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Research Article

Habitat Ecology and Propoxur Susceptibility of Anopheles Mosquito Larvae in Lafia Metropolis, Nasarawa State, North-Central Nigeria

James Ishaku Maikenti[1]*, Akwashiki Ombugadu[1], Sumayyat Umar Muhammad[1], Fatimah Abubakar[1], Hussein Oshomah Ahmed[1], Samaila Kuna Yusuf2, Scholastica Onyaweyo Ayuba[1], Mohammed Ahmed Ashigar[1], Sunday Kajibia Dogo[1] and Victoria Adamu Pam[1]

[1]Faculty of Life Sciences, Department of Zoology, Federal University of Lafia, Nigeria

[2]National Biotechnology Research and Development Agency (NBRDA), Umaru Musa Yar'adua Way Lugbe, Nigeria

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***Corresponding author:** James Ishaku Maikenti, Faculty of Life Sciences, Department of Zoology, Federal University of Lafia, Nasarawa State, Nigeria

ABSTRACT

The species of mosquitoes of the genus Anopheles are primarily responsible for spreading malaria, a disease that causes high morbidity and mortality in Nigeria. With the evolution of insecticide resistance among the malaria vector mosquito population, control measures for the vector become less effective. Therefore, this study sought to determine the habitat ecology of the larvae of Anopheles mosquitoes in Lafia metropolis of Nasarawa State, Nigeria, as well as the susceptibility profile of the emerged adults to Propoxur (carbamate insecticide). Female Anopheles mosquito larvae were collected from three communities of Lafia metropolis, namely, Gandu, Agyaragu-Tofa Road, Mararraba, using the standardized dipper. Four different habitat types; tyre tracks, marshy ground, hoof prints and river banks, were used for larvae collection. Parameters such as water temperature, dissolved oxygen, pH, turbidity and alkalinity of water were measured. Larvae were grown into adults under insectary conditions. CDC bottle bioassay test was done using Propoxur at the diagnostic dose to assess knockdown rate and mortality in 2 to 5 days old, sugar-fed females. Recording of mosquito mortality was done every 15 minutes within the 30 minutes diagnostic time. Breeding index was computed to calculate the risk of malaria transmission by the mosquitoes. In total, 1,973 Anopheles mosquito larvae were collected. The majority of the larvae, 1,637 (82.97%) were collected from tyres tracks while only two larvae (0.10%) from river bank. A significant difference ($\chi^2 = 181.12$, df = 3, $p < 0.001$) was found between the number of larvae collected across habitat types. The number of larvae collected per community showed that Gandu community had significantly more larvae, 1,706 (86.47%). Breeding index value of 16.44 was above 5% threshold indicating a higher transmission risk. The results from physicochemical parameter analysis showed strong inverse correlation between number of larvae collected per type of habitat (turbidity: r = -0.140, pH: r = -0.995 and dissolved oxygen: r = -0.789). However, weak positive correlations were observed between the number of larvae collected per type of habitat for temperature (r = 0.213) and alkalinity (r = 0.11). A 100% mortality and knockdown at 30 minutes occurred, showing complete susceptibility of Anopheles mosquitoes to Propoxur.

Keywords: Anopheles, Physicochemical parameters, Propoxur, Insecticide susceptibility, CDC Bottle Bioassay, Malaria vector control, Lafia, Nigeria

1. Introduction

Indeed, malaria continues to be one of the biggest public health problems in the sub-Saharan region of Africa and accounts for about 92% of malaria infections and 93% of deaths from the disease around the globe[1]. The Plasmodium parasites are responsible for this mosquito-borne disease with a great toll among young children under five years old and pregnant mothers[2]. It is evident that despite efforts over many years, the global malaria burden has never been so high and this indicates the need to take another approach to managing the disease. Indeed, mosquito prevention is part and parcel of malaria control programs and LLINs and IRS are the primary methods of vector control[3]. However, their success is currently endangered by increasing insecticide resistance among the vectors.

