.

**v. Enugu state**

Statistical analytic method not stated.

### 3. Results

Across the six entomological sentinel sites being reviewed, there was a uniform predominance of the *Anopheles gambiae s.l.* vector, the highest percentage prevalence being >93% in Lagos, and the lowest, 53.9% in Enugu State. The peak vector density varied across the states, with some having twin peaks (Rivers: May and October); (Enugu: March and October); (Nasarawa: May and July) and Lagos (April and September). Plateau’s peak was in July and Jigawa in August. Table 2 below shows the results.

**Table 2. Trend of Vector Density Across the Sites.**

<table>
<thead>
<tr>
<th>CRITERIA</th>
<th>RIVERS</th>
<th>PLATEAU</th>
<th>LAGOS</th>
<th>JIGAWA</th>
<th>ENUGU</th>
<th>NASARAWA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prevalent Specie</td>
<td>An. gambiae s.l. 92.75%</td>
<td>An. gambiae s.l. (&gt;93%)</td>
<td>An. gambiae s.l. 86.2%</td>
<td>An. gambiae s.l. 53.9%</td>
<td>An. gambiae s.l. 68%</td>
<td></td>
</tr>
<tr>
<td>Peak Vector Density</td>
<td>May &amp; October</td>
<td>July</td>
<td>April &amp; September</td>
<td>August</td>
<td>March &amp; October</td>
<td>May and July</td>
</tr>
<tr>
<td>Lowest Vector Density</td>
<td>March</td>
<td>March</td>
<td>July</td>
<td>June</td>
<td>May</td>
<td>October</td>
</tr>
</tbody>
</table>

Table 3 below indicates the trend of vector behaviour and feeding time of the *Anopheles spp.* Jigawa, Enugu and Plateau States had similar peak periods for indoor and outdoor activities, while Lagos and Rivers States had different peak periods. Interestingly, Nassarawa had a wide range of both indoor and outdoor biting activity, about 9 hours of active biting.

**Table 3. Trend of Vector Behaviour and Feeding Time.**

<table>
<thead>
<tr>
<th>Criteria</th>
<th>Rivers</th>
<th>Plateau</th>
<th>Lagos</th>
<th>Jigawa</th>
<th>Enugu</th>
<th>Nassarawa</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indoor Peak Activity</td>
<td>3-5am</td>
<td>7-8pm; 11pm-12midnight</td>
<td>12midnight-1am, 1-2am and 4-5am</td>
<td>3am-5am; 5am-7am</td>
<td>3-4am; 5am-6am</td>
<td>7pm-8am</td>
</tr>
<tr>
<td>Outdoor Peak Activity</td>
<td>12midnight-1am, 1-2am and 4-5am</td>
<td>12midnight-1am, 1-2am and 4-5am</td>
<td>1-2am and 4-5am</td>
<td>1am-2am and 4-5am</td>
<td>1am-2am and 4-5am</td>
<td>7pm-8am</td>
</tr>
</tbody>
</table>

Trends on the relationship between Environmental Parameters and Vector Abundance (VA) were mainly non-significant, hence they were not captured in this narrative.

Table 4 and figure 11 below represent the susceptibility of the vector to a range of insecticides as measured by their mortality in the first hour, except for Plateau which was measured in 24 hours. Plateau was not captured in the bar chart due to non-comparability.

Overall, DDT, an organochloride, had the least sensitivity, recording as low as 17% vector mortality in Lagos State. Conversely, the carbamate class of insecticides had the highest sensitivity, recording 98.88 to 100% vector mortality. The pyrethroids show various degrees of resistance, recording 27.5% to 100% vector mortality.

**Table 4. Trend of percentage mortality due to effect of insecticides.**

<table>
<thead>
<tr>
<th>Insecticide</th>
<th>Class of insecticide</th>
<th>Rivers (% in 1 hr)</th>
<th>Plateau (% in 24hr)</th>
<th>Lagos (% in 1 hour)</th>
<th>Jigawa (% in 1 hour)</th>
<th>Enugu (% in 1 hour)</th>
<th>Nasarawa (% in 1 hour)</th>
</tr>
</thead>
<tbody>
<tr>
<td>DDT (40%)</td>
<td>Organochloride</td>
<td>78</td>
<td>63.16</td>
<td>17</td>
<td>41</td>
<td>31.25</td>
<td>27.5</td>
</tr>
<tr>
<td>Permethrin (0.75%)</td>
<td>Pyrethroid</td>
<td>42</td>
<td>94.57</td>
<td>64</td>
<td>50</td>
<td>71.25</td>
<td>27.5</td>
</tr>
<tr>
<td>Propoxur (0.05%)</td>
<td>Carbamate</td>
<td>100</td>
<td>98.88</td>
<td>100</td>
<td>99</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Bentiocarb (0.13%)</td>
<td>Carbamate</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Lambda-cyhalothrin (0.5%)</td>
<td>Pyrethroid</td>
<td>75</td>
<td>100</td>
<td>90</td>
<td>86.9</td>
<td>70</td>
<td>98.75</td>
</tr>
<tr>
<td>Deltamethrin (0.05%)</td>
<td>Pyrethroid</td>
<td>56</td>
<td>84.21</td>
<td>69</td>
<td>62.8</td>
<td>82.69</td>
<td>32.5</td>
</tr>
<tr>
<td>Primiphos-methyl (0.25%)</td>
<td>Organophosphate</td>
<td>100</td>
<td>71.43</td>
<td>98</td>
<td>61.8</td>
<td>12.5</td>
<td>46.25</td>
</tr>
</tbody>
</table>

Legend: >98% Mortality = Susceptible; 80-98% Mortality = Verification Required; and <80% Mortality = Resistant Individuals Present.