According to WHO, insecticide resistance has developed to at least one insecticide class in 78 countries with active malaria transmission[3]. Pyrethroid insecticides are the only group used on LLINS and there is an increased resistance to these chemicals. Propoxur is a carbamate pesticide that inhibits acetylcholinesterase and, therefore, prevents neurotransmission, making it important in rotation programs[4]. Nigeria is a country that carries the greatest malaria burden in the world, with almost 27% of worldwide infections recorded in this country[2]. Despite the fact that Nasarawa state experiences perennial malaria transmission, there is inadequate information on the susceptibility of the local Anopheles population to Propoxur. This project intends to address this gap by investigating the susceptibility profile of emerged adult Anopheles mosquitoes cultured from wild field larvae in Lafia metropolis of Nasarawa State, Nigeria, to Propoxur.

2. Materials and Methods

2.1. Study area

The experiment was conducted in the city of Lafia (8°28′59.99″N; 8°31′0.01″E), which is the capital of the state of Nasarawa, in Nigeria **(Figure 1)**. Lafia experiences a savanna climatic zone and it consists of two seasons, namely wet season (from April to October) and dry season (November to March). Average rainfall in Lafia varies between 1,100 mm and 1,500 mm, while the temperature.

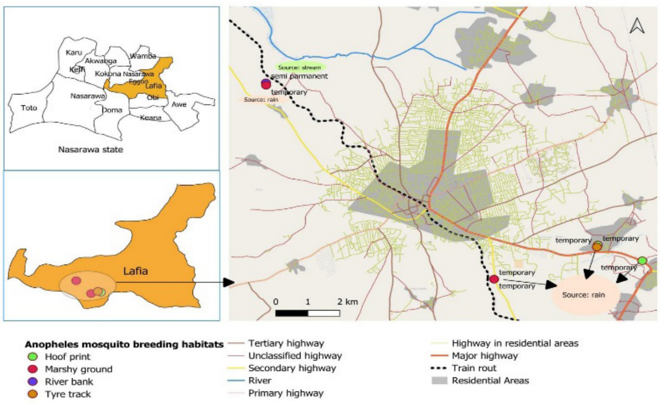

Figure 1: Anopheles Larvae Collection Sites in Lafia Local Government Area, Nasarawa State, Nigeria.

2.2. Larval collection

Mosquito larvae were collected between 7:00 and 9:00 AM through the use of a dipper of 350 mL as recommended by Williams and Pinto[5]. A pipette of 5 mL was used where there was shallow water. Four habitat categories were used for collecting larvae namely: tyre tracks, marshy ground, hoof prints and river banks. The collected larvae were then placed in the respective labelled bottles filled with the water from the habitat and brought to the Zoology laboratory of Federal University of Lafia.

2.3. Rearing to adulthood

The larvae were reared in breeding bowls containing their own breeding water for 24 hours after which they were transferred to deionized water. The larvae were given food consisting of yeast and biscuits flakes on daily basis. Room temperature was maintained during the process of rearing and the emerging adults were provided with 10 percent sugar solution.

2.4. Adult mosquito identification

Adult female mosquitoes were morphologically identified using standard taxonomic keys[6]. Only Anopheles species were included.

2.4.1. CDC bottle bioassay for propoxur susceptibility: CDC bottle bioassay method was used following the guidelines as specified by the CDC[7]. For the test, Wheaton bottles of 250 ml capacity were coated with 1 ml of Propoxur insecticide solution using the diagnostic dosage (12.5 µg/bottle), while for the control bottle, 1 ml of acetone solution was coated. In the test, 25 two to five-day-old unfed female Anopheles mosquitoes each were placed inside four Propoxur coated bottles and one control bottle. Observation for mosquito mortality was done after intervals of 15 minutes within the 30 minutes diagnostic time. Mosquitoes were considered dead if they failed to stand and coordinate movements.