1 Where <95% Mortality occurs at the diagnostic time in bioassays that have been conducted under optimum conditions and with a sample size of >100 mosquitoes, then resistance can be strongly suspected.
4. Discussion

Synthesis of results from this study revealed that *Anopheles gambiae sensulacto* (s.l), as determined by Pyrethrum Spray Catches, is the main malaria vector across the geographical zones. This correlates with other studies done across the country [4-7]. They are an efficient vector in Nigeria due to favourable climatic conditions characteristics of the zone and abundance of breeding habitats.

In agreement with previous studies, vector density of the *Anopheles* species were higher at the onsets of the rains in Nigeria as attested to by the twin peak periods observed in some of the sites [8, 9]. However, there were variations across the zones with peaks spreading across the rainy season and ranging from April to October. However, the lowest vector densities were recorded during the dry season in March, and rainy season in June, July and October. This could account for the characteristic all-round endemicity in Nigeria. It is worthy of note that rainfall provides breeding sites for mosquitoes to lay their eggs and ensures a suitable relative humidity.

The vector behaviour demonstrated by the CDC-Light Trap Catches indicate the tendency of the Anopheline malaria vectors to feed in the late night, but some feed early as shown by a wide range of variation, with Jigawa, Enugu, Nasarawa and Plateau sites showing similar bite times indoors and outdoors. However, Rivers and Lagos recorded different bite times indoors and outdoors. Though the endophilic (indoor biters) vectors showed peak activity after mid-night, the exophilic (outdoor biters) vectors’ activity was spread all through the night, from 7pm. A Burkina Faso study showed that *Anopheles gambiae* s.l. had a high biting activity that was high throughout the night, at indoor and outdoor posts alike [10]. The implication for this is the risk of transmission of the malaria parasite outdoors before one sleeps inside the Long-Lasting Insecticidal Nets. It also calls to question, heavy reliance on LLINs as a major control intervention, without a corresponding behavioural change among the populace. In Nigeria, families majorly tend to spend some time outside the house before retiring to bed due to a host of factors ranging from socialization to climatic convenience.

Susceptibility to insecticides showed an interesting perspective. The vectors are increasingly developing resistance to a wide range of insecticides, except those in the class of the carbamates (propoxur and bendiocarb) which have not been used for control programmes in Nigeria. The worst resistance was recorded with DDT; with the pyrethroids, as a class, and the different insecticides, showing a wide range of resistance across the zones in Nigeria [11, 12] and in Africa [13-15]. Interestingly, only the pyrethroids have been approved for impregnation of LLINs for the region, though there is currently a concerted effort by the National Malaria Elimination programme to introduce piperonyl butoxide (PBO)-based LLINs to address the growing resistance to pyrethroids in Nigeria. PBO is often combined with natural pyrethrins or man-made pyrethroids and has demonstrated effectiveness in certain regions of the world.
[16-19]. How effective this will be in malaria control programme, perceiving the fact that a component, the pyrethroid, is already resistant remains to be monitored, and vigorously, at that. This is a call for a definitive review of insecticides for use in malaria control in Nigeria and neighbouring regions if we aim to achieve the target 3.3 of the Sustainable Development Goal, “By 2030, end the epidemics of AIDS, tuberculosis, malaria and neglected tropical diseases and combat hepatitis, water-borne diseases and other communicable diseases [20].”

5. Conclusion

From the foregoing, it has been demonstrated that *An. Gambiae s.l.* is the predominant vector of malaria in Nigeria. The vector has demonstrated considerable activity indoors and outdoors, and across the rainy season of Nigeria. The mosquitoes were more active in the night, into early mornings. High susceptibility of the vector *An. gambiae s.l* is to the carbamates: bendiocarb and propoxur; with degrees of resistance to the pyrethroids, organochlorines and organophosphates.

Recommendations

1. Based on the present study, it is suggested that the area under the influence of *An. gambiae s.l* should be covered under a carbamate-based IRS programme.

2. There is a need to consider implementing the larval source management strategy.

3. The current pyrethroid-based LLIN ought to be reviewed.

Acknowledgements

This is to acknowledge the management of National Malaria Elimination Programme for granting access to data for this study; and to the following: Ebere, N., Mwansat, G., Awolola, S., Manu, A., Chukwuekezie, C. and Yako, A. for providing technical assistance in the field and laboratory towards the completion of the various studies.

Availability of Data

The Sentinel Surveillance data used to support the findings of this study have been deposited at the National Malaria Elimination Programme, Abuja.

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