Resistance classification[8]:

- Mortality 98–100%: susceptible
- Mortality 90–97%: suspected resistance (needs confirmation)
- Mortality <90%: resistant

2.4.2. Physicochemical parameters: At each larval collection site, water temperature (°C), pH, turbidity (NTU), alkalinity (mg/L) and dissolved oxygen (mg/L) were measured in situ using a handheld multi-meter.

Entomological Index – Breeding Index (BI)

The Breeding Index was calculated as:

$$BI = \frac{TLP}{ND \times BP}$$

Where:

TLP = total number of larvae and pupae collected

ND = total number of dips performed

BP = Number of breeding sites sampled

Other parameters are available to estimate the occupation of breeding sites by mosquitoes

A BI >5% was considered indicative of high transmission risk[5].

2.5. Data analysis

Analysis of data obtained from the experiment involved the use of R program version 4.3. Analysis involved the use of Chi-square test to compare larval abundance in different community structures and habitats. Analysis also included calculation of Pearson's correlation between larval abundance and physicochemical properties. Significance level was set at $P < 0.05$.

3. Results

3.1. Larval abundance by community

A total of 1,973 Anopheles mosquito larvae were collected in different breeding sites within the study location. The highest abundance was in Gandu community (1,706; 86.47%), followed by Agyaragu Tofa Road (180; 9.12%) and Mararraba (87; 4.41%) as shown in **(Table 1)**. The difference was statistically significant ($\chi^2 = 127.39$, df = 2, $P < 0.001$).

Table 1: Abundance of Anopheles **Larvae in Relation to Communities within** Lafia Metropolis.

Community	Number of Larvae	Abundance (%)
Gandu	1,706	86.47
Agyaragu-Tofa Road	180	9.12
Mararraba	87	4.41
Total	1,973	100

$\chi^2 = 127.39$, df = 2, $P < 0.001$

3.2. Larval abundance by habitat type

(Table 2) shows that tyre tracks yielded the highest number of larvae 1,637 (82.97%), followed by marshy ground 178 (9.02%), hoof prints 156 (7.91%) and river bank 2 (0.10%). The difference was significant ($\chi^2 = 181.12$, df = 3, $p < 0.001$).

Table 2: Abundance of Anopheles Larvae in Relation to Habitat Type.

	Communities				
Habitat Type	**Gandu**	**Mararraba**	**Agyaragu-Tofa Road**	**Total**	**%**
Tyre track	1,637	0	0	1,637	82.97
Marshy ground	0	0	178	178	9.02
Hoof print	69	87	0	156	7.91
River bank	0	0	2	2	0.1
Total	1,706	87	180	1,973	100

$\chi^2 = 181.12$, df = 3, $p < 0.001$.

3.3. Sex ratio of emerged adults

Among emerged adults, females (177; 87.19%) significantly ($\chi^2 = 55.324$, df = 1, $p < 0.001$) outnumbered males (26; 12.81%).

3.4. Physicochemical parameters and larval abundance

Correlation analysis showed weak positive correlations between larval abundance and temperature ($r = 0.213$) and alkalinity ($r = 0.11$). But negative correlations were observed with dissolved oxygen ($r = -0.789$) and pH ($r = -0.995$). Also, turbidity showed a weak negative correlation ($r = -0.140$) as shown in **(Figure 2)**.

3.5. Breeding index

The Breeding Index was calculated as:

$$BI = \frac{TLP}{ND \times BP}$$

$$BI = \frac{1973}{10 \times 12}$$

BI = 16.44

This value exceeds the 5% threshold, indicating high potential for mosquitoborne disease transmission.

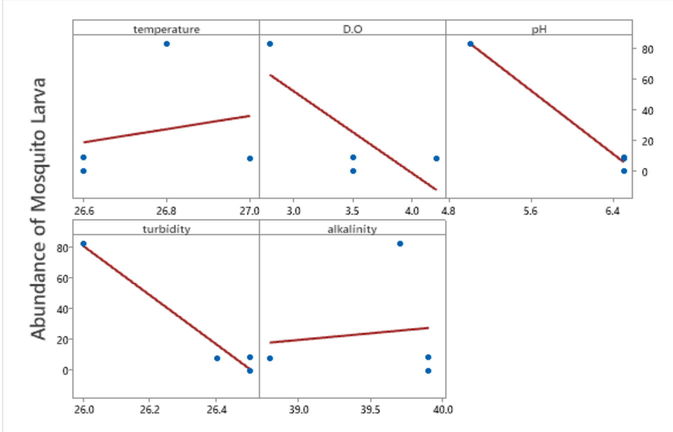

Figure 2: Scatter plots showing relationships between larval abundance and physicochemical parameters.

3.6. Ropoxur susceptibility

CDC bottle bioassays showed progressive knockdown: 0% at 0 minutes, 97% at 15 minutes and 100% at 30 minutes. All mosquitoes (100%) remained dead at the 24hour mortality check. Control bottles had 0% mortality. According to WHO criteria, mortality ≥98% indicates full susceptibility. Therefore, the Anopheles population in Lafia is fully susceptible to Propoxur **(Table 3)**.

Table 3: Susceptibility of female Anopheles **mosquitoes to** Propoxur.

Exposure time (min)	Number assayed	Knockdown (%)	Status
0	100	0	–
15	100	97	–
30	100	100	Susceptible
Control (acetone)	25	0	–

4. Discussion

The current study provided baseline information on larval ecology and Propoxur susceptibility status of Anopheles in Lafia metropolis, Nasarawa State. The prevalence of high larval densities in tyre tracks (82.97%) is consistent with the previous reports on urban areas that tyre tracks retain rainwater which create an optimal habitat for mosquito larvae[9,10]. The presence of tyre tracks as breeding habitats is indicative of poor waste disposal and represents a potential site for intervention through larval source management (LSM) in Lafia metropolis, Nasarawa State. The significantly high abundance of mosquito larvae in Gandu community (86.47%) in comparison to other locations could be attributed to the increase in anthropogenic activities such as open water containers, construction as a result of increasing population density and poor drainage[11]. The high Breeding index (BI) value (16.44) indicates a high risk of malaria transmission; the same has been documented by Igba, et al.[12] in Nasarawa State. The results of physicochemical properties of water showed that larvae abundance was inversely associated with high pH and organic rich condition, a phenomenon well established in previous studies[9,13]. On the other hand, positive correlation with temperature is predictable owing to faster development in warm waters.

The results of this research indicate that there is no sign of resistance to Propoxur in Anopheles mosquitoes in Lafia Nasarawa state, as evidenced by complete susceptibility to this insecticide (100% mortality in 24 hours). This is consistent with previous findings reported by Hassan, et al.[14] in Keffi, Nasarawa State, who reported the high efficacy of Propoxur on female Anopheles mosquito. Although resistance to pyrethroid class has been extensively reported throughout Nigeria[15,16], continued susceptibility to Propoxur is encouraging as it offers an alternative insecticide for use in IRS programmes to help delay evolution of pyrethroid resistance. Even though susceptibility does not necessarily translate into effective use in vector control operations, several other factors come into play. Nonetheless, Propoxur can be considered for IRS programmes in Lafia metropolis, Nasawara State, particularly as part of insecticide rotation. No resistant individuals were detected during testing, but continued monitoring is essential as IRS programmes generate selection pressure for resistance. Anopheles mosquitoes in Lafia metropolis remained susceptible to Propoxur insecticide. This insecticide can be considered as one of the options for use in IRS in addition to others as an insecticide rotation strategy for combating pyrethroid resistance.

5. Conclusion

This study confirms that tyre tracks are the dominant breeding habitats for *Anopheles* larvae in Lafia, with Gandu community exhibiting the highest abundance and transmission risk. The complete susceptibility of local *Anopheles* populations to Propoxur provides a valuable insecticide option for rotation strategies, though continued resistance monitoring remains essential for sustainable vector control.

